

# **Regional Review Workshop on Completed Research Activities**

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# **Dairy Animals Research Results**

# Effect of Replacement of Cowpea hay (*Vigna Unguiculata*) for Concentrate Feed on Milk Yield and Composition of Lactating Borana Cows

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## Abstract

The study was carried out in Yabello Pastoral and Dry Land Agricultural Research Center to investigate the effect of replacement of cowpea (*Vigna Unguiculata*) for concentrate mix on milk yield and composition in lactating Borana cows fed rhodes grass hay as a basal diet. Four lactating Boran cows of similar milk yield, body weight and stage of lactation (early lactation), but different in parities were arranged in 4 x 4 Latin square design. The treatments included offering rhodess grass hay a basal diet ad libitum with different proportion of concentrate mix and cowpea hay. T1= 100% concentrate mix and 0% cowpea hay (control), T2= 80% concentrate mix and 20% cowpea hay, T3= 65% concentrate mix and 35% cowpea hay and T4= 50% concentrate mix and 50% cowpea hay. The concentrate mix consisted of 49.5% wheat bran, 49.5% noug seed (*Guizotia abyssinica*) cake and 1% common salt. There were no significant differences ( $P>0.05$ ) in dry matter and nutrient intakes among treatment groups, while milk yield was significantly different ( $P<0.05$ ) among treatments. The overall mean of daily milk yield was 4.05 kg/day. Cows that were receiving 100% concentrate mix (T1) produced the highest milk yield (4.18 kg/day) followed by cows receiving the diet in which 50% of the concentrate mix was replaced by cowpea hay (T4). Except on milk fat, there were no significant effects ( $P>0.05$ ) of treatments on milk protein, ash, lactose, solids not fat and total solid contents. The highest net return (15,744.4 Birr) was obtained from cows fed the diet in which cowpea hay replaced 50% of the concentrate mix (T4). As conclusion, cowpea hay can substitute up to 50% of the concentrate mix without adversely affecting feed intake, nutrient intake, milk yield, and milk composition of lactating Borana cows fed rhodes grass hay as basal diet. This substitution level was also found to have economic benefits. Therefore, smallholder farmers can improve milk production of their cows on poor pasture by feeding cowpea hay up to 50% to replace the concentrate mix, especially during dry seasons.

**Keywords:** Cowpea, Hay, Milk Yield, Lactating cows, Rhodes grass

## Introduction

Ethiopia has the largest livestock population in Africa estimated at about 70.29 million head of cattle, 42.91 million sheep, 52.46 million goats, 10.79 million donkeys, 2.15 million horses, 0.38 million mulls, 8.1 million camels, 56.99 million poultry and 6.99 million beehives (CSA, 2021). Despite the large number of livestock population, the productivity of animals is very low. However, with rapid population and income growth, and increasing urbanization, the demand for livestock and livestock products is growing, and these present huge opportunities for the sector mainly the dairy sector (Yilma *et al.*, 2006). In Ethiopia, domestic dairy production largely depends on traditional way of production under different systems. The production systems belong to the rural smallholder (mixed crop–livestock) production, pastoral and agro–pastoral production, urban and peri-urban small holder dairy production, or specialized

commercial dairy production system. Pastoral and agro–pastoral production system comprises the vast lowland areas of the country where arid and semiarid agro–climates dominate and subsistence is mainly based on livestock and livestock products, milk being the main source of food, except in agro–pastoral areas where some crops are produced. In all dairy production system of the country, productivity of indigenous dairy cow is very poor.

In addition to animal health problems, lack of adequate quantity and quality of feed is a major cause of poor livestock productivity. It was reported that the use of improved feeds is limited (0.3%) in rural areas of Ethiopia. Native pasture grass is the major feed resource (56.23%) followed by crop residue (30.06%). Hay and by-products are also used as animal feeds comprising about 7.44% and 1.21% of the total feeds, respectively (CSA, 2015). Earlier report by Dejene *et al.* (2014) on dairy cattle feed recourses in Borana areas during dry season, revealed that the main source of feed for animal was standing hay, which was poor in quality and limited in supply. This leads to dry season feed shortage in quantity and quality. The digestibility and intake of these feeds are low thus resulting in poor performance of the animals (Wamatu *et al.*, 2019). Despite their potential economic benefits, cereal grain and concentrate as supplements to low-quality feeds are unaffordable by smallholder farmers, in addition to their scarcity because of grain use as human food. Therefore, there is a need to look for other protein sources that farmers/pastoralist could grow on their own farm with minimum cost.

Borana pastoral area is a part of the lowlands of Ethiopia where livestock husbandry is the essential part of the production system, and livestock serve as a source of cash income and food supply for households. Feed shortage is one of the critical factors that affect milk production and productivity of dairy cattle in Borana pastoral areas. Recommended incorporation of improved forage legumes into the existing production systems is a practically appropriate and valuable economic option to purchasing a protein or energy rich concentrates for a smallholder dairy producers. Forages are effective in increasing milk yields by as much as 50% and also reduce the pressure on pasture (ILRI, 2009).

Cowpea is one of the forage legumes used for livestock feeding and is the most popular legume used for its grain in Africa, particularly in Sub-Saharan. Cowpea forage is usually superior to other forage legumes in terms of both quantity and quality (Koralagama *et al.*, 2002). Sekota dry land research center has recommended two varieties of cowpea which have potential to produce high biomass ranging from 1.8 to 2.1 ton DM/ha (SDARC, 2008). Most of the farmers grow local cultivar for seed production and biomass and used the haulms for feeding selected animals such as sick, lactating and castrated animals. Cowpea is an excellent source of protein containing 19.5 -26%. This justifies that it could be a good substitute for the more expensive concentrates (Owolabi *et al.*, 2012). A study by Tesfaye *et al.* (2016) showed that cowpea hay can replace up to 50% of concentrate mix without any significant reduction in milk yield under semi-arid conditions and valuable economic alternatives to purchased protein or energy rich concentrates as a practical solution for smallholder dairy production. As a result, this study was designed to evaluate the effect of substitution of concentrate mixture with cowpea hay as a protein source on milk yield and composition in lactating Borana cows.

## **Materials and Methods**

### ***Description of the study site***

The experiment was conducted in Borana zone at Yabello Pastoral and Dryland Agriculture Research Center (YPARC) which is located about 567 km to the south of Addis Ababa on the main road to Moyale. There are four major seasons in the Borana area: (1) - Ganna (March-May), the long rainy season; (2)- Adoolessa (June–August), the cool dry season; (3)-Hagayya (September- November), the short rainy season; and (4) Bona (December-February), the warm dry season. The average annual rainfall ranges from 350 and 900 mm. Livestock production is the main stay of the pastoral and agro-pastoral community of the Borana people. The major livestock raised are cattle, camel, goat, and sheep. Crop production is also practiced in the area (Coppock, 1994).

### ***Experimental Animals and Management***

A total of four Borana breed cows were used for the experiment. Experimental cows with the same stage of lactation, similar body weight, but with 2<sup>nd</sup> and 3<sup>rd</sup> parities were selected from the total dairy herd available in YPDARC and used for experiment. All cows were dewormed with broad-spectrum antihelminthics (Albendazole 500 mg) and sprayed with diazinol for external parasites prior to commencement of the experiment. Careful observation and follow up has been undertaken for the occurrence of any health problem and disorders during the experimental period. The experimental animals were individually managed in a well-ventilated barn with concrete floor and appropriate drainage slope and gutters. Throughout the experimental period, animals had free access to water while basal and treatment diets were offered in the milking parlor individually. The cows were hand-milked twice daily at approximately 12-hour intervals in milking room.

### ***Feed Preparation and Feeding Management***

Rhodes (*Chloris gayana*) grass hay was established at Yabello Agricultural Research Center on-station on four hectares of land. Fertilizer (DAP) at the rate of 100 kg/ha was applied during the year of establishment in 2020. Partially dried Rhodes (*Chloris gayana*) grass hay was offered ad-libitum (adjusted up to 20% refusal) in the morning hours after it was chopped to 10 cm-20 cm in order to minimize selection. On other hand, Cowpea hay was harvested when 25% of the pods were colored. Then it was chopped and stored under a hay shade and used throughout the experimental period. The amount of cowpea hay was given depending on the percentage of crude protein it is supposed to replace in the concentrate feed. In other words, the amount of cowpea hay to be given was adjusted depending on the CP in the formulated concentrate mixture in such a way that an equivalent CP was supplied by the cowpea hay. The concentrate was purchased from Adama town and the quantity of the concentrate mix offered daily was at the rate of 0.5 kg/l of milk produced by each cow with equal portions during the morning and evening milking time. Feed refusals were collected and weighed before the next feeding. Feed intake was calculated as the difference between the quantity of feed offered and feed refused.



### ***Experimental Design and Treatments***

At the beginning of the experiment, five cows were randomly assigned in 4X4 Latin square design to the control and intervention diets that consisted of three levels of cowpea hay replacement for concentrate mix. Supplementation diets were offered during milking time. There were four periods each consisting 21 days. During the first 7 days of each period, animals were acclimated to the experimental diet and the remaining 14 days were used as experimental period when data collection was undertaken. Hence, the experiments took 84 days; being started in March 2021 and finished in May 2021. *The four dietary treatments were:*

T1= Concentrate mix (100%) (0.5 kg/1kg of milk) + RGH *ad libitum* (control)

T2= Concentrate mix (80%) + Cowpea hay (20%) + RGH *ad libitum*

T3= Concentrate mix (65%) + Cow pea hay (35%) + RGH *ad libitum*

T4= Concentrate mix (50%) + Cowpea hay (50%) + RGH *ad libitum*

The concentrate mixture was composed of 49.5% wheat bran + 49.5% noug seed cake + 1% salt. The basal feed was offered *ad libitum* at a 20% refusal rate and the offer was adjusted every four days. Adjustments for concentrate offer was made at the end of each period and for each treatment based on the actual milk produced. The amounts of cowpea given were calculated depending on the amount of CP in the concentrate to be replaced thus making the diets iso-nitrogenous.

### ***Experimental measurements***

Chemical analysis of the feed samples was undertaken at Hawassa University, Animal Nutrition Laboratory. The samples were dried in an oven at 105<sup>0</sup>C overnight in a forced draft oven to determine the DM contents of the feed. Other feed samples were partially dried at 65°C and ground to pass through 1mm size screen for chemical analysis. Neutral Detergent Fiber (NDF), Acid Detergent Fiber (ADF) and Acid Detergent Lignin (ADL) were determined following the procedures of Van Soest and Roberson (1985). The ash and Nitrogen (N) content were analyzed according to the procedure outlined by AOAC (1990).

Milk yield from each cow was measured using a graduated measuring cylinder. Lactating cows of each group were manually milked twice a day at 8:00AM and 6:00PM. Milk consumed by the calves was calculated as the difference in the weight calves before and after suckling.

For analyzing milk chemical compositions, representative milk samples of 100 ml were taken from each cow in the morning and afternoon twice every week during the experimental period. Then keeping in refrigerator for a while, milk fat, protein, total solid, ash and lactose percentage were determined by using a Lacto Scan milk analyzer (Milkotronic Ltd, Nova Zagora, Bulgaria). In addition, the following simple arithmetic was used to calculate some chemical entities of milk.

$$\text{SNF} = (\text{TS} - \text{fat}) \times 100 \text{ (O'Mahoney, 1988)}$$

$$\text{Percent ash} = \frac{\text{Weight of residue}}{\text{Weight of sample}} \times 100 \text{ (Richardson, 1985)}$$

***Percent lactose =Percent total solids-(% fat+ % protein+ % total ash) (O'Mahoney, 1988)***

### ***Partial budget analysis***

The partial budget analysis and marginal rate of return were calculated to determine the profitability of the different supplemental feeds. The partial budget analysis involved the calculation of variable cost and benefits. According to (Ehui *et al.*, 1992) Net income (NI) was calculated as the amount of money left when total variable cost (TVC) was subtracted from total returns (TR). In this experiment the variable costs included estimated purchase price of the supplemental feed, and labor cost for preparation of the supplemental feeds. While total return (TR) was estimated as the average sale price of milk produced per animal per day during the experimental period Net income (NI) = TR TVC).

### ***Statistical Analysis***

Data of milk yield and compositions were subjected to GLM procedure or Latin Square Design using Statistical Analysis System (SAS, 2008). Treatment means were separated using Least Significant Difference (LSD) at  $\alpha = 0.05$ . The model used for analysis of the data was:

$$Y_{ijk} = \mu + C_i + P_j + T_k + E_{ijk},$$

Where;  $\mu$  = Overall mean;  $C_i$  = Cow effect (parity);  $P_j$  = Period effect;  $T_k$  = Treatment effect;  $E_{ijk}$  = Experimental error

## **Result and Discussion**

### ***Chemical Composition of the Experimental Feeds***

Chemical composition of the experimental feed is presented in Table 1. The DM content was almost similar for feeds used in the experiment. DM content of the formulated concentrate mix in the current study agrees with the mean DM content of 91.09% reported by Geleta and Demissu (2020). On the other hand, the DM content of cowpea hay was found to be similar with the finding of Tesfaye *et al* (2016) in which the DM content of 90.64% was observed. The highest CP content was noted in Nougseed cake, followed by concentrate mixture and Cowpea hay in that order. The CP content of cowpea hay in the current experiment is within the range of 18.78 to 20.22% observed by Hasan *et al.* (2010) for cowpea forage fertilized with different levels of fertilizer. However, in comparison to the CP contents of 21.03% reported by Tesfaye *et al* (2016), the cowpea hay in the current study exhibited lower CP contents. This might have resulted from the stage of harvest and loss of nutrients during the hay making process in the present trial. The supplements of concentrate mix and Cowpea hay have lower NDF and concentrations relative to the Rhodes grass hay used. The low levels of NDF in both supplements are indicative of high cell soluble matter. The same trend was observed for ADF contents with the highest value recorded for rhodes grass hay followed by cowpea hay and the noug seed cake.

The neutral detergent fiber (NDF) of grass hay was higher than the value of 70.22% reported by Diribe *et al.* (2016) and lower than the value of 80.8 % reported by Geleta and Demissu (2020). This difference in neutral detergent fiber DF content might be attributed to the difference in stage of maturity at the harvest, type of soil, seasonal variation and the difference in management during conservation of Rhodes grass for hay.

Table 1. Chemical composition of the experimental feed

Feed Type	DM	Ash	CP	NDF	ADF	ADL	Hemi.	Cell.
RGH	91.46	11.22	8.14	72.59	47.36	5.92	24.45	41.44
Cowpea hay	90.34	12.10	19.03	48.18	31.42	5.78	16.76	25.64
Wheat bran	90.75	6.87	17.21	44.04	12.76	4.1	31.28	8.66
NSC	92.97	9.55	31.03	40.20	29.65	7.95	10.55	21.7
Conc.mix	91.67	6.83	20.79	28.99	13.01	5.38	15.98	7.63

*ADF = Acid detergent fiber; ADL= Acid detergent lignin; Cell. = Cellulose Conc. mix = Concentrate mix CP = Crude protein; DM = Dry matter, Hemi. = Hemicellulose; NDF= Neutral detergent fiber, NSC = Noug seed cake, OM = Organic matter, RGH=Rhodes Grass Hay*

### **Dry matter and nutrients intake**

The overall mean feed dry matter and nutrient intakes of cows in the feeding treatments are presented in Table 2. There were no significant differences ( $P>0.05$ ) in dry matter and nutrients intake among the treatment groups. The present study is disagreement with the findings of Tesfaye *et al.* (2016) in which the replacement of concentrate with cowpea hay at the rate of 50% increased the total DM intake of cows significantly ( $P<0.05$ ) over those cows received cowpea hay at other lower rates of replacement.

Crude protein and MEI intakes have followed the same trend as dry matter intake which is not significant ( $P>0.05$ ) among treatment group. The highest MEI (63.29 MJ/head/day) obtained from 100% Concentrate mix (T1) is far from the estimated daily ME requirement (97.6 MJ/head/day) of lactating cows weighing 400 kg and producing 8-10 kg milk of 4.5% butter fat (ARC, 1990). But the mean ME intake obtained in this study is sufficient to meet the daily requirement for ME of cows with a mean daily milk yield of 4.05 kg/cow/day. Neutral detergent fiber and Acid detergent fiber intakes were also no significantly different ( $P>0.05$ ) among treatment groups.

Table 2 Dry matter and nutrients intake

Intake (Kg/day)	Treatments						
	T1	T2	T3	T4	Mean	SL	SEM
Total DMI	7.59	7.48	7.46	7.55	7.52	ns	±0.43
DMI (% LW)	2.76	2.72	2.71	2.75	2.74	ns	±0.11
Total CP	0.95	0.90	0.88	0.92	0.91	ns	±0.08
Total ME(MJ/day)	63.29	62.1	62.05	62.85	62.57	ns	±2.34
Total NDF	4.56	4.18	4.16	4.51	4.26	ns	±0.02
Total ADF	3.76	3.21	3.26	3.39	3.41	ns	±0.01

*Treatments within row are not significantly different ( $P > 0.05$ ); ADF = Acid detergent fiber; CP = Crude protein; DM= Dry matter; ME = Metabolisable energy NDF = Neutral detergent fiber; NS = Not significant SEM = Standard error mean .*

### ***Milk yield***

Mean milk yields of the cows in each treatment are presented in Table 3. Milk yield was significantly different ( $P < 0.05$ ) among treatments. Cows those received the 100% concentrate mix (control) (T1) produced the highest milk yield followed by those received the 50% level of cowpea hay replacement for concentrate mix (T4). The lowest milk yield was recorded for cows fed the 20% and 35% level cowpea hay replacement for concentrate mix. Tesfaye *et al.* (2016) also found no significant difference ( $P > 0.05$ ) in daily milk yield between cows fed 100% concentrate mix and those fed cowpea hay up to 50% level of replacement for concentrate mix.

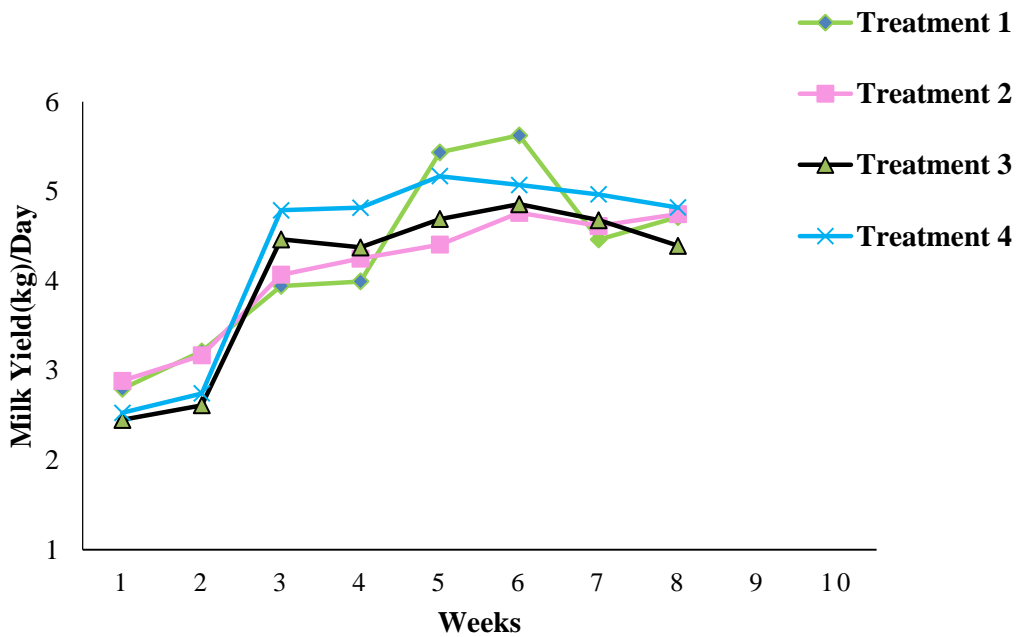
Table 3. Effect of replacement of concentrate mix with different proportions of Cowpea hay on milk yield in lactating Borana cows fed rhodess grass hay.

Treatment	Milk Yield
T1	4.18 <sup>a</sup>
T2	3.96 <sup>b</sup>
T3	3.95 <sup>b</sup>
T4	4.10 <sup>a</sup>
Mean	4.05
Significance level	**
SEM	0.05

<sup>a-b</sup> means within column having different superscript are significantly different at; (\*\*) =  $P < 0.05$ ; SEM = standard error of mean

The lactation curve in Figure 1 represents the milk yield for a lactation period of 56 days. Cows on all dietary treatments reached peak milk yield between 5<sup>th</sup> and 6<sup>th</sup> weeks, but retained that peak lactation for short duration (Figure 1).

Figure 1. Lactation curve of lactating cows fed on ad libitum Rhodes grass hay supplemented with different proportions of cowpea hay as partial replacement to concentrate mix.



### ***Milk composition***

The mean values for CP, fat, total solids, solid-no-fat, ash and lactose contents of milk from cows in different treatments are presented in Table 4. Except for milk fat, treatment effects were non-significant ( $P>0.05$ ) for milk protein, ash, lactose, solids not fat and total solid components. The highest (4.37%) and lowest (3.77%) values of fat content were observed for T3 and T1 respectively. Khalili and Sairanen (2006) observed higher milk fat content for cows allocated only green hay. This is due to fact that, rations with more concentrates may result in changes in proportion of ruminal VFA, which in turn can result in the reduction of milk fat like in T1 of the current study. But difference between T2, T3 and T4 in fat percent might be due to the difference in the forage characteristics such as forage particle size, maturity, and fiber content of the forage. However, the higher level of CP intake by the animals did not significantly affect the concentration of milk protein in the current trial. This study agrees with the conclusion made by Getu *et al.* (2010). The mean value of lactose observed in the present study is (4.64%) is slightly higher the findings by these authors. The total solid percentage (12.80%) observed in the present study contrasts with the value of 15.05% reported earlier by Tesfaye *et al.* (2016). This difference could be associated with the difference in breeds of cows used in the two experiments.

Table 4. Effect of replacement of concentrate mix with different proportions of cowpea hay on milk composition in lactating Borana cows fed rhodess grass hay

Treatment	Milk Composition (%)					
	Fat	protein	Ash	Lactose	SNF	Total Solid
T1	3.77 <sup>b</sup>	3.18	0.77	4.81	8.76	12.75
T2	3.99 <sup>b</sup>	3.13	0.74	4.72	8.59	12.36
T3	4.37 <sup>a</sup>	3.37	0.64	4.88	8.89	13.26
T4	4.17 <sup>a</sup>	3.17	0.74	4.77	8.68	12.85
Mean	4.08	3.21	0.72	4.64	8.73	12.80
SEM	0.3	0.06	0.3	0.06	0.12	0.42
SL	*	ns	ns	ns	ns	ns

<sup>a-b</sup> means within column having different superscript are significantly different at (\*) =  $P<0.05$ ; SEM = standard error of mean

### ***Partial budget analysis***

The partial budget analysis for feeding lactating Borana cows with basal diet of rhodes grass hay supplemented with different proportions of cowpea hay as replacement for concentrate mix is summarized in Table 5. In this study, though the difference among treatments was not significant, highest net return (15,744.4 ETB) was obtained from cows fed with 50% cowpea hay replaced concentrate mix (T4) than cows in other treatment groups. This might be due to the fact that the higher milk yield obtained from cows in this treatment combined with the lower cost incurred during cowpea hay production than buying concentrate mix.

Table 5. Partial budget analysis of lactating Borana cows fed basal diet of rhodes grass hay supplemented with different proportions of cowpea hay as replacement for concentrate mix.

Variables( birr)	Treatments			
	T1	T2	T3	T4
Days of Experiment (days)	70	70	70	70
Feed Cost/Cow/Day	38.66	30.09	25.18	21.08
Milk Yield/Cow/Day(liter)	4.18	3.96	3.95	4.1
Feed Cost Per kg of milk/day	9.25	7.6	6.37	5.14
Gross Income (GI) /day	250.8	237.6	237	246
Total Cost/cow/Experimental Period	2706.2	2106.3	1762.6	1475.6
Total Revenue	17,556	16632	16590	17220
Net Return	14,849.80	14,525.70	14,827.40	15,744.40
Benefit Cost Ratio - BCR =TR/TC	6.49	7.9	9.41	11.67
Rate of Return - RR=NR/TC	5.49	6.9	8.41	10.67
Gross Ratio - GR= RR/BCR	0.85	0.87	0.89	0.91

### Conclusion and Recommendation

The result indicated significantly higher daily milk yields for T1 and T4 compared to T2 and T3. Except Fat, all milk composition was not significance difference ( $P>0.05$ ) among all treatment groups. The partial budget analysis also indicated that cows receiving 50% level cowpea hay replacement for concentrate mix (T4) provided highest net return and thus being economically beneficial than the cows those received the other two levels of cowpea hay replacement and the control group (T1).

In general, cowpea hay up to 50% as substitute for concentrate mix without adversely affecting feed intake, nutrient intake, milk yield and composition while highest net return was gained on cows fed on T4 (50% cowpea level). Therefore, considering these potentials, smallholder dairy farmers can use cowpea hay up to 50% to replace the concentrate mix for cows on poor pasture, especially during dry season to improve their cows' milk production.

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# **The effect of substitution of Lablab for concentrate on milk yield and milk composition of lactating F1 Arsi Friesian dairy cows**

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## ***Abstract***

*The objective of the present study was to evaluate the effect of partial substitution of Lablab purpureus for concentrate in the diet of growing F1 (Arsi x Holstein Friesian) lactating cows and to evaluate the economic advantage of substitution of lablab purpureus for concentrate. Four lactating cross bred cows with first parity were allocated to four treatments of Lablab purpureus substitution at the rate of 0, 25, 50 and 75% using switch over arrangement of four by four latin squire design. The experiment lasted for four months with 15 days of adaptation period and 15 days of feeding and data collection during each period of the switch over. The result indicated no statistically significant difference ( $P < 0.05$ ) in feed intake among the four treatments. In addition there was no statically significant difference among treatments with 0, 25% and 50% lablab substitution both in milk yield and milk composition. The net income recorded were 114.56, 112.63, 111 and 82 Birr per cow per day for 50%, 25%, 0% and 75% lablab purpureus substitution for concentrate, respectively. It can be concluded that substitution of lablab purpureus for concentrate in the diet of lactating F1 cows up to 50% will be economical without affecting the biological responses.*

**Key words:** feed intake, milk yield, supplement, milk composition

## **Introduction**

In most tropical regions, the majority of animal feed is derived from poor quality crop residues and agro-industrial by-products. Most of the resource poor farmers, which constitute the bulk of the livestock farmers in the tropical regions, are unable to afford good quality feeds due to their high cost, leading to sub-optimal productivity of their animals (FAO, 2012).

Conventional feeds are feed stuffs which are commonly utilized by farmers or used for preparing formulated feeds by commercial feed manufacturers. Availability of conventional feed resources is declining due to shrinking grazing lands as a result of expansion of cropping and urbanization. High cost and non-availability of concentrates and protein source feeds necessitates the use of locally available supplements for dairy cows. These should be supplied in higher quantities as a replacement to concentrates to reduce the feed costs (Goswami et al., 2013, seyoum et.al.2018). It is suggested that the use of improved forage legumes integrated into existing farming systems are valuable economic alternatives to purchased protein or energy rich concentrates as a practical on-farm solution for smallholder dairy production (Denbela 2017).

At present, the larger proportion of livestock products is produced by smallholder crop-livestock mixed farmers operating under such poor quality feeds in mixed farming systems. .As much of the arable land is already under cultivation, increased livestock productivity will thus have to come through enhancing qualities of these fibrous feed resources. One way to achieve this is through integration of low cost feed technologies that are easy and suitable, and within the limits of the resource poor farmers.

Supplementation of crop residues with plant protein sources such as leguminous forage crops may alleviate protein deficiency as these contain medium to high levels (12–25%) of CP (Solomon, 2004). Supplementing ruminant livestock with feeds that are affluent in protein and energy improves intake, digestibility and growth performance of ruminants fed on low quality forages. Lablab is among the legume forages used as protein supplements in ruminant nutrition to improve their production sustainability of the ruminant livestock (Solomon, 2004).

Feeding dairy cows with locally produced forages but with adequate nutrients fulfilling the animal requirement is an indispensable task to have a profitable and sustainable dairy farm. Farmers in rural areas with adequate arable land can produce these animal feed resources as one means of supplementing their animals by practicing forage development and/or intercropping them with other crops. The use of locally produced forages as a means of supplement has tremendous advantages with respect to cost of the supplemental feed without affecting the biological requirement of the animals. Unfortunately, very few works were done with respect to the use of lablab for milking animals and hence the current study was initiated to address this issue.

### ***Objectives***

- To evaluate the supplementary value of graded level of lablab in the diet of lactating dairy cows
- To assess the economic advantage of using the graded level of forage legume as a supplementary feed for dairy cows

### **Materials and methods**

#### ***Description of the study area***

The experiment was conducted at Adami Tulu Agricultural Research Center (ATARC), which is located 167 km south of Addis Ababa at an altitude of 1650 m above sea level in mid rift valley. The agro-ecological zone of the area is semi arid and sub humid with acacia woodland vegetation type. The mean annual rain fall is 760 mm. The mean minimum and maximum temperatures are 12.6 and 27<sup>0</sup>c, respectively. The soil type is fine, sandy loam with sand: silt: clay ratio of 34: 38: 18, respectively. (ATARC, 1998).

#### ***Experimental treatment set up and animal management***

Four cows were randomly blocked in a switch over 4 x 4 Latin square design composed of 15 days of adaptation and 15 days of treatment period. The experimental animals were then randomly allotted to one of the four dietary treatments given below.

T1= Chopped Maize stover *ad libitum* + concentrate (Noug cake + wheat bran + salt ) mix (control)

T2= Chopped Maize stover *ad libitum* + 25% of the concentrate mix replaced by lablab hay

T3= Chopped Maize stover *ad libitum* + 50% of the concentrate mix replaced by lablab hay, and

T4= Chopped Maize stover *ad libitum* + 75% of the concentrate mix replaced by lablab hay.

All the experimental animals were treated for internal and external parasites before the commencement of the experiment. Feed formulation was done based on the requirement of the animals (weight and milk yield) and the quantity of supplemental diet was at the rate 0.5 kg/lit of milk produced each day. And the concentrate was delivered in two equal portions in the morning and the evening.

### ***Feed sample analysis***

Chemical composition of the supplemental diets was analyzed. Representative samples of 100 g of the ingredients were collected once in a week when the feed is mixed. For the individual feed components, sample from the upper, middle and bottom parts of their container was taken to make the sample representative. The feed samples, 250 gm from each ingredient and treatments, were partially dried at 65°C for 48 hrs and ground to pass through 1mm sieve in Holeta Agricultural Research Center laboratory. Chemical analysis was also conducted at this laboratory. Crude fiber, Dry matter, Nutrient free extract, Ether extract and Ash were determined using proximate procedures (Van Soest and Robertson 1985). Nitrogen was determined according to Kjeldhal procedure and crude protein was calculated as N x 6.25. In vitro dry matter digestibility was determined by the two stage method developed by Tilly and Terry (1963). Rumen fluid was collected from three rumen fistulated steers before morning feeding. The steers were fed on natural pasture hay ad libitum and two kg concentrate per head per day.

### ***Gross margin Analysis***

The partial budget analysis and marginal rate of return was calculated to determine the profitability of the four different supplemental feeds used in this study. According to (Ehui et al. 1992) Net income (NI) is calculated as the amount of money left when total variable cost (TVC) is subtracted from total returns (TR). In this experiment the variable costs include estimated purchase price of the basal and supplemental feed, labour cost for preparation of the supplemental feed and cost for medicaments and treatments. Total return (TR) was estimated from the average selling price of milk produced per animal per day during the experimental period.  $NI = TR - TVC$ .

**Statistical analysis:** Switch over Latin square design was applied in the experiment

$$\text{Model: } X_{ijk} = \mu + \alpha_i + B_j + Y_k + E_{ijk}$$

where,  $X_{ijk}$  = response variable,  $\mu$  = overall mean,  $\alpha_i$  = treatment effect,  $B_j$  = block effect,  $y_k$  = column effect and  $E_{ijk}$  = error term.

Analysis of variance (ANOVA) following the General Linear Model (GLM) procedure of SAS (SAS, 2002, version 9.1.3) was used to analyze the data. Differences among treatment means were separated using LSD at 5% level of significance.

## Result and discussion

### *Composition of experimental Feeds*

The dry matter (DM), Ash, organic matter (OM), Neutral Detergent Fibre (NDF), Acid Detergent Fibre (ADF), Lignin, Crude Protein (CP) of all the feed ingredients and the treatment diets are presented in Table 1.

Table 1. chemical composition of experimental feed

Feed sample	DM (%)	Nutrient composition					
		Ash	CP	NDF	ADF	Legnin	OM
Feed ingredients							
Lablab	91.50	9.50	20.01	41.02	37.58	7.90	90.50
Noug cake	91.98	12.91	31.54	39.64	31.56	9.16	87.49
Wheat bran	90.56	4.58	15.98	46.08	14.74	3.93	95.45
Treatments							
Treatment 1	90.67	7.05	22.08	68.54	52.19	6.74	92.85
Treatment 2	90.75	7.08	22.08	67.89	52.86	8.38	92.42
Treatment 3	90.94	7.10	22.07	67.56	52.59	8.05	92.65
Treatment 4	90.08	7.14	22.06	67.49	53.74	9.79	92.29

DM= dry matter, OM= organic matter, NDF = neutral detergent fibre, ADF= Acid detergent fibre, CP= crude protein

The crude protein (CP) content of the lablab was slightly higher than the value of 19.93% reported by Abuye et.al (2018) and slightly lower than the value of 20.2% report by workenesh (2014) and the value of 21% report by Chala Duguma et.al (2019). The dry matter (DM) and organic matter (OM) content of the lablab was comparable with the result obtained by Abuye et.al (2018) and worknesh (2014).

### *Milk yield and feed intake*

Milk yield and feed intake of the experimental animals during the experimental period is presented in Table 2.

Table 2. Daily feed intake and milk yield ( $\pm$  S.E) of the experimental animals

	Treatments	Fee intake (kg)	Milk yield (kg)
5	Treatment 1	5.11 $\pm$ 0.35	6.53 $\pm$ 0.32 a
6	Treatment 2	5.06 $\pm$ 0.37	6.3 $\pm$ 0.3 a
7	Treatment 3	4.94 $\pm$ 0.34	6.07 $\pm$ 0.39 a
8	Treatment 4	4.05 $\pm$ 0.27	4.6 $\pm$ .21b

a, b= means with in the column with different letter show significant difference at  $p < 0.01$

The result indicated that the mean milk yield among different levels of lablab supplementation was significantly different ( $P < 0.01$ ). On the other hand, even if the difference is not significant there is a bit difference in the mean DM intake among the treatments. The mean milk yield is less than the milk yield reported by (Nyambati, 2003) which was 9.5 $\pm$  0.05 kg for Holstein cows fed on Napier grass and lablab. However, the result in the current study is in agreement with the finding of Getu et.al. (2010) in which 6.68 to 6.45 kg milk yield was observed in substitution of legume hay for concentrate at the rate of 0 to

50%.with But the current milk yield for 75% substitution of lablab (4.6±0.21kg) is lower than the figure reported by the same author for 75% of substitution of concentrate with legume. On the other hand, the present result is higher than the milk yield reported by Tesfaye et.al. (2016) which was 3.08, 3.31 and 2.75 kg milk/cow/day for substitution rate of 25, 50 and 75% legume hay for concentrate, respectively for lactating Horro cows.

### ***Milk composition***

Treatment effects on milk composition of the experimental animals is given in Table 3. Of all the milk composition, fat and total solid were found to be significantly different ( $p < 0.05$ ) among the treatments. As the lablab substitution level in the supplementary feed increased, both the fat and the total solids increased. The increase in fat content might be due to the increase in the fiber component of the experimental feed, which in turn had the same effect on the total solid.

Table 3. Effects of treatments on milk composition (%) for the experimental animals

Treatment	Fat	Protein	Lactose	Ash	Total solid	P - value
T1	3.84±0.35b	2.65±0.05	3.98±0.005	0.71±0.01	11.3±0.075b	0.025
T2	3.83±0.14b	2.74±0.015	4.12±0.02	0.75±0.01	11.44±0.05b	
T3	4.52±0.01a	2.67±0.02	4±0.03	0.77±0.01	11.87±0.02b	
T4	5.24±0.45a	2.85±0.02	4.08±0.03	0.75±0.005	12.92±0.038a	

<sup>a,b</sup> Parameters with in the column with different letters are significantly different  $p < 0.05$ .

### ***Gross margin analysis***

Results of the partial budget analysis are shown in Table 4. Substitution of concentrate with the lablab at the rate of 50% resulted in the highest return followed by the 25% substitution, whereas supplementation with 75% substitution of concentrate with lablab gave the lowest rate of return.

Table 4. Gross margin analysis

Particulars	Treatment 1	Treatment 2	Treatment 3	Treatment 4
Supplemental feed cost/trt/day	47.4	38.875	27.95	18.435
Labour cost/trt/day	33.325	33.325	33.325	33.325
Medicaments/trt/day	4.1667	4.1667	4.1667	4.1667
Total input cost/trt/day	84.89167	76.3667	65.4417	55.9267
Total variable cost/trt/day	84.89167	76.3667	65.4417	55.9267
Total Return/trt/day (sale of milk)	195.90	189	180	
Net income/trt/day	111.0083	112.63	114.5583	82.0733

\*The calculation was done on the bases that 1lt is milk is sold for 30 birr

### **Conclusion and recommendation**

The present experiment was initiated with the objective of identifying the best level of substitution of lablab for concentrate in the diet of lactating cross bred Arsi X Holstein Friesian cows with out affecting milk yield of the cows. Accordingly the substitution concentrate with 25% and 50% lablab did not show

significant difference in milk yield as compared to cows to fed only with concentrate mix. But the 75% substitution level showed significantly lower milk yield as compared to cows supplemented with concentrate mix. The milk composition result indicated that fat content of the milk increased significantly ( $p < 0.05$ ) with the increase in lablab supplementation level starting from 50% substitution level. The net incomes per cow per day were decreasing for 50, 25%, 0% and 75% substitution in that order. Generally the 50% lablab substitution level is recommended as it resulted in the best biological performance of the cows as well as the higher level economic return.

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# Assessment of Boran Cattle Calf Management and Associated Risk Factors in Selected Districts of Borana Zone

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## Abstracts

*Study was conducted with the objectives to assess calves management practices, constraints and associated risk factors in Borana zone. A total of 180 respondents were selected. The study revealed that 81.11% of respondents had awareness on importance of feeding colostrum to newborn calves though 18.89% of them had no knowledge about importance of it for calves. Out of total, 67.79% of respondents allowed calves to start other feeds after 2 weeks of birth. The study indicated that the major sources of calf feed were natural pastures and crop residues. Respondents explained that they also supplemented their calves with seasonal crop residues. Ponds bore hole and pump were sources of water for calves. . The average weaning age of calves was revealed to be  $6.7 \pm 1.80$  months. The major weaning criteria for calves were age of calves and body condition their dam (61.70) as per this study. The major problems of calve raring were feed and water followed by calf diseases. Amongst the diseases were FMD, heart water, lumpy skin disease, and CBBP were the major ones according to respondents. and , pasteurellosis, diarrhea and liver fluke. On the other hand, liver fluke was ranked 1<sup>st</sup> disease cause calf mortality followed by heart water, contagious bovine pleuropneumonia, diarrhea, pasteurellosis, foot mouth disease and blackleg, respectively. In general, the feed shortage and health problems in pre weaned calves were the major findings in this study which might affect the herd replacement of the dairy cattle.*

**Keywords:** Calf, Risk factors, Borana zone, Management, Assessment

## Introduction

Livestock provides an enormous service in the Ethiopian household economy by providing food, input for crop production and soil fertility management, cash income as well as in promoting savings, fuel, social functions, and employments (Land O'Lakes Inc., 2010). With this wide array of functions, livestock can be considered as a vehicle for improving food security and better livelihood of the rural population (Land O'Lakes Inc., 2010). Like in other pastoral and agro-pastoral production system, livestock production forms the basis of the economy, especially the main source of food and income supporter of Borana zone. Borana pastoralists have various indigenous practices such as herd mobility, herd splitting, feeding and watering, and breeding, which have direct and/or indirect influence on the productivity their animals regardless of the variation in the magnitude and intensity. Such practices are depend on climatic factors, availability of pasture and water, and culture (Ayana and Fekadu, 2003). Boran cattle and other cattle type of neighboring zone are the predominant species and strain of cattle, which is reared for various purposes (Zelalem, 2012).

The huge and diverse cattle population, varied and favorable agro-ecology for dairying, increasing demand for dairy products in urban and peri-urban areas, long- standing culture of dairy products consumption, and favorable policies are indicators of the importance and potential of dairying in the

Ethiopia (Tegegne *et al.*, 2013). Cows contribute to about 95% of the total annual milk produced and the remaining 5% comes from camels from the pastoralist areas of the country (CSA, 2010). A successful dairy farm operation requires that a large percentage of cows wean a live healthy calf every year. According to Godden (2008), rearing healthy dairy calves to weaning time requires maximizing the calf's level of immunity against disease while minimizing its exposure to infectious agents. However, among the factors that hinder success of dairy industry, morbidity and mortality of calves is the one (Acha *et al.*, 2004). High mortality and/or morbidity rates of newborn calves, caused mainly by failures in the transfer of passive immunity, are the main causes for such inefficiency in the dairy activity. Calf morbidity and mortality are problems of major concern in all countries where cattle is raised under extensive husbandry practices (Radostits, O.M, 2001). In addition, previous report concluded that calf morbidity and mortality were found to be relatively high in the examined area and can have short-term and long-term detrimental effects on dairy production by suppressing growth rate of the calves and replacement capacity of the herd (Asefa and Ashenefi, 2016).

In spite of advancement in dairy husbandry practices, clinical medicine and diagnostic techniques, the morbidity and mortality rates of dairy calves are still unacceptably high even on many advanced dairy farms in developed countries (Mee, 2008). Close look at management practices can help to identify risk factors that are responsible for dairy calf morbidity and mortality in order to design and implement possible solution strategies. It was reported that calf management problems such as poor housing, keeping calves with adults, absence of calving pen were amongst the prevalent factors, and hence was depicted that one third of the calves affected by at least one disease condition (Tekalign, 2020).

Raising calves on the low land areas of Borana is a very important, because pastoralists' life in the low land area almost depends on livestock production and productivities. Having a successful calf raising techniques is not only important financially, but also important for the sustainable conservation of gene of cattle breeds. Understanding Boran cattle calf management practices, identifying problems and designing strategic solution is the first step in village based dairy cattle breeding program. Deciding that right management practices for the cow and calf immediately after birth, proper nutrition for the young calf and applying best way to keep the calf healthy is difficult prior without research based information. Identifying management practices and controlling factors those expose to diseases that affect dairy calves can provide economic, health and welfare benefits in the dairy production system. Therefore, this study was designed to assess information gabs on calf management practices and associated risk factors in Borana management practices.

## **Materials and methods**

### ***Description of the study areas***

The study was carried out in Yabello, Dire and Gomole districts of Borana zone, south part of Ethiopia. These districts were selected based on the availability of livestock and its population. The Borana plateau is the portion of the Southern Ethiopia rangelands which is generally semi-arid with annual rainfall range of 500 mm in the South and 700 mm in the North. The altitude of the area ranges from 1000m in the South to 1500m in the Northwest while the rainfall is bimodal but erratic in distribution. Fifty –nine percent (59%) of annual precipitation occurs from March to May and 27% from September to November

with annual mean daily temperature varying from 19 to 24°C. There are four major seasons in the Borana plateau: (1) - Ganna (March-May), the long rainy season; (2)- Adoolessa (June–August), the cool dry season; (3)-Hagayya (September- November), the short rainy season; and (4) Bona (December-February), the warm dry season (Coppock, 1994)

### ***Data collection methods***

A survey was conducted in three selected districts of Borana zone. From these districts 180 respondents were interviewed using open and closed structured questionnaires to determine calf management practices and its associated risk factors in the study areas. Data on management practices such as feeding, housing, watering, colostrum feeding, disease (both infectious and non-infectious) and other managements were collected for new born to pre-weaned calves. To validate the information obtained from individual interview, focus group discussion was undertaken with elder pastoralists who have better knowledge on livestock production of the study area.

### ***Site selection and Sampling procedure***

Districts of the zone were purposively selected by considering availability of livestock and their agro-ecologies. Selection of the study area was carried out based on the secondary data that were gathered from zonal development livestock and fishery resources development office. Accordingly, two (Pastoral Association) PAs were selected randomly from each district. Sampling was undertaken by simple random selection of households. Depending on the availability of livestock, 180 respondents were selected from the three districts. The components of the study subjects were based on preliminary survey, group discussion and interview of owner using semi-structured questionnaires.

### ***Experimental animals and clinical examination***

During the study period, 92 calves were born and registered. Before starting clinical observation, all selected calves were given identification name based on their owner, date of birth and sex followed by registration of the identification name on data collection record sheet. The calves were regularly monitored and clinically examined for a period of 6 months.

Examination of vital signs and other overt clinical signs like coughing, diarrhoea, skin condition, joints, and feet examination, deprecation, poor suckle reflex, weakness, and recumbence found during the examination were recorded on a predesigned format. Moreover, the calves' health was evaluated via objective criteria of appetite, fecal consistency, hydration status, and behavior. A total of 92 fecal samples were collected from households of selected calves in the study area. In each selected PA animal health expert were assigned to the selected animals for follows up of the animals and record any visible sign. The researcher supervised the animals and data collection process by the animal health expert four times during the study period. Additionally, animal owners both female and males participated in the reporting of any illness and mortality of calves. Fecal samples were collected directly from the rectum into a universal container and later on transported to Yabello Pastoral and Dryland Research Center Laboratory for laboratory analysis. Fecal samples were examined for identification of internal parasite's eggs using sedimentation, and flotation techniques following standard laboratory procedures.

### *Data management and statistical analysis*

Microsoft excel sheet and Statistically Package for Social Science (SPSS, Version 20) were employed for data management and statistical analysis, respectively. Ranking of morbidity and mortality diseases indicated by respondents was done. Ranking was done after calculating the weighted average score for each disease and then dividing the sum by the total number of respondents. Chi-square test was used to determine differences in morbidity as well as mortality between the different reported diseases.

## **Results and Discussion**

### *General information of respondents and calves management practices*

General information on respondents is shown in Table 1. Most of the respondents in the study areas were males (67.78%) and the remaining (32.22%) were females. The age distributions of respondents were as follows: 25-45 (32.22%), 46-65 (44.44%) and 46-65 (23.34%) years old. Majority of the respondents were illiterate 154 (85.56%); whereas 11.11% and 3.33% had educational level of 1-8 and 9-12, respectively (Table 1).

Table 1. General information on respondents

<b>Parameter</b>	<b>Frequency</b>	<b>Percentage</b>
<b>Sex of respondents</b>		
Male	122	67.78
Female	58	32.22
<b>Age of respondents</b>		
25-45	58	32.22
46-65	80	44.44
>66	42	23.34
<b>Respondents education</b>		
Illiterate	154	85.56
1-8 grade	20	11.11
9-12 grade	6	03.33

Regarding calf management, the study showed that majority of calves' management was undertaken by women(51.7%) followed by all family members (18.9%), adult female and boy (12.8%), adult boy (8.9%) and husband (7.8%) in decreasing order (Figure 1)..

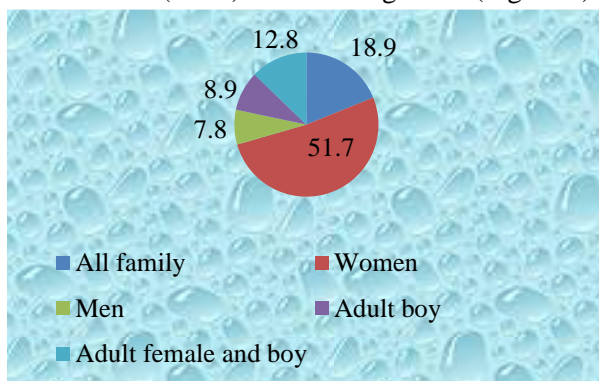


Figure 1: Family member participation in calf management activities

### *Calf housing and colostrum feeding*

The current study showed that the types of calf house in the study area individual pens housed outside (22.8%) and individual pens housed outside inside (2.8%). Majority (66.7%) of respondents used outside group housing. With regard to calves, 5.6% of the respondents kept their calves in group housing inside. Moreover, 2.2% of calf owners used locally constructed barns (Table 2).

Nevertheless, respondents indicated that they have fairly high degree (81.11%) of awareness on importance of feeding colostrum to newborn calves while 18.89% of respondents had no awareness on importance of colostrums feeding. In terms of time of colostrum feeding, 60% respondents of respondents fed their new born calf within 1 hours, whereas 28.89% fed between 2 to 4 hours, and 11.11% of them fed their calves in after 5 hours of birth (Table 2). According to the respondents, most (76.70%) of them allowed colostrum feeding until the calf is satisfied. Out of the total respondents, 2.78 % and 16.67% allowed calves feed colostrum until cow refuses suckling and until the calves stop sucking by itself, , respectively (Table 2). From the analysis output, 67.79% of respondents allowed calves to start other feeds after 2 weeks of birth, whereas 26.67% and 5.56% of respondents provide feeds after 3 and 4 weeks of birth, respectively (Table 2).

Table 2. Calf management practice

<b>Variables</b>	<b>Number of respondents</b>	<b>Percentage</b>
<b>Calf housing</b>		
Individual pens housed outside	41	22.8
Individual pens housed inside	5	2.8
Group housing outside	120	66.7
Group housing inside	10	5.6
Locally constructed barns	4	2.2
<b>Awareness on importance of colostrum</b>		
Yes	146	81.11
No	34	18.89
<b>First time of colostrum feeding after birth</b>		
In 1 hours	108	60.00
2- 4 hours	52	28.89
> 5 hours	20	11.11
<b>Administration of Colostrum feeding</b>		
Until the calves are full	138	76.67
Still the teat is empty	5	02.78
Until the calves stand by	30	16.67
No time limitation	7	03.89
<b>Time to start other feed</b>		
Two weeks	122	67.79
Three weeks	48	26.67
Four weeks	10	5.56

### ***Calf feeds and water management***

According to the result of this survey majority calf feed sources were natural pastures like grasses, legumes, herbs, shrubs and trees foliage as well as crop residues such as straws of teff, wheat, maize and sorghum, haulms of haricot beans. In addition, industrial by product like wheat bran was supplemented during strict feed shortage. As per result from this study, 7.80% and 15.60% of respondents used concentrates and both roughages and concentrates, respectively (Table 3). It was reported earlier that feed recourses of dairy cattle in Borana area particularly basal diets like common natural pastures (grasses, legumes, herbs, shrubs and trees foliage) and recently crop residues (straws of teff, wheat, maize and sorghum, haulms of haricot beans) were available in lowland (Dejene *et al.*, 2014).

A successful dairy production requires that a large percentage of cows wean live and healthy calves every year for herd replacement and income generation. Rearing healthy dairy calves to weaning time requires maximizing the calf's level of immunity against disease while minimizing its exposure to infectious agents (Godden, 2008). As per respondents' response, majority (61.70%) of weaning criteria of calves was based on age of calves and body condition of their dams. The remaining percentages were shared by availability of feed for calves (20%) and willingness of cow to nourish milk to her calf (18.30%).

Table 3: Sources of feed, water and weaning criteria

<b>Sources of feed</b>	<b>Frequency</b>	<b>Percentage</b>
Roughages	138	76.70
Concentrates	14	07.80
Both	28	15.60
<b>Sources of water</b>		
Bore holes	12	06.70
Bore holes and pond water	35	19.40
Pond water	88	48.90
Pump and pond water	45	25.00
<b>Age at weaning of calves</b>		
5-6 Months	111	61.67
7-8 Months	42	23.33
9-10 Months	15	8.33
11-12 Months	12	6.67
<b>Weaning criteria for calves</b>		
Availability of feed for calves	36	20.00
Cows willingness	33	18.30
Age of calves and body condition dam	82	61.70

### ***Knowledge on Calf Management and Sources of problem***

It was revealed that most of the respondents (78.33%) used their acquired indigenous knowledge of calf management while 13.89% and 7.78% of them used previous training. The rest 7.78% have no any information about calf management (Figure 2).

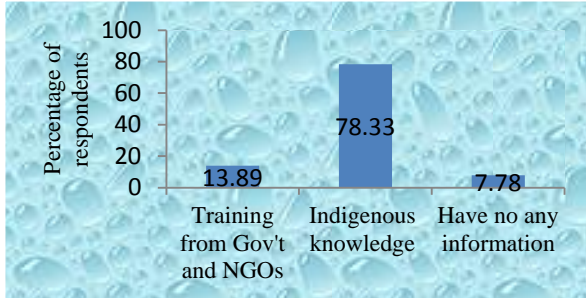


Figure 2. Sources of information on calf management

The major problems of calf management according to the study result were shortage of calf feeds including water, calves diseases, drought, shortage of grazing land and lack of government attention (Figure 3).

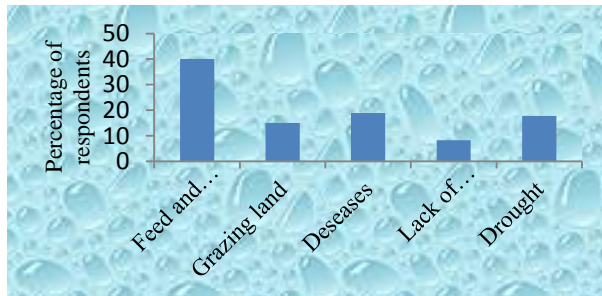


Figure 3. Major problems of calf management

### *Causes of calf morbidity and mortality*

The most common diseases of calf were listed in the table 4 below. Based on owners, response, FMD was ranked first followed by heart water and others (Table 4). According to Lorenz *et al.* (2011), calf morbidity and mortality have short-term and long-term detrimental effects on performance of a dairy farm. Among the factors that have been hindering success of dairy industry, morbidity and mortality of calves is one that causes major concern (Acha *et al.*, 2004). It was also noted that morbidity and mortality are important causes of economic losses on dairy farms worldwide (Phiri, 2008).

Table 4: Most common disease ranked by pastoral and agro-pastoral that causes calf morbidity

Main disease	Rank	X <sup>2</sup>	P-value
Foot moth disease	1	24.653	0.017
Heart water	2	29.290	0.004
Lumpy skin disease	3	9.407	0.668
Contagious bovine pleuropneumonia	4	15.297	0.226
Pasteurellosis	5	7.656	0.811
Diarrhea	6	18.570	0.099
Liver fluke	7	13.311	0.347

X<sup>2</sup> = Chi-square; P-value, significant at p < 0.05

According to survey respondents on diseases of calves, it was revealed that liver fluke is the first followed by heart water; CBPP, Diarrhea, Pasteurellosis, FMD and Blackleg in the decreasing order (Table 5). The report of Wudu (2008) revealed that calf diseases that cause morbidity and mortality are the results of complex interaction of the management practices, environment, infectious agents and the calf itself. Different management and environmental factors significantly affect replacement stock calf morbidity and mortality are colostrum feeding, housing, calving assistance, production system, herd size, season and hygiene of micro- environment. Similarly, this study indicated that season, mobility time, disease, colostrum feeding, housing and calving assistance were associated risk factors under extensive management practices in the study areas.

Table 5: Most common disease ranked by pastoralists and agro-pastoralists

Major disease	Rank	X <sup>2</sup>	P-value
Liver fluke	1	34.553	0.000
Heart water	2	31.320	0.002
Contagious bovine pleuropneumonia	3	7.647	0.812
Diarrhea	4	5.193	0.951
Pasteurellosis	5	47.377	0.000
Foot moth disease	6	10.878	0.539
Blackleg	7	16.626	0.083

X<sup>2</sup> = Chi-square; P-value, significant at p < 0.05

#### *Clinical observation and laboratory examination of calf diseases*

During observational study on clinical signs of calf diseases, 61.40% (57/92) of calves were diseased. Fecal examination for calves under age of 4 months showed that all were affected by eimeria and nematodes. On the other hand, all calves between 4-6 months of age were affected by eimeria, nematodes and cestodes (Table 6).

Table 6. Health condition of calves

Condition of diseases	Number	Percentage
Diseased calves	54	61.4
Health calves	38	34.6
Total	92	100

Table 7. Calves parasitological result

Factors	Categories	Parasitological examination result			X <sup>2</sup>	P-value
		cestodes	Eimeria	Nematodes sp.		
Age/month	<1	0	3	2	4.863	0.302
	2-4	0	4	3		
	4-6	6	8	15		
Diseases condition	Health	4	2	8	6.016	0.049



	Diseases	2	13	12		
Sex	Female	4	5	10	2.125	0.346
	Male	2	10	10		
Body condition	Good	0	1	3	4.704	0.319
	Medium	2	3	9		
	Poor	4	11	8		

## Conclusion and Recommendations

Interview results about calf management showed poor housing where owners pooled calves together and grouped outside in barn without shade. Though majority of owners have awareness about the importance of colostrum and right time of feeding, some of the owners had little knowledge about it. In the study area, calves start feeding 2 weeks after birth, and supplemented with crop residues during feed shortage. The source of water for calves is pond, bore holes and pump. The average weaning age of calves is  $6.7 \pm 1.80$  months. Majority of weaning criteria of calves' age of calves and body condition their dam. The major problem of calve rearing was feed shortage followed by calf diseases.

Morbidity was associated with foot mouth disease, heart water, lumpy skin disease, contagious bovine pleuropneumonia, pasteurellosis, diarrhea and liver fluke. Liver fluke was the 1<sup>st</sup> ranked disease cause of calf mortality followed by heart water, contagious bovine pleuropneumonia, diarrhea, pasteurellosis, foot mouth disease and blackleg. According to laboratory fecal examination result, all were affected by eimeria and nematodes. Based on the above remarks of conclusion, the following recommendations are forwarded:

- Training and advising should be given to pastoralists and agro pastoralists on the importance of proper management of calves,
- Further study other than parasitological investigation is recommended to identify the causative agents involved in the major health of calves in the study area

## Acknowledgements

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# Identification and characterization of toxic plants of livestock in Borana zone, Southern Oromia, Ethiopia

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## **Abstract**

*A study was conducted to identify and characterize toxic plant poisonous to livestock in Borana Zone. A total of the 120 (46 (38.33%) female and 74 (61.67%) male) interviewed individuals through semi structured questionnaires, 95% respondent was reported the presence of toxic plant in the area and suggest considerable health impacts on the livestock and specified 31 plants as poisonous to livestock in the study area. Among identified toxic plant, Pavetta gardeniifolia (Gaadallaa) (23.63%), Loudetia flavida (Seerrichaa) (10%), Euphorbia tirucalli (1.36%), Solanum somalense (Hiddii gaagee) (3.2%), Eragrostis cilianensis (Ardaa) (17.72%), Sorghum arundinaceum (Fincoo) (17.72%), Acokanthera schimperi (Qaraaruu) (4.1%), Capparis tomentosa (Ogoraa gaalaa) (3.63%), Teclea salicifolia (Hadhaa/hadheessa) (2.27%) were the poisonous plants complained mainly and frequently by the respondents. Major factor expose livestock to toxic plant were rainy season 41.1% (Gannaa and Hagayya), forest area/plateau (42.9%) where animal are frequently graze, shortage of feed in combination with nutritional defecincy (73.6%), ingestion (98.2%) of fresh evergreen and matured whole part (52.6%) and cattle (72.8%) to be the most frequently affected animals. Common clinical symptoms complained by the respondent on the poisonous plants were bloating, diarrhea, depression, incoordination, weakness, salivations, bloody urine, thorn tongue and mouth, loss of appetite, restlessness, unable stand, depression and coma. This study showed toxic plants have major health impact on the livestock. Therefore more intervention is needed on the major toxic principles and phytochemistry of the identified plants. Prevention and control measures which are traditionally practiced among livestock owners should also be supported with further studies to overcome impacts of poisonous plants.*

**Key words:-** Borana, Characterization Identification, Toxic Plant,

## **Introduction**

Varieties of poisonous plants have caused extensive losses to the livestock industry in many parts of the world mainly in East Africa including Ethiopia. They have still significant impacts in numerous areas. Poisonous plants produce their toxic effects that include physical upset, loss of productivity and death after being ingested and/or absorbed by animals (Mekonnen, 1994). Plant poisoning is due to either accidental ingestion along with grass or willful consumption of poisonous plants during feed shortage. It is also more likely to occur in animals that have been moved from one part of the country to another (Bah 2013). Livestock poisoning occurs when animals consume these plants in large amount in a short period or graze over a prolonged period of time (Panter *et al.*, 2007). Animals are exposed to consumption of poisonous plants particularly during drought periods when palatable forages become scarce in the pastureland (Hart and Carpenter 2001; Botha and Penrith 2008). Most of the poisonous plants remain

green and attractive for hungry and thirsty animals. These problems may be aggravated when animals suffer from deficiencies of phosphorus or vitamin A, which can greatly affect grazing behavior of the animals (Hart and Carpenter 2001). The effect of poisonous plants is complex and can affect various organ systems because of presence of several toxic principles in some plants that affect different systems. The dominant effect depends on several factors such as condition, growth stage or part of the plant, the amount ingested, the species, and susceptibility of the victim (Botha and Penrith 2008).

Animals under nutritional stress may be less able to detoxify plant toxins and may suffer relatively greater harm from the metabolic effects of the toxins. Thus, proper observation of grazing animals, good grazing management with prior knowledge of poisonous plants in the rangelands, and strategic supplemental feeding may help to mitigate the problem. This is because separation between losses due to diseases, accidents, predators, ingestion of poisonous plants can be difficult. Low reproductive performance and weight loss can be caused by disease and inadequate nutrition as well as by consumption of poisonous plants. Some adverse effects of poisonous plants such as birth defects occur long after ingestion of poisonous plants (Holechek, 2002).

Toxic plants are still cause significant problems in numerous areas. Poisonous plants produce their toxic effects after being ingested and/or absorbed by animals that include irreversible, or worse, continues damage to escalate progressive liver or kidney disease, physical upset, loss of productivity and death. In lowland area due to the toxic plant problem, owners face the risk of animal pain management and the quality of life for their animal is significantly decreased. In the pastoral area, livelihood is based mainly on livestock and livestock products. The lowland face various life threatening hazards notably infectious and noninfectious like metabolic diseases, poisoning and others of miscellaneous origin (Tashale, 2004). Even though previous report (Bedane *et al.*, 2008) indicated presence of toxic plants in Borana zone, is few information on identification of toxic plant in the area though there is some verbal information about presence of some poisonous plants used for criminal poisoning. Therefore, this study was designed to identify and characterize toxic plants, to assess consequences of poisonous plants, to identify factors that predispose livestock to major toxic plants, and to design control and prevention method of toxic plant in Borana zone, southern oromia.

## **Materials and Methods**

### ***Study Area***

The study was conducted in four districts of Borana zone of Oromia Regional State, southern Ethiopia, which is located at 570-775 Kms South of Addis Ababa, bordering northern Kenya and the Somali Regional State of Ethiopia. The Borana plateau, the portion of the Southern Ethiopia rangelands, comprises an area of about 95,000km<sup>2</sup> (Coppock, 1994). The rainfall is bimodal but erratic and unreliable in distribution. Its distribution is season as long rainy season “*Gannaa*” from March to May, a cold dry season “*Adoolessa*” from June to August, short rainy season “*Hagayya*” from September to November, and a long dry season “*Bonaa*” from December to February.

### ***Sampling Method***

Districts were purposively selected by considering the proximity to the Research center, and availability of of plant; and multi-stage sampling procedure was used to take samples from PAs, ollas, households.

### ***Study population and sample size***

A total of 120 populations were interviewed using a semi-structured separate questionnaire for each group including animal health practitioners in Moyale, Dubluq, Yabello and Dire district of Borana zone

### ***Data collection method***

A total of 120 individuals (30 per kebele) were interviewed by using a semi-structured questionnaire and focus group discussion. Separate semi-structured interviews, field observations, and plant sample collection were undertaken during the study time. All relevant information including the local name of the plants, poisonous parts of the plant, factors exposing livestock to the toxic plants, species of animals poisoned, age group of exposed animals, the effects of poisoning and its control and prevention were gathered during the interviews. Following standard procedure, identification of scientific names of the plants was undertaken and recorded on the data collection sheet.

### ***Data Analysis***

The collected data by questionnaires, names of collected plants and other possible information were managed using Ms-Excel spread sheet, and then analyzed by SPSS version 20. Descriptive statistics was used in the analysis of the data. .

## **Results and Discussion**

### ***Availability of toxic plant***

From those respondent 46 (38.33%) were female and 74 (61.67%) were male (Table 1).and specified 31 plants as poisonous to livestock in the study area. Out of the total of 120 interviewed individuals, 95% reported presence of toxic plants in the area.

Table 1. Proportion of respondents reported the presence of poisonous plants to livestock in relation to districts according to sex division of respondent.

Districts	% of respondents on availability of toxic plant		Sex of respondent		Total
	No	Yes	Female	Male	
Dire	-	25	14 (11.7)	16 (13.3)	30
Dubuluk	4.16	20.83	9 (7.5)	21 (17.5)	30
Moyale	-	25	13 (10.83)	17 (14.2)	30
Yabelo	0.83	24.16	10 (8.3)	20 (16.7)	30
Total	5	95	46 (38.33%)	74 (61.67%)	120

### ***Risks Factor of Exposure***

According to respondents, animal exposure to toxic plant was 41.1% (Gannaa and hagayya), 40.3% (Bonnaa ), 16.6% (All seasons)and 1.7% (cold dry season called adolessa) based on seasonal difference in decreasing order. This agrees with the finding in Horo Guduru Wollega (Diriba and Debela 2019), Wondo Genet ( Abdallahi *et al.*,2020) and peri-urban *kebeles* of Woliso and Wonchi (Biruk *et al.*, 2018) and this might be due to the extensive growth and evergreening nature of the plant during animal feed shortage. Most of the poisonous plants were reported to occur in forest area/plateau (42.9%) where animals frequently graze followed by Farm land (24.5%), Rangeland/road side (15.7%), dung decomposed fertile area (15.7%) and water points (0.8%) were the next areas where these toxic plants were claimed to be found, in decreasing order. The present study agrees with the study undertaken in Woliso and Wonchi towns by Biruk (et al., 2018) and lower than the finding of (Abdallahi *et al.*, 2020) whose report showed 39.2% (rangeland) and 31.4% (farmland) , and higher than forest (20.4%)

The present study showed that shortage of feed in combination with nutritional deficiency (73.6%), sudden consumption with grass (22.8%), shortage of water (2.6%) and accidental body contact (0.8%) were major predisposing factor of animals to the poisonous plants. This also agrees with previous works (Yoobsan Fikaduand and Girma Kebede, 2018, Abdallahi *et al.*, 2020) and this may due growth of together with major forage plants and readily accessible to grazing animals (Marczewski, 1983; Panter *et al.*, 2011). Hungry animals are less selective of forages and consume large quantities of toxic plants within a short period of time (Rook *et al.*, 2004) while animals on a good plane of nutrition are less likely to eat poisonous plants and are better able to detoxify the small amount consumed (Lopez-Ortiz *et al.*, 2004). Nutritional deficiency can also increase the probability of livestock ingesting toxic plants. The result of this study was also in agreement with the findings reported in the Sokoto state, Nigeria (Ebbo *et al.*, 2003, Onyeyili *et al.*, 1996) who reported similar risk factors, and the finding reported in North Central America by Martison *et al.* (2006).

Regarding with poisoning of livestock by parts of plants, the study revealed that ingestion or contact of fresh evergreen and matured whole part contribute 52.6% followed by Leaves and seed (34.2%), fresh and dry leaves (11.4%), Seed (1.7%) of toxic plant (Table 2). This is in agreement with research findings from Nekemte town area (Kebede., *et al.* 2015), central Ethiopia (Abera D., *et al.*, 2014) and Wondo Genet (abdallahi *et al.*, 2020).

According to respondents, cattle are the most frequently affected animals. This agrees with the finding of Dereje *et al.* (2015). Large animals spend more time foraging and are less selective. In contrast, small ruminants requires less feed, but are more selective feeders and spend more time searching for high-quality forage (Lyons and Machen, 2000; Mosavat and Chamani, 2013); therefore, are less sensitive to poisonous plants (Hernandez and Sanchez, 2014; Dereje *et al.*, 2015).

Table 2. Summary of risks of exposure to toxic plants

Variables	Risk factor for toxic plant exposure	No of respondent	Percent
<b>common habitat /Distribution/</b>	Everywhere/range	18	15.7
	Forest area/plateau	49	42.9
	Farm land	28	24.5
	Fertile area decomposed with dung	18	15.7
	Around ela/water point	1	0.8
<b>Toxic part</b>	Leaves	13	11.4
	evergreen and matured whole part	60	52.6
	Leaves and seed	39	34.2
	Seed	2	1.7
<b>Abundant season</b>	Hagayya	3	2.6
	Bona	46	40.3
	Ganna	44	38.5
	All season	19	16.6
	Adolessa	2	1.7
<b>Mode of infection</b>	Ingestion	112	98.2
	Body Contact	2	1.7
<b>Effective form</b>	Leaves	9	7.8
	fresh and matured all part	37	32.4
	matured seed	68	59.6
<b>Reason of exposure</b>	deficiency/shortage of feed	84	73.6
	sudden consumption with grass	26	22.8
	accidental body contact	1	0.8
	shortage of water	3	2.6
<b>Frequencies of exposure</b>	Repeated exposure/multiple	83	72.8
	Single	31	27.2
<b>Animal species</b>	Bovine	83	72.8
	Camel	6	5.3
	Camel, Bovine, Caprine	11	9.6
	Bovine and Caprine	12	10.5
	All Animals	2	1.8

### ***Toxic Plants Reported in the study area***

Among identified toxic plants, *Pavetta gardeniifolia* (23.63%), *Eragrostis cilianensis* (17.72), *Loudetia flavida* (10%), *Acokanthera schimperi* (4.1), *Capparis tomentosa* (3.63), *Solanum* sp (3.2), *Teclea salicifolia* (2.27), *Euphorbia tirucalli* (1.36), were some of the poisonous plants complained mainly and frequently by the respondents. Most of these identified plants in this study were similar to those plants identified in Borana Zone and Some Liben zone of Moyale area by Aster Abebe et.al (2011) and documented finding of Temesgen Kassa and Wondimu Debash (2019) in Holeta. Gaaddallaa, Marra dhigaa, Qorsa diidaa, Mishingaa, Ardaa, Tabari, Ogora gaalaa, Hadha/hadheessa/, Bobiya, Finchoo, Aannoo, Hidi, Qobboo were reported from the present study were also reported in the previous study of Aster *et al.*(2011) in Borana Zone

Some of the identified poisonous plants in the study area were also reported from other parts of Ethiopia were *Datura stramonium*, *Sorghum bicola*, *Solanum incanum* *Ricinus communis*, *Amaranthus spp* (Abdallahi et al., 2020, Yoobsan and Girma, 2018, Biruk et al., 2018, Diriba and Debela, 2019, Angesom Desta, 2019), *Euphorba tirucalli* (Biruk et al, 2018, Abdallahi et al., 2020), *Olea africana* (Biruk et al., 2018), *Aloe species* (Abdellahi et al., 2020), *Euphorbia sp* (Diriba and Debela, 2019) were reported from Woliso and Wonchi towns, Bako, Horo Buluk and Afar respectively.

Common clinical symptoms complained by the respondent on the poisonous plants were bloating, diarrhea, depression, incoordination, weakness, salivations, bloody urine, thorn tongue and mouth, loss of appetite, restlessness, inability to stand, depression and coma. Other symptom were draining foam from mouth, enlarged belly for longtime, vomiting and diarrhea, bad smells and bitter taste of milk, constipation and inability to defecate to dry feaces, blindness, discomfort, loss of lip movement, foam discharge, inability to stand, Pain, irritation and swelling of skin, abortion, respiratory problem, muzzle dryness and respiratory disturbance (Table 5).

The dominant effect depends on several factors such as condition of plant, stage of plant growth, part of the plant, the amount ingested, species of the plant, and susceptibility of the victim (Botha and Penrith, 2008). Most of the respondents in study area reported that animals poisoned by *P. gardeniifolia* do not show any symptoms before death and that the poisoning can be confirmed by observation of seed of the plant in gastrointeste of the animal. However some of the respondents complained that animals poisoned by the plant show symptoms like bloating and diarrhea. *Sorghum arundinaceum* and *Loudetia flavida* cause animal to urinate blood and hiddii gaagee make animals blind. This finding is similar to the finding of Aster Abebe et.al (2011) in Borana Zone and Some Liben zone of Moyale area and the finding of Temesgen Kassa and Wondimu Debash (2019) in Holeta.



Table 3. Summary of poisonous plants and their characteristics

Local name	Scientific name	Frequency %	flowering time	Flower colour	Distribution/common habitat	Toxic part	Abundant season
Gaadallaa	<i>Pavetta gardeniifolia</i>	52 (23.63)	bona	white	forest	leaves and seed	bona
Ardaa	<i>Eragrostis cilianensis</i> (All.) Vign. ex Janchen	28 (17.72)	bona	white	fertile cattle barn area	all part	gana
Fincoo	<i>Sorghum arundinaceum</i> (Desv.) Stapf	28 (17.72)	adolessa	white	farm land	all part	gana
Seerricha/marra dhiigaa	<i>Loudetia flavida</i> (Stapf) C. E. Hubb	22 (10)	bona	red	forest	all part	all season
Tabarii	NA	15 (6.82)	bona	white	Everywhere/rang	all part	all season
Qaraaruu	<i>Acokanthera schimperi</i> (A. DC.) Schweinf	9 (4.1)	bona	white	forest	leaves and seed	all season
Ogoraa gaalaa	<i>Capparis tomentosa</i> Lam	8(3.63)	adolessa	yellow	Forest	leaves	bona
Hiddii gaagee	<i>Solanum somalense</i> Franchet	7 (3.2)	bona	white	everywhere/range	only seed	bona
Hadheessaa	<i>Teclea salicifolia</i> Engl.	5 (2.27)	adolessa	white	forest	Leaves	adolessa
Aannoo	<i>Euphorbia tirucalli</i> L	3 (1.36)	adolessa	white	everywhere/range	all part	adolessa
Ejersa	<i>Olea europaea</i> subsp. <i>cuspidata</i> (Wall. Ex. G. Don) Cif.	3 (1.36)	bona	white	forest	leaves	adolessa
Banjii	<i>Datura stramonium</i>	3 (1.36)	bona	white	Range	Leaves/seed	Gana
Kuubaa	<i>Partinum hysterophorus</i>	3 (1.36)	bona	white	everywhere/range	all part	bona
Qobboo	<i>Ricinus communis</i> L	3 (1.36)	bona	white	on the fertile area of removed barn	can be leaves and/or seed	bona
Argeessa	<i>Aloe species</i>	2 (0.9)	bona	red	everywhere/range	leaves	bona
Boobiyaa	<i>Kalanchoe</i> sp	2 (0.9)	No flower	no any colour	forest	all part	bona
Doobbii	<i>Tragia pungens</i> (Forssk.)	2 (0.9)	all season	white	removed cattle barn	all part	bona

	Mu'll. Arg.				fertile with dung		
Halquuqaa	<i>Haricot bean</i>	2 (0.9)	bona	red	Farm land	only seed	bona
Haxaawwoo	<i>Becium verticillifolium</i> (Bake) Cufod.	2 (0.9)	bona	white	farm land	leaves	bona and ganna
Qorsa diidaa	<i>Euphorbia</i> sp	2 (0.9)	bona	red	removed cattle barn fertile with dung	all part	adoolessa
Darguu	<i>Tephrosia pentaphylla</i> (Roxb.) G. Don.	1 (0.45)	bona	white	Water point	leaves	bona
Gaalee	<i>Psydrax schimperiana</i> (A. Rich.) Bridson	1 (0.45)	bona	white	forest	all part	bona
Gambora	Cactus	1 (0.45)	bona	red	everywhere/range	leaves/spine	adoolessa
Hadaa	<i>Osteospermum vaillantii</i> (Decne.) T. Norl.	1 (0.45)	bona	yellow	everywhere/range	leaves	bona and ganna
Hiddii loonii	<i>Solanum giganteum</i> Jacq.	1 (0.45)	bona	yellow	everywhere/range	only seed	bona
Hiddii waatoo	<i>Solanum incanum</i> L.	1 (0.45)	adolessa	white	everywhere/range	leaves	bona
Illaadduu	<i>Echinochloa haploclada</i> (Stapf) Stpaf	1 (0.45)	adolessa	green	around ELA	leaves	gana
Luuccolee	<i>Bothriochloa insculpta</i> (A. Rich.) A. Camus	1 (0.45)	bona	red	forest	all part	adoolessa
Raafuu	<i>Amaranthus thunbergii</i> Moq.	1 (0.45)	bona	white	forest	leaves and seed	gana
Qundhii		1 (0.45)	bona	red	around ELA	leaves and seed	gana
Missingaa	<i>Sorgum bicolar</i>	1 (0.45)	hagayya and ganna	white	farm land	leaves and seed	gana
Tummaa		1 (0.45)	No flower	no colour	everywhere/range	leaves	gana

Table 5. Effect of poisonous plant on livestock

Local Name	Scientific name	Animal Species	Age	Mode of infection	Clinical Sign	Treatments
Gaadallaa		Bovine	all age	Ingestion	bloating, diarrhea, unable stand, depression, coma death	
Ardaa		Bovine	all age	Ingestion	Bloating, draining foam from mouth and incoordination, sometimes cause death	
Fincoo		Bovine	all age	Ingestion	Bloody urine, bloating, weakness, incoordination, , salivation and death	
Seericha		Bovine, Caprine	all age	Ingestion	Urination with blood, causes diarrhea, incoordination, tongue trough sometimes bloating, finally death	
Tabarii		Camel, Bovine, Caprine	all age	Ingestion	enlarged belly for longtime and staggering walk and unable to stand from the hind	
Qaraaruu		Bovine, Caprine	all age	Ingestion	Seed make animal to vomit and diarrhea, leaves change the milk taste	
Goraa gaalaa		Camel	young	Ingestion	bloating, diarrhea, depression	
Hiddii gaagee		Camel, Bovine, Caprine	all age	Ingestion	bloating, incoordination, red urine, constipation, unable to defecate, sometimes make blind	
Hadheessaa		Ovine	all age	Ingestion	Loss of appetite Weakness	
Aannoo		Camel and caprine	young	Ingestion	Stress and discomfort	
Ejersa		Bovine, Caprine	all age	Ingestion	diarrhea	
Banjii		Caprine	adult	Ingestion	Stress	
Kuubaa		Bovine	all age	Ingestion	milk has Bitter taste and smells bad and animal loss body weight	
Qobboo		Bovine	all age	Ingestion	Incoordination	
Argeessa		Bovine	all age	Ingestion	Incoordination and vomiting	
Boobiyaa		Camel	adult	Ingestion	loss of lip movement, foam discharge, lastly unable to stand	
Doobbii		All Animal	all age	Contact	Pain, irritation and swelling of skin over contacted area	
Halquuqaa		Caprine	all age	Ingestion	bloating, diarrhea, restlessness, stomach ache and death	
Haxaawwoo		Bovine	adult	Ingestion		

Qorsa Diidaa		Bovine, Caprine	young	Ingestion	Constipation and death	
Darguu		Camel	adult	Ingestion	swelling around belly region, un able to defecate dry feaces	
Gaalee		Bovine	all age	Ingestion	Slow digestion	
Gambora		Bovin/caprine	all age	Ingestion	Bleeding from mouth, animal stand and unable to eat	
Hadaa		Bovine	adult	Ingestion	Diarrhea	
Hadhawa		Bovine	all age	Ingestion	Bloating, if animal take to high level it cause incoordination	
Hiddii loonii		Bovine	all age	Ingestion		
Hiddii Waatoo		Camel, Bovine, Caprine	all age	Ingestion	Animal may abort	
Illaadduu		Bovine, Caprine	all age	Ingestion	Bloating	
Luuccolee		Bovine	all age	Ingestion	Diarrhea	
Raafuu		Bovine, Caprine and ovine	all age	Ingestion	Bloating, Loss of appetite	
Qundhii		Bovine	adult	Ingestion	Thorn cause mouth bleeding and Emaciation	
Missingaa		Bovine, Caprine	all age	Ingestion	Bloating, diarrhea, restless and incoordination, respiratory problem and death	
Tummaa		Bovine	all age	Ingestion	muzzle dryness, respiratory disturbance, depression	

## Conclusion and Recommendations

In the study area thirty one poisonous plants to livestock were identified. Most frequently mentioned includes *Pavetta gardeniifolia*, *Loudebia flavida*, *Euphorbia tirucalli*, *Solanum somalense*, *Eragrostis cilianensis*, *Sorghum arundinaceum*, *Acokanthera schimperi*, *Capparis tomentosa*, *Teclea salicifolia*. The common symptoms animals show during exposure to the plants as respondents claimed are bloating, diarrhea, depression, incoordination, weakness, salivations, bloody urine, thorn tongue and mouth, loss of appetite, restlessness, unable stand, depression and coma. As part of recommendation preventive ways and use of remedies which are traditionally practiced by livestock owners should be supported, Intervention on the major toxic principles and phytochemistry in the different identified plant species is needed, and Further study on plant dynamics and compatibility with livestock production is required. to overcome impacts of poisonous plants.

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# Veterinary Drugs Handling and Antibiotic Resistance Assessment in Borana Zone, Southern Ethiopia

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## **Abstract**

*A study was conducted on in Borana zone with the objectives of assessing veterinary drug handling and identifying causes of antimicrobial resistance. Semi-structured questionnaires and Kirby–Bauer disc diffusion method were used during the study. According to respondents tetracyclines (94.4%), procaine penicillins (91.1%), ivermectin (78.3%), albendazole (63.3%) diazinon (25%) and trimethoprim-sulfonamides (18.9%) were the most dominantly used classes of veterinary drugs drugs in the study area. Out of the total respondent pastoralists 66.7% buy and administer veterinary drugs by themselves. With regard to antibiotic use 71.7% of the respondents use them to treat bloat, 66.7% use to treat any diseases of animals , 76.1% for parasitic cases, 53.3% in the absence of any disease symptoms From this study, most of pastoralists use antibiotics to treat any diseases of animals (Majority (89.4%) of the respondents consume milk within a day of treatment without considering drug withdrawal period and only 45.6% of the respondents have awareness on drug residue in food items of animal origin. Regarding drug handling, 68.9% of respondents stored antibiotic leftover for next use. Only 57.2% of pastoralists dispose of expired drugs in inappropriate place. 80% of animal health professionals in the study area transport veterinary drugs by public transport and 24% of them store drugs according to manufacturer. Moreover, 20%, 92% and 64% of animal health providers follow-up of treated animals, inform drug withdrawal periods pastoralists and manage safe handling of drugs starting from acquisition to handover to pastoralists, respectively. Generally, inappropriate use of antimicrobials like sharing antibiotics between humans and animals and the awareness related to the antimicrobial residues in animal food items (milk, meat and eggs) have been take placed in the way that encourage the emergence of Antimicrobial resistance. Inclusively, drugs Handling and management plus treating sick live stocks either by health professional or pastoralists have been found very poor. Awareness creation on proper drug transportation and storage, practice of selling drugs with prescription, proper disposal of expired drugs, dose determination, follow-up of treated animals and determining drug withdrawal periods starting from acquisition to end users should to be made*

**Key words:** - Antibiotic Resistance, Assessment, Borana, Handling, Veterinary Drugs,

## **Introduction**

Antibiotics serve many purposes beyond treating “routine” bacterial infections. Antibiotics are often used after a medical treatment, as well as an important addition to the treatment. Thus, antibiotics are essential to saving individuals from infection (Piddock, 2012). Loss to the ability of antibiotic against bacteria and other microbes develop when they resist the effects. A primary characteristic of antibiotics is that they lose their effectiveness over time. Appropriate use of existing classes of antibiotics could improve the lifespan of these drugs. The side effects of antibiotic resistance include reduced patient outcomes and increasingly potent disease states. They have been used to improve animal health for food production



(Amyes, 2000). Using antibiotics is a complex subject involving the prescriber, the dispenser, the patient, and pharmaceutical institutions. It is influenced by factors such as drug availability, the prescriber's experience and knowledge of dispensers.

Inappropriate drug use is a problem of the whole world; however, the degree of the problem is higher in developing countries like Ethiopia (Sisay et al., 2017; Mensa et al., 2017). Improper use of drugs may cause ineffective treatment and unnecessary wastage of resources and may harm the patient (DACA, 2006.) Antimicrobials are mainly used in the production of swine, cattle, and poultry and, recently, in aquaculture and crop production (Ilbergeld *et al.*, 2008; Carlet et al., 2012). To reduce the volume of antibiotics used in veterinary medicine and curtail the selection of resistant bacteria, scientists and the WHO (WHO, 2011) have suggested improved hygiene-based husbandry methods, veterinary supervision, and antibiotic dispensing under prescriptions only to policymakers and governments. Because of the central role of antibiotics in disease management, a study of prevalent diseases and their management practices will aid in informing practical interventions that will aim at reducing the bacterial use in every parts of case (Abebe, 2003).

Antibiotics tend to break down with time. This occurs by oxidation-reduction reactions, hydrolysis, biodegradation, or photo degradation (Halling-Sørensen, 2000; Chen *et al.*, 1997). Such processes reduce the concentration of the drugs and their ability to kill the bacteria they come in contact with, allowing the exposed bacteria time to develop resistance. Environmental factors influence these processes, making storage conditions of antimicrobial very important (Thiele-Bruhn, 2003). Consequently, describing and containing the level of such dissemination are critical to safeguarding the environment from these health hazards. Antibiotic resistance occurs when bacteria or other microbes have the ability to resist the effects of an antibiotic. Bacteria or microbes change and reduce the overall effectiveness of drugs or the chemicals in the drug itself (Hopkins et al., 2013). Further, microbes can develop resistance to drugs when ability of drugs is reduced as results of storage and transportation under adverse conditions which damages quality of antibiotics.

Manufacturers' recommendations concerning storage temperatures should be observed and this may involve the use of specialized storage and transport facilities. Temperature monitoring devices should be used to demonstrate compliance with the designated temperature ranges (Hughes, 2011). Moreover, the accumulation of antibiotics in adipose tissues (Rang *et al.*, 2003) after their administration (antibiotic residues) has been shown to further the selection of resistant phenotype (Wageh *et al.*, 2013); Borgen *et al.*, 2000). To tackle these side effects, proper antibiotic administration practices like accurate dosing, route of administration, observance of withdrawal periods, and strict adherence to manufacturers' instructions major detrimental effect.

In Ethiopia, conventional veterinary services have been playing a paramount role in the control and prophylaxis of livestock diseases in the last three decades. However, they cannot yet deliver complete coverage in preventive and curative health care practices because of inadequate labor, logistical problems, an erratic supply of drugs, and the high cost of drugs and equipment. Consequently, the majority of those raising stock in rural areas especially are far from the site of veterinary stations, and those who have access to veterinary services may not be able to afford to pay for them. Additionally, reduced funding for animal disease control is an issue in Ethiopia and is likely to influence the incidence of some serious

livestock diseases (Abebe, 2003, Sori, 2004). Pastoralists are raising the problem of veterinary drugs handling and antimicrobial resistance which are widely occurring events. Therefore, the general objective of this study is to assess the handling and usage of veterinary drugs and development of antimicrobial resistance in Borana zone southern Ethiopia.

## **Material and Methods**

### ***Description of Study Areas***

The study area was conducted in Borana zone in Elwaye, Dire, Dubluk, Yabello and Gomole districts. The area is located 4.6<sup>0</sup>N and 36.42<sup>0</sup>E sloping from 1600 meter above sea level (masl) in the north-east to about 1000 masl in the extreme south that borders northern Kenya. Spatial and temporal variability in both the quality and distribution of rainfall renders the area semi-arid, with an average annual rainfall varying from 353 to 873mm per annum (McCarthy *et al.*, 2002).

### ***Study Population and Data Collection***

By using cross sectional study design, the study was conducted by including 180 respondents from both pastorals and agro-pastorals and animal health professionals in five districts of Borana zone.

### ***Methods of Data Collection***

#### ***Questionnaire survey***

Semi-structured questionnaire surveys were undertaken to assess the handling and usage of veterinary drugs and antimicrobial resistance in the study area. The questionnaires were pre-tested with special attention on information about veterinary drugs handling and management practices during acquisition, transportation, storage, dispensing, disposal of expired drugs and drug resistance problem.

### ***Antimicrobial Susceptibility Test (AST)***

Bacteriological isolation and identification of the common bacterial causative agents of mastitis was undertaken on 50 milk samples that had CMT scores of more than 2 according to National Mastitis Council guideline (Quinn, 2010). Briefly, a loopful (10 µL) of milk sample was inoculated into blood agar with 5% sheep blood (blood agar plates) (Oxoid, Basingstoke, UK) and incubated at 37<sup>0</sup>C for 24–48hrs. Plates were evaluated for the growth of bacteria at 24 and 48 h of incubation and colony characteristics, pigment production, and hemolysis were recorded. Sub-culture was performed on nutrient agar (Oxoid, Basingstoke, UK) to get pure colonies. Gram staining was performed per pure culture and differentiated into Gram-positive and Gram-negative bacteria. The pure colonies were sub-cultured on selective media for both gram positive (staphylococcus and streptococcus species) and gram negative bacteria (*Enterobacteriaceae*). The biochemical tests used included lactose fermentation, indole production, the methyl red test, Voges–Proskauer, citrate utilization, hydrogen sulfide in TSI agar, lysine decarboxylase, and urease activity. Each pure bacterial isolate was inoculated into tryptic soy broth (TSB) (Oxoid) and grown overnight at 37 °C, and 500 µL of the overnight culture (16–18 h) for additional tests. Following selective sub-culture, biochemical tests were undertaken for catalase and oxidase tests.

Catalase positive cocci were considered as staphylococci while oxidase negatives were considered as Enterobacteriaceae.

Antimicrobial susceptibility testing was conducted for 12 bacterial isolates from 50 milk samples and obtained of *S. aureus*, *S. intermedius* and *E. coli* using the Kirby–Bauer disc diffusion method on Mueller–Hinton agar (Wayne, 2018). The isolates were tested against a panel of five antimicrobials (Oxoid): Penicillin, Amoxicillin, Doxycycline, Nocardin and Gentamycin. For each isolate, the inhibition zone diameter was measured to the nearest millimeter by using digital caliper, and the result was interpreted as susceptible (S), intermediate (I), or resistant (R) (Wayne, 2018).

**Statistical Analysis:**

- The data was managed Microsoft Excel spread sheet and analysed by SPSS Version 20.

**Results**

**Demographic Information of the respondents**

Out of 180 of respondents, 128 (71.1%) were male and 52 (28.9%) were female. Majority of the respondents (45%) were in the age range of 25-55 years. Amongst the respondents, 94.4% were those married, 88.3% were illiterate and 77.8% were Waaqeffataa (Table 1.)

Table 1. Demographics of the livestock owners

Variable	Categories	Number	%
Sex of respondent	Male	128	71.1
	Female	52	28.9
Age of respondent	≤25	53	29.4
	25-55	81	45.0
	≥55	46	25.6
Marital status	Married	170	94.4
	Unmarried	4	2.2
	Widowed	6	3.3
Educational status	Never go to school	159	88.3
	primary school	19	10.6
	secondary school	2	1.1
Religion	Muslim	25	13.9
	protestant	15	8.3
	Wakefata	140	77.8

**Commonly Used Veterinary Drugs in the Study area**

Tetracyclines (94.4%), Penicillins (Penstrip interchangeably used by the owner) (91.1%), Ivermectin (78.3%), Albendazole (63.3%) Diaznonen (25%) and trimethoprim-sulfonamides (18.9%) were the most dominantly used classes of drugs reported by the livestock owners (Table 3).

Table 3. Common veterinary drugs used by the livestock owner (N=180)

Commonly Used Drugs	Frequency	Percentage
Oxytetracycline	170	94.4
Penstrip/penicillin/	164	91.1
Ivermectin	141	78.3
Albendazole	114	63.3
Diaznone	45	25
Trimethoprim-sulfonamides	34	18.9
Multivitamin	10	1.1

### *Veterinary Drug Handling Practices in the Study Area*

Majority of pastoralists (66.7%) buy drugs for for treatment of bloating of animals (71.7%), in parasitic cases (76.1%), in the absence of diseases (53.3%) and to treat any kind of diseases (66.7%). Inappropriate use of antimicrobials, especially antibiotics shared between humans and animals, plays a leading role in the emergence of AMR (Alanis, 2005). The awareness related to antimicrobial residues in the animal food items (milk, meat and eggs) was 45.6%. On the other side, 63.9% of respondent immediately free their animal after treatment without quarantine. Additionally, 89.4% consume milk within a day of treatment without considering drug withdrawal period. According to respondents, 68.9% store of owners store leftover antibiotics for next use Owner follow various systems for expired drugs: 35.6% dispose add in the hole 35% dump in the toilet, 10.6% leave drugs in the space, 10% store to use in the future and 2.2% are not concerned about expire of drugs ( (Table 4).

Table 4. Knowledge and practice of veterinary drug handling and management by animal owners

Focused Questionnaires Item	Response Categories	Frequency	Percentage
Treatment option in sick animals	Take to veterinary clinic	40	22.2
	Buy drugs for animals	120	66.7
	Ask CAHWs	20	11.1
Human drugs usage in treating sick animals?	Yes	30	16.7
	No	150	83.3
Immediate release of treated animals?	Yes	115	63.9
	No	65	36.1
Awareness and knowledge of antimicrobial residues	Yes	82	45.6
	No	98	54.4
Use of Antibiotics to treat bloated animals	Yes	129	71.7
	No	51	28.3
use of antibiotics In Parasitic diseases animals	Yes	137	76.1
	No	43	23.9
Use of antibiotic in the absence of disease.	Yes	96	53.3
	No	84	46.7
Milk consumption within a day after	Yes	161	89.4

treatment of animals with antibiotics?	No	19	10.6
Use of Antibiotics to treat any kind of diseases	Yes	120	66.7
	No	60	33.3
keeping leftover antibiotic for future use	Yes	127	70.6
	No	53	29.4
consumption of meat from animals that were just treated with antimicrobials?	Yes	162	90
	No	18	10
Source of animal drugs	Market/roadside	40	22.2
	vet drug shop	60	33.3
	vet clinic	20	11.1
	CAHWS	10	5.6
	Human pharmacy	30	16.7
	Both private and vet clinic	20	11.1
transportation of drugs to your area	On foot	125	69.4
	By public transport	14	7.8
	motor cycle	41	22.8
storage methods of drugs in home	Box	119	66.1
	Utensil/qarxaasa/	27	15
	Bag/basket	30	16.7
	Store in the maize storage	4	2.2
Disposal methods of expired drugs?	toilet	63	35
	Not concerned	4	2.2
	Drop in the hole	64	35.6
	leave in the environment	19	10.6
	Use nest time	18	10
	Toilet and hole	12	6.7

### ***Handling and Management of Drugs by Animal Health Professionals***

From the total of animal health professionals included in the study, 80% use public vehicle to transport drugs. Only 24% store drugs properly according to the manufacturer's directions. Moreover, study revealed that 20%, 92% and 64% use follow-up of treated animals, inform drug withdrawal periods to end users and manage safe handling of drugs starting from acquisition by owners to end of drugs, respectively (Table 5).

Table 5:- Knowledge and practices of animal health professionals on safe handling and management of drugs (N=25)

<b>Focused questionnaire items</b>	<b>Response category</b>	<b>Frequency</b>	<b>Percent</b>
Transportation of drugs	Public transport	20	80
	Special vehicle	0	0
	Motor cycle	5	20
Proper drug storage practice	Proper	6	24
	Improper	19	76
Selling drugs without prescription	Yes	25	100
	No	0	0
Diagnosis of animal diseases	Tentative	21	84
	Confirmatory	4	16
Dose determination using age and body weight	Yes	18	72
	No	7	28
Follow-up of treated animals	Yes	5	20
	No	20	80
Informing drug withdrawal periods to end users	Yes	23	92
	No	2	8
Knowledge of safe handling and management of drugs starting from acquisition to end users	Yes	16	64
	No	9	36
Disposal of expired drugs	Burning	3	12
	Burying	18	72
	Not yet	4	16

### ***Antibiotic resistance***

Bacterial isolates were detected in 24% of the milk samples collected from the cows. The highest resistance was observed to amoxicillin (100%; n = 12), followed by penicillin which is 83%, doxycycline (50%), nocardin (41.67%) and gentamycin (16.67%) as shown in table 7. From the most bacterial isolates Antimicrobial susceptibility test was on the three isolates of bacteria (*S. aureus*, *S. intermedius* and *E. coli*) showed that gentamycin is effective (16.67% resistant) drug of mastitis compared to the other in controlling the bacterial isolates in the milk samples.

Table 7. Antimicrobial resistance to bacterial isolates

<b>Bacteria isolates</b>	<b>Number of isolate resistant on Antimicrobial disc</b>				
	Penicillin	Amoxicillin	Doxycycline	Nocardin	Gentamycin
<i>S. auerus</i> (7)	5(71.4%)	7(100%)	2(28.6%)	1(14.3%)	0
<i>S. intermedius</i> (2)	2(100%)	2(100%)	1(50%)	1(50%)	0
<i>E.coli</i> (3)	3(100%)	3(100%)	3(100%)	3(100%)	2(66.7%)
<b>Total</b>	10	12	6	5	2

## Result and Discussion

Regarding antimicrobial usage in livestock, this study found that major pastoralists rely on their own judgment to treat sick animals, buy drugs without prescriptions and not concerned about drugs withdrawal period and expired date. Access to antimicrobials without prescriptions resulted in increased risk for antimicrobial resistant pathogens. This result agrees with previous reports from elsewhere in Africa (Hair, 2007, Wageh *et al.*, 2013).

The use of tetracyclines groups (94.4%), penicillins (Penstrep interchangeably used by the owner) (91.1%) and trimethoprim-sulfonamides (18.9%) were the most dominantly used classes of antibiotics in the study area. This agrees with the findings of (Hair, 2007; Chen *et al.*, 1997, Sori, 2004, Biruk *et al.*, 2020) in the pastoral production systems and elsewhere who reported that antimicrobials are frequently used in food animals in Africa. Inappropriate use of antimicrobials, especially antibiotics shared between humans and animals, plays a leading role in the emergence of antimicrobial resistance (Alanis, 2005). Antimicrobial use may vary widely between and within countries, species, production systems, and individual farms (Thiele-Bruhn, 2003). Despite the frequent use of antimicrobial usage by pastoralists to maintain good livestock health and production in the studied settlements, there was overall low level of knowledge and awareness about proper usage. Such misuse of drugs might result in antimicrobial resistant by pathogens and emergence of drug resistant pathogens. The poor knowledge in the use of veterinary drugs for food animals in the study area as per the result leads to the risk of exposure of consumers to unwanted hazards.

It is noted that public transport (80%) and motor cycle (20%) without referring the manufacturer's direction can expose the drugs to sunlight, unadjusted temperature, humidity, and other conditions including physical damage to the containers which disturb the stability of the drugs. The other most unethical practices in the study area were selling antibiotics without the prescription, improper disposal of expired antibiotic, and drugs storage methods. These findings were inconsistent with finding of Gameda *et al.*, 2020 in Ethiopia. Poor drug storage conditions further enhance the risk of antimicrobial resistance, which agrees with Thiele-Bruhn, 2003 Without considering the drug withdrawal period the drug residue available in milk/meat may expose human to various chemicals which may cause hypersensitivity/allergic reactions, mutagenicity, and cancer (Hughes, 2011).

From the bacterial isolates, antimicrobial susceptibility testing was performed on the three isolates of bacteria (*S. aureus*, *S. intermedius* and *E. coli*) mastitis compared to the other in the bacterial isolates of the milk samples. These results were relatively similar to the previous report by Regassa *et al.*, 2013, Balemi *et al.*, 2021 who reported the highest prevalence of *S. aureus*, 12.8% and 2.9%, at the animal and quarter levels, respectively, in camels in the Borana Zone. Similarly, Haftu *et al.*(2012) also reported *S. aureus* (36%) and *E. coli* (27.3%) as the major isolates from cases of mastitis in dairy cattle. Abebe *et al.*, (2016) reported that *S. aureus* was isolated from 51.2% of milk samples cultured and 73.2% of the herd were affected with mastitis. The high prevalence of *S. aureus* in this study might be associated with the absence of hygienic milking practices, a lack of culling of cows chronically infected with *S. aureus*, and consistent hand-milking practices throughout the dairy herds. Since *S. aureus* is usually found on the udder or teat skin surface of infected animals, the primary source of transmission from infected udders to uninfected is usually by the milkers' hands during hand-milking.

*S. aureus* isolates in this study showed high sensitivity to gentamycin, and less resistant to Nocardin (14.3%). This might be due to limited usage of these antimicrobials for the treatment of diseases of these species of dairy animals, including mastitis. *S. aureus* isolates from cows were resistant to Penicillin (71.4%), Amoxicillin (100%), and Doxycycline (28.6%). These results were in agreement with reports from Tassew et al., (2017) in and around Assosa that suggested a possible development of resistance from prolonged and indiscriminate use of these antimicrobial drugs. Relatively similar to the report of Getahun et al., 2008 the present findings of all *S. intermedius* isolates from cows showed that 100% resistance to penicillin and amoxicillin, and 50% susceptible to Doxycycline and Nocardin relative to 100% susceptible to gentamycin.

## Conclusion and recommendations

As a conclusion, the findings of current study signifies as utilization of drugs and treating any sick livestock are out of the established health protection procedures. Overall there is no any strong drug utilization and professional codes sated to encourage the exercise of legal drug and treating responsibilities. As a result, majority of pastoralists buy drugs for their sick animals and treat by themselves, use any of antibiotic to treat any kind of diseases. Generally, inappropriate use of antimicrobials like sharing antibiotics between humans and animals and the awareness related to the antimicrobial residues in animal food items (milk, meat and eggs) have been take placed in the way that encourage the emergence of AMR. Inclusively, drugs Handling and management plus treating sick live stocks either by health professional or pastoralists have been found very poor. Therefore the following points need to be addressed if the improvement of the situation is required. Awareness creation on proper drug transportation and storage, practice of selling drugs with prescription, proper disposal of expired drugs, dose determination, follow-up of treated animals and determining drug withdrawal periods starting from acquisition to end users should to be made .A strong and objectively designed manual that comprehensively considered drug handling and utilization management that involved all stakeholders and addresses all the major issues starting from acquisition to end users must be prepared and made available.

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# **Meat Animals Research Results**

# Study on Sero-prevalence and Associated Risk Factors of Camel Brucellosis in Selected Districts of Borana, Oromia Regional State, Ethiopia

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## **Abstract**

*This study was conducted with the objectives of assessing the epidemiological distribution of the disease, identifying the risk factors involved and to know the perception of the community about the disease. Sera samples were collected from a total of 621 camels from four randomly selected districts. The samples were screened using Rose Bengal Plate Test (RBPT) and confirmed by competitive Enzyme-Linked Immunosorbent Assay (c-ELISA). Out of the total 621 sera samples tested 44 (7.09%; 95% CI= 5.20-9.40%) and 15 (2.42%; 95% CI= 1.36-3.95%) were positive for antibodies by RBPT and c-ELISA, respectively. The prevalence of camel brucellosis was 3.82% in Gomole (95% CI= 1.42-8.13%), 3.27% in Yabello (95% CI= 1.07-7.50%), 1.32% in Arero (95% CI= 0.16-4.67%) and 1.26% in Elwoya (95% CI= 0.15-4.47%) districts in decreasing order; with insignificant difference between the districts ( $P > 0.05$ ). Age, history of reproductive health problem and parity of the animal were associated with prevalence of the disease ( $P < 0.05$ ) while sex and herd size of the animals were not associated ( $P > 0.05$ ) with the occurrence of brucellosis. In general, this study indicates that camel brucellosis was prevalent in all four districts of the study area and hence awareness creation for pastoralists in the study area and designing appropriate control methods has to be undertaken to improve the health and productivity of camel as well as prevention of community from such zoonotic disease.*

**Keywords:** Camel, Brucellosis, Risk factors, Prevalence, Borana, Ethiopia

## **Introduction**

Worldwide there are roughly 30 million dromedary camels, with highest number in Africa and the Middle East (Zhu et al., 2019). Interest in dromedary camel (one-humped camel) rearing has increasingly developed in arid countries. Camels play a vital role in food security and national economy (Mohamud et al., 2021). Ethiopia is one of the largest camel's populated countries in the world with 1,102,119 numbers of camels that rank third in Africa next to Somalia and Sudan. The camel is kept in the arid and semi-arid lowlands of Borana, Ogaden and Afar, which accommodate 50% of the pastoralists areas in Ethiopia (Jara et al., 2020).

Brucellosis is a globally important zoonotic disease that affects cattle, sheep, goats, camels, pigs and wildlife as well as humans (Khan et al., 2020) and caused by some bacteria in the genus *Brucella* (Wainaina et al., 2020). Brucellosis is an infectious disease of livestock and wild animals and is the second most important zoonotic disease after rabies throughout the world (Gondal et al., 2017). The disease was first reported in camels in 1931 and since then has been testified by all camel rearing countries like Sudan, Ethiopia, Somalia, Kenya, Nigeria, Jordan and Egypt but not Australia (Khan et al., 2020).

Brucella infection in camels is mainly caused by *B. abortus* and *B. melitensis*. Camels are not known to be the primary host for Brucella spp., and infection in camels depends on contact with other primary host animals (Mohamud et al., 2021). The clinical picture of brucellosis in camels vary from asymptomatic to abortion, retention of fetal membranes, weak offspring, impaired fertility and delayed sexual maturity in females and orchitis accompanied by lameness in males (Khan et al., 2020).

The disease is well controlled in many countries but is still endemic in many others with high records in humans in the Middle East and central Asia regions (Khan et al., 2020). Humans get exposed to Brucella spp. through consumption of contaminated raw or poorly cooked animal source foods (ASF), contact with infected tissues such as aborted fetuses and at the time of taking care of sick animals (Wainaina et al., 2020). The disease is highly important especially in pastoral areas where there is close contact between livestock and people, because Borana is one of the pastoral area where people are totally dependent on livestock amongst which camel is one, it is important to generate information about camel brucellosis in the area. Therefore the study was conducted with the objectives: to determine the sero-prevalence of camel brucellosis, to identify potential risk factors associated with camel brucellosis in the study and to know the level of pastoralists’ awareness about camel brucellosis.

## Materials and Methods

### *Description of the study area and period*

The study was conducted in four districts of Borana zone namely Arero, Elweya, Gomole, and Yabello (Figure 1). The mean annual rainfall of the area ranges from 250 to 700 mm. The annual mean temperature varies from 19 to over 25 °C. The area has a bimodal rainfall distribution; i.e. the long rainy season called “Ganna” and the short rainy season called “Hagaya”(Teshome *et al.*, 2019; Teshome *et al.*, 2022).

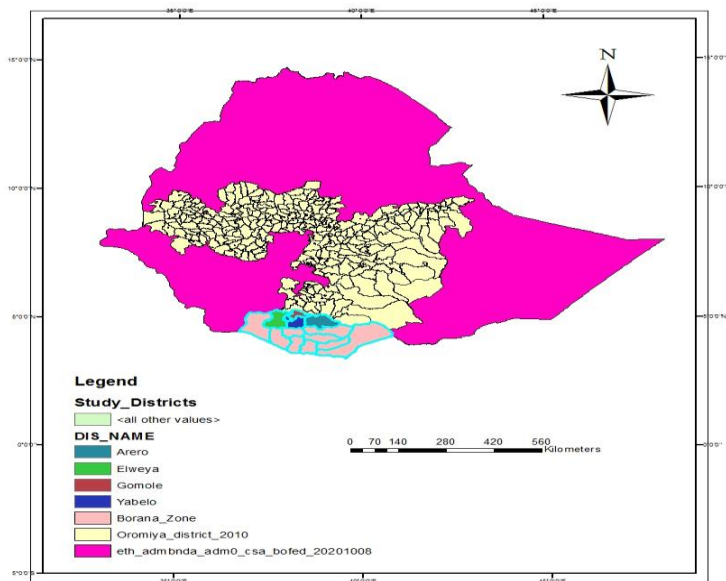


Figure 1: Study site

### ***Study Animals***

The study animals were camels which were kept under pastoral and Agro pastoral communities of Borana Zone. All camels with age of 6 months or above were included in the study.

A cross-sectional study design was used for both questionnaire survey and serological tests (RBPT and cELISA).

### ***Sampling method and sample size determination***

Multistage random sampling coupled with systematic sampling method was applied to select the study animals. Primary sampling units were a list of 13 districts found in the zone followed by random selection of four districts from them (Arero, Elwoya, Gomole, and Yabello). Then two villages per each district was randomly selected. Following this, camels of varying age were systematically selected from each village for serological sampling. The determination of sample size for serum collection was based on the formula given by Thrusfield (2005) as follows:

$$n = \frac{1.96^2 P_{exp} (1 - P_{exp})}{d^2}$$

Where, n = required sample size,  $P_{exp}$  = expected prevalence, d = desired absolute precision (5%)

For sample size determination prevalence of 50% considered and thus, 50% was used as the expected prevalence. Using the 95% confidence level and an expected prevalence of 50% and 5% absolute precision a sample size of 384 camel were obtained. However, for this study 621 camels were used.

### ***Questionnaire Survey***

In this study, a semi structured questionnaires were prepared and administered to randomly selected livestock owners in the village and animal health practitioners. The questionnaires were pre-tested on few of the respondents before actual data collection, evaluated and then data were collected. Data on breed, sex, age, herd size, abortion, and presence of swollen joints, animal management, and importance of the disease for human health were collected. Moreover, the history of reproductive problem in the study population was also collected.

### ***Blood sample collection and serological analysis***

A volume of 10ml of blood sample was collected from the jugular vein of each animal using plain vacutainer tube and each sample was labelled by using codes describing the specific animal. After that the collected samples were allowed to clot overnight in a slant position at room temperature. Then sera removed from the clot in to Cryovials with plastic pipettes; unretracted bloods were centrifuged, by siphoning into another sterile test tube and finally the serum samples were stored at  $-20^{\circ}\text{C}$  until tested by RBT.

### **Screening test and laboratory confirmation of samples**

All serum samples were screened using the RBPT, according to the procedures described by Alton *et al.* (1975) and the *Manual of Diagnostic Tests and Vaccines for Terrestrial Animals* of the World Organisation for Animal Health (OIE, 2018). The rose Bengal antigen constituted a suspension of *B. abortus*. The RBPT was carried out at Yabello veterinary laboratory. Thirty  $\mu$ l of serum was mixed with an equal volume of antigen suspension on a glass plate and agitated. After four minutes of rocking, any visible agglutination was considered a positive result followed by confirmation of positive samples by cELISA at National Animal Health and Diagnostic and Investigation Center (NAHDIC), Sebata, Ethiopia.

### **Data Management and Statistical Analysis**

The collected data was checked, coded and entered in to Microsoft excel and saved until analysis. The data was analyzed by stata statistical software (version 13.0). Brucella prevalence was determined by dividing the number of positive samples by the total number of animals examined and expressed as Percentages (%). Fishers' exact test was used to measure association between prevalence of the disease and risk factors. Confidence level of 95% and statistical significance at P-value less than 0.05 were considered during analysis.

### **Results**

The result of this study showed that out of 621 camels screened by RBPT, 44 (7.09%) were positive for Brucella infection. Out of 44 positive animals, 15(2.42%) were confirmed positive by using cELISA. With respect to study districts, the study showed a prevalence of 3.82% in Gomole (6/157; 95% CI= 1.42-8.13%), 3.27% in Yabello (5/153; 95% CI= 1.07-7.50%) and, 1.32% in Arero (2/152; 95% CI= 0.16-4.67%), and 1.26% in Elwoya (2/159; 95% CI= 0.15-4.47%). There was no statistically significant difference ( $P>0.05$ ) among the districts. The result also showed that, out of 504 female camels and 117 male camels, 14(2.78%) and 1(0.85%) were positive by cELISA, respectively. The study revealed that prevalence of the disease was associated with age, history of reproductive problem, herd size and parity of the camel ( $P<0.05$ ), but not with sex of the camels (Table 1).

Table 1: Sero-prevalence and Associated Risk Factors Camel brucellosis

Risk Factors	Number of tested	Number of positive	Percent	(95%Conf. Interval)	P-Value
<b>District</b>					
Arero	152	2	1.32	(0.16 – 4.67%)	0.368
Elwoya	159	2	1.26	(0.15 – 4.47%)	
Gomole	157	6	3.82	(1.42 – 8.13%)	
Yabello	153	5	3.27	(1.07 – 4.50%)	
<b>Sex</b>					
Female	504	14	2.78	(1.53 – 4.62%)	0.325
Male	117	1	0.86	(0.02 – 4.67%)	
<b>Age</b>					
Adult ( $\geq 4$ )	318	12	3.77	(1.96 – 6.50%)	0.034
Young ( $\leq 4$ )	303	3	0.99	(0.21 – 2.87%)	



<b>Herd size</b>					
Large	125	7	5.60	(2.28 – 11.20%)	0.021
Medium	251	6	2.39	(0.88 – 5.13%)	
Small	245	2	0.82	(0.099 – 2.92%)	
<b>History of reproductive problems</b>					
Yes	220	11	5.00	(2.52 – 8.77%)	0.004
No	401	4	0.997	(0.27 – 2.53%)	
<b>Parity</b>					
0	167	2	1.20	(0.15 – 4.26%)	0.021
≤2	224	4	1.79	(0.49 – 4.51%)	
≥3	113	8	7.08	(3.11 – 13.47%)	
<b>Total</b>	<b>621</b>	<b>15</b>	<b>2.42</b>	<b>(1.36-3.95%)</b>	

Result of multivariable for fitting regression model for risk factors statistically significant for Fisher's exact test indicated that the odds of brucellosis sero-prevalence in adult camel was twice (OR= 2.177) that of young camels. The likelihood of getting infection was 3.65 times higher in animals with history of reproductive health problems to the odds of camels without previous history of reproductive health problem. The current study also showed that animals reared in large and medium herd size had about 7.05 and 3.28 times higher odds of being positive to brucella, respectively than those animals from small herd size, respectively (Table 2).

Table 2: Result of multivariable logistic regression for significant factors

<b>Risk Factors</b>		<b>Odds Ratio</b>	<b>Std. Err.</b>	<b>z</b>	<b>P&gt;z</b>	<b>(95% Conf. Interval)</b>
<b>Age</b>	Young	Ref.				
	Adult	2.177	1.570	1.080	0.281	(0.530 - 8.946)
<b>HRHP</b>	No	Ref.				
	Yes	3.645	2.381	1.980	0.048	(1.013 – 13.113)
<b>Herd size</b>	Small	Ref.				
	Large	7.050	5.747	2.400	0.017	(1.426 – 34.843)
	Medium	3.284	2.714	1.440	0.150	(0.650 - 16.592)
<b>Constant</b>		0.002	0.002	-6.530	0.000	(0.000 – 0.014)

HRHP=History of reproductive health problem

From the result of questionnaire survey the respondents revealed that about 26.67%, 80.83%, 95.83%, 100% were only have awareness about zoonosis, disposed aborted fetus and fetal membrane with bare hand, consume raw milk and assist delivery during parturition with bare hand respectively (Figure 2).

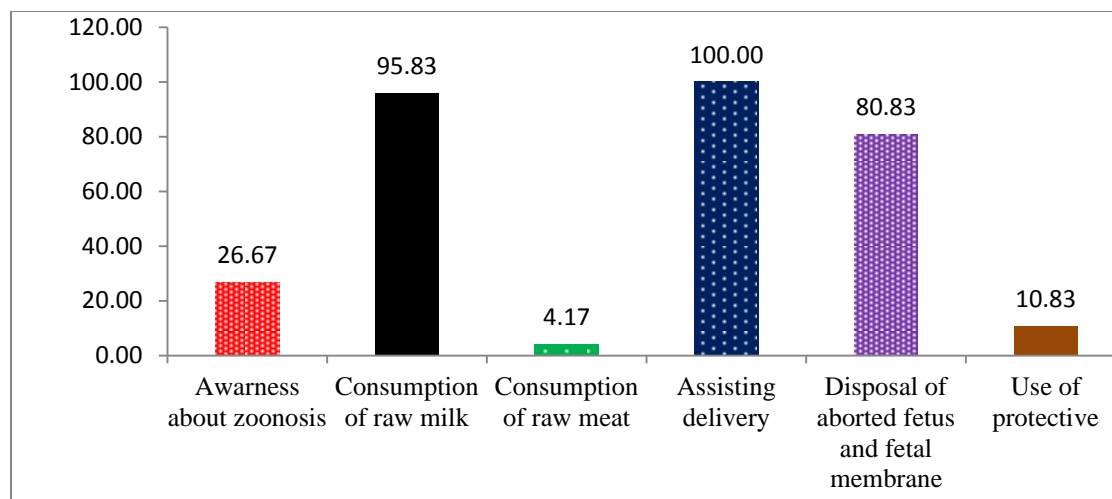


Figure 2. Perception of Pastoralists towards the disease and their practices

## Discussion

In the current study, the overall sero-prevalence of camel brucellosis in selected districts of Borana was 7.09% by the RBPT and 2.42% by cELISA. The current result agrees with the report (2.43%) of Tilahun *et al.* (2013) in Jigjiga and Babile districts, Eastern Ethiopia and report (2.09%) of Zeru *et al.* (2016) in Afar. The present study result is higher than previous finding by Mohammed *et al.* (2011) in Dire Dawa (1.6%) and Megersa *et al.* (2005) in Borana (1.8%). However, it is lower than the result (4.2%) of Teshome *et al.* (2003) in Borana, Hadush *et al.* (2013) in Afar (4.1%), Zewolda and Wereta (2012) in afar (5.7%). The difference observed among these studies could be due to the variation in animal management, topography, animal movement during drought in search of feed and water. There is no statistical difference between sexes. This is might be due similar animal management and movement.

Age showed statistically significant difference. This finding was in line with study conducted by (Megersa *et al.*, 2005; Tilahun *et al.*, 2013 and Alrawahi *et al.*, 2019). The odds of positivity to brucella antibody in adult camel are 3.92 times higher than the young ones. Sexually matured animals are more prone to *Brucella* infection than sexually immature due to sexual mature animals participate on mating and sugar erythritol development increase, which favors the multiplication of pathogen. As age increase the probability of getting expose to infection increase and young animals tend to be more resistant to *Brucella* infection and frequently eliminate the infection (Poester *et al.*, 2013 and Alrawahi *et al.*, 2019).

In the present study, camels with larger herd size were more likely to be infected by brucella as compared to small herd size. Similar observations are reported by (Mohammed *et al.*, 2011; Hadush *et al.*, 2013; Alrawahi *et al.*, 2019).

History of reproductive Problem shows significant difference to sero positivity to brucellosis and the chance of getting infected for animals having history of reproductive problem was 5.22 more likely to test seropositive than animals with no history of reproductive problem (Radiostits *et al.*, 2010). As camels with history of reproductive problem (HRP) increases the chance of contact between animals increases leading to more chances of infection particularly during calving and abortion.

Parity also shows significant association and the she-camels with parity greater than three were 4.24 times the more chance in getting positive to brucellosis and it is in line with Jara *et al.* (2020). However due to parity and age was multi-collinearity it was not included in multivariable regression model.

## Conclusion and Recommendations

Based on questionnaire survey and serological test; the present study indicated that camel brucellosis is prevalent in the study area. Age, history of reproductive health problem and parity were found to be risk factors of camel brucellosis in the area. Shortage of awareness on zoonotic nature of the disease and lack of knowledge on prevention methods to decrease disease transmission from animal to human and from animal to animal have were the common problems raised the participants. Based on the findings; the following recommendations are forwarded:-

- An integrated control and prevention approach on camel brucellosis is warranted
- Strengthening of the community awareness about the disease, source, transmission, prevention and its zoonotic nature
- Age of the animal should be considered during control and prevention

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# Supplementation of alfalfa hay as a substitute for Noug seed cake in concentrate based feeding of yearling Arsi-Bale Sheep

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## **Abstract**

*The study was conducted at Adami Tulu Agriculture Research Center to evaluate supplementation of alfalfa hay as a substitute for noug seed cake on the growth performance and carcass parameters of yearling Arsi-Bale sheep. The experimental feeds were formulated in such a way that they are iso-nitrogenous. Ingridients of the formulated experimental feeds were wheat barn, noug seed cake and alfalfa hay. The alfalfa hay was set in treatments with proportion of 0%, 25%, 50%, 75% and 100% of the Noug cake in the concentrate diet. Thirty yearling Arsi-Bale sheep were purchased and treated for anti-parasites. The sheep were allocated to the treatments in a randomized complete block design with five treatments replicated six times. The feeding trial lasted for hundred days followed by slaughter of representative animals for evaluation of carcass traits. The final body weight, total body weight gain, average daily weight gain, hot carcass weight, dressing percentage and most of the non-carcass traits of the sheep were not significantly varied among the treatments. The overall final body weight, total body weight gain and average daily weight gain of the experimental sheep were 29.23 kg, 10.76 kg and 107.54g/day, respectively. The overall of hot carcass weight and dressing percentage of the experimental animals were 10.78 kg and 37.34%. All treatments had positive effects on the sheep growth performance and carcass traits. The partial budget analysis indicated that animals fed high proportion of alfalfa hay fetched more income than those fed low proportion. Therefore, alfalfa hay as a protein supplement for yearling Arsi-Bale sheep can be demonstrated for wider use in areas where growing alfalfa is possible.*

**Keywords:** *Alfalfa hay, Noug seed cake, Arsi-Bale sheep*

## **Introduction**

The local sheep population is above 42 million per head (CSA, 2021) and they play significant roles in the Ethiopian economy. However, the productivity of local sheep in terms of live weight gain and carcass yield is very low, mainly due to inadequate nutrition (Gezahegn *et al.*, 2020) associated with reliance on sole natural pasture, crop residues and/or stubble grazing. The dry forage from grazing areas and crop residues have below 7% CP which indicate poor nutritive value not cable of meeting microbial requirements (Van Soest, 1994). The oil crop by-products have high crude protein in dry mass. But the prices of protein source feeds are becoming increasing in the country. Feed cost is the main factor that affects animals' farm economic return. Therefore, looking for other protein sources which are relatively cheaper and which could be substitutes for oil crop byproducts is inevitable.

The dietary nutrients like energy and protein are the major factors affecting productivity of sheep (Worknesh and Getachew, 2017). There are several complementary and alternative strategies that can be pursued in tropical regions with the objective of making low quality feeds more useful for production of meat and milk. According to Preston and Leng (1987), concentrate feeding is one strategy which increases intake and digestibility. However price of concentrate is increasing in alarming rate. The

utilization of local protein sources such as leguminous forages is one option to maximize the productivity of animals (Popi and McLennan, 1995). It plays an important role in supplementing diets of growing lambs as an alternative to the oil seed cake supplements (Worknesh and Getachew, 2017).

Legume forages can substitute the protein sources, particularly noug seed cake, in the diet of animals (Ajebu *et al.*, 2013). Among the forage legumes, Alfalfa is a perennial forage legume known for its high forage quality and positive effects on soil fertility. Alfalfa withstands long periods of water deficit by impeding its vegetative growth (Annicchiarico *et al.*, 2010). It accesses water from depth through its deeper root system (Voltaire, 2008).

Alfalfa is a common feed stuff included in the diets of many livestock species (Sen *et al.*, 1998). Alfalfa leaves have a high protein content whereas its stems are high in fiber (Palmonari *et al.*, 2014). However the CP content of alfalfa varies depending on the maturity stage. Alfalfa forage has an important role in ruminant production. Farmer can produce easily by rain or irrigation on his farm. It is characterized as a low cost protein sources and can be used as a self-sufficient substitute for expensive concentrate (Wang *et al.*, 2019). Therefore, the study was designed with the objectives of evaluating the feeding of different levels of alfalfa hay on growth performances and carcass parameters of Arsi-Bale sheep and the profitability of its substitution for noug seed cake..

## **Materials and Methods**

### ***Study area***

The experiment was conducted at Adami Tulu Agricultural Research Center, which is located in mid rift valley 167 South of Addis Ababa at an altitude of 1650 meter above sea level. The agro-ecological zone of the area is semi-arid and sub humid with acacia woodland vegetation type. The mean annual rain falloff the area is 760 mm. The minimum and maximum temperatures are 12.6°C and 27°C, respectively. The soil type is fine, sandy loam with sand: silt: clay ratio of 34: 38: 18, respectively.

### ***Feed preparation***

Seeds of adapted variety of alfalfa (Hunter River) were collected from Werer Agriculture Research Center. The seeds were sown at a rate of 10 kg/ha on well prepared land. All the necessary agronomic practices were conducted and plant was harvested at 50% flowering and dried under shade for hay preparation. The hay was manually chopped to a size of 2-3 cm by a grinding baler machine to encourage intake. Wheat bran and noug seed cake were purchased from markets.

### ***Animals and experimental design***

A total of 30 yearling Arsi-Bale sheep were purchased from the local markets nearby the center. Ages of the sheep were estimated based on dentition techniques and information obtained from the owners of the sheep (seller). All sheep were ear-tagged and treated for internal and external parasites. The pens were disinfected before the animals were moved in and then cleaned daily. Sheep were kept in individual pens during the experimental period. The experiment was conducted as a randomized complete block design with five treatments replicated six times. The experimental feeds were formulated based on the iso-nitrogenous principle. The control group (treatment 1) was fed 50% wheat bran, 49% *noug* seed cake and 1% salt. The alfalfa hay was made to replace the 25%, 50%, 75% and 100% of the target feed (*noug* cake)

in treatment 2,3, 4 and 5, respectively. . Rhodes hay was given *ad-lib* for all experimental treatments. . Water was given freely for the animals.

### ***Animals feeding and live weight gain***

Animals were fed on the experimental feed for 14 days adaptation period to acclimatize them to the feed as well as to the pens and experimental procedures. The formulated feeds were supplemented for experimental sheep at the rate of **2.5%** of their body weight during the whole growth periods. The animals were weighted at 14 days intervals. The amount of feeds supplemented to the animals were adjusted based on their body weight gain at 14 day interval. All animals were weighed in the morning hours after overnight fasting using a weighing scale. The actual feeding trail was conducted for hundred days. The total weight gain of the experimental sheep was calculated by subtracting initial weight from final body weight and the daily body weight gain was calculated as the difference between final live weight and initial live weight divided by the number of feeding days.

### ***Carcass parameters***

After the completion of the feeding trial, three experimental sheep were selected randomly from each feeding group. The selected sheep were fasted overnight and slaughtered for evaluation of carcass parameters at Adami Tulu Agriculture Research Center slaughter house. Slaughter body weights were taken before slaughtering the animals. After the animals were slaughtered, the weights of kidney, lung with trachea, heart, liver, spleen, kidney fat, heart fat, full gut, empty gut, penis, testicle, skin with feet, tail and head with tongue were taken using digital weighing balance. The hot carcasses were split along the vertebral column into the left and right sides and then weighed. The right sides were chilled for 24 hours at 4°C before being weighed. The chilled carcass was deboned into lean, fat, and bone components. Chilling loss was calculated as the percentage of the difference between hot carcass weight and chilled carcass weight divided by hot carcass weight multiplied by 100. Slaughtering dressing percentage = hot carcass weight divided by slaughtered body weight multiplied by 100.

### ***Partial budget analysis***

The variable costs such as feed cost, animal purchase, veterinary cost, animals transportation cost, labor cost and transportation different items were included in the partial budget analysis. The land rent price, ploughing cost, weeding cost, harvesting cost, and seed price were included in the alfalfa production costs. At the end of the trial, sheep selling prices were estimated by known skilled traders. The gross return was calculated by subtracting the total cost from the revenue.

### ***Method of data analysis***

The data were subjected to analysis of variance in a randomized complete block design using the linear model procedure of SAS (2009). The treatment means were separated by LSD. test. The model used for data analysis is:  $Y_{ij} = \mu + t_i + e_{ij}$ , Where:  $Y_{ij}$  = response variable;  $\mu$  = over all mean;  $t_i$  = treatment effect;  $e_{ij}$  = random error.

## Result and discussion

### *Live weight gain*

The effects of feeding different levels of alfalfa hay as a replacement to noug seed cake on the growth performances of Arsi-Bale sheep are given in Table 1. The average initial weights of the sheep were 18.33, 18.66, 18.33, 18.33 and 18.66 kg for animals in T1, T2, T3, T4 and T5, respectively.

Table 1. Effect of different levels of alfalfa hay on growth performance of sheep

Treatments	Growth parameter			
	IBW(kg)	FBW(kg)	TWG(kg)	ADG(g/day)
T1	18.33±1.05	28.50±1.26	10.16±0.60	100.00±5.72
T2	18.66±0.84	29.16±1.01	10.50±1.05	105.00±10.06
T3	18.33±0.49	29.00±0.68	10.66±1.02	106.7±9.73
T4	18.33±0.61	29.66±0.61	11.33±0.66	113.33±6.35
T5	18.66±0.42	29.83±0.70	11.16±0.83	111.66±7.94
Overall mean	18.46±0.30	29.23±0.38	10.76±0.36	107.54±3.47

IBW: initial body weight, FBW:Final body weight, TWG: Total body weight, ADG: Average daily weight gain

The observed values of final body weight, total weight gain and daily weight gain did not show significant differences among the treatments. This might be due to the fact that the feed formulation was on the basis of crude protein content of the ingredients in such a way that dietary rations were iso-nitrogenous. The overall final body weight gain observed in this trial was higher than the value of 25.3 kg reported by Aman *et al.* (2019) for yearling Arsi-Bale rams fed on wheat bran 50% and 49% noug seed cake for 75 days with a value of . at Adami Tulu Agriculture Research Center. The overall final body weight of the experimental sheep in this study was lower than what was observed for yearling Afar Sheep that were fed on different wheat bran and leucaena leaves mixture and reached 30 kg in 98 days at Werer Agriculture Research Center (Abebe *et al.*, 2013). The final body weight of the animals in the current study was similar with that of the Arsi-Bale sheep fed 400 g concentrate and Desho grass at Debra Zeit Agricultural Research Center (Worknesh *et al.*, 2021). However, the daily weight gain of the animals in the current finding was lower than the value of 111.9 g/day reported by Tedesse *et al.* (2014) for Arsi-Bale sheep fed on concentrate blocks at Southern part of the country. Similarly, the observed average daily gain in the current study was relatively similar to the finding of Abebe *et al.* (2010) in which 104g/day was reported for Arsi-Bale lamb fed on linseed cake and wheat bran diet. From the points mentioned above, it can be said that supplementation of alfalfa hay in this study had positive effect on the weight gains of the yearling Arsi-Bale sheep.

### *Carcass characteristics*

The effects of supplementation of different levels of alfalfa hay on the carcass parameters of the experimental sheep are indicated in Table 2. The slaughter weight, hot carcass weight, cold carcass weight, chilling loss and dressing percentage of the lambs did not vary ( $P<0.05$ ).among the treatments The dressing percentages, based on the slaughtering body weight, varied from 36.05 to 38.87%with the highest value observed for treatment 2 and the lowest for treatment 5..



Table 2. Carcass characteristics of sheep fed on different level of alfalfa hay

Parameter	Treatments					Overall
	T1	T2	T3	T4	T5	
SBW(kg)	29.16±1.05	28.16±0.88	29±1.00	29.5±1.45	29.33±0.57	29.30±0.53
HCW(kg)	10.93±0.63	10.96±0.82	10.90±0.37	10.85±0.42	10.23±0.34	10.78±0.22
Right side HCW	5.45±0.25	5.48±0.39	5.47±0.16	5.42±0.24	5.15±0.15	5.39±0.10
Left side HCW	5.48±0.38	5.48±0.43	5.47±0.23	5.43±0.19	5.08±0.19	5.39±0.12
Right side CCW	5.36±0.43	5.23±0.39	5.25±0.13	5.27±0.19	5.12±0.20	5.25±0.11
Chilling loss%	1.51	4.56	3.83	2.76	1.55	2.85
DP%	37.47	38.87	36.56	36.76	36.05	37.34

SW: slaughtering weight, EBW: empty body weight, HCW: Hot carcass weight, DSW: dressing percentage of slaughtering weight, DEBW: dressing percentage of empty body weight

The carcass yield observed in the study was in the range of 10.23-10.96 kg. The average carcass yield of the experimental animals was higher the finding of Abdi *et al.*, (2019) who reported the average carcass weight of 8-9 kg per sheep for indigenous sheep at yearling age. The carcass yield observed in the current experiment was similar to the 10-11 kg reported by Abebe *et al.* (2010) for Arsi-Bale lamb fed linseed cake and wheat bran. Carcass yield is affected by the nutrition quality, genotype, season, age and sex of the animals. The chilling loss percentage observed in this study was similar to the finding of Getahun *et al.* (2020) for Arsi-bale sheep fed on the sugar tops silage. The chilling loss had minor effect in lowering the dressing percentage (Getahun *et al.*, 2020). The observed values for dressing percentage in the current study was found to be in the range of 36-38% reported by Ayele and Urge (2019) for Arsi-Bale sheep fed on urea treated barley straw supplemented with 300-400 g concentrate. Similarly, Girma *et al.*, (2010) reported dressing percentages of 36.2 -38.5% for yearling Arsi-Bale lambs raised on pasture.

### ***Carcass compositions***

The effects of different alfalfa hay on carcass composition are presented in Table 3. The muscle, fat and bone weights did not significantly varied among the experimental groups. The absence of variations among the experimental treatments in carcass composition might be correlated to the similarity in nutritional composition of the feeds. The observed values showed that the proportion of muscle was higher followed by the bone and fat in the carcass. The values of muscles, fat and bone observed in this experimental were closer to the report by Getahun (2015) for Arsi-Bale sheep fed different dietary crude protein levels.

**Table 3:** Composition of muscles, bone and fat of the different treatments

Parameter	Treatments					Overall
	T1	T2	T3	T4	T5	
Muscles wt (kg)	3.36±0.24	3.23±0.44	3.33±0.07	3.18±0.10	3.00±0.5	3.22±0.07
Fat wt(kg)	0.65±0.15	0.76±0.18	0.65±0.13	0.78±0.11	0.68±0.08	0.71±0.05
Bone wt(kg)	1.28±0.08	1.23±0.04	1.18±0.08	1.33±0.14	1.40±0.03	1.28±0.04
Muscles%	63.65	61.74	64.68	60.24	59.05	<b>61.87</b>
Fat%	12.03	14.34	12.41	14.71	13.38	13.37
Bone%	24.32	23.92	22.91	25.05	27.57	24.75
Muscle : Bone ratio	2.64	2.64	2.84	2.43	2.14	2.54
Muscle : Fat ratio	5.62	4.65	5.62	4.18	4.49	4.91

**Non-carcass parameters**

The effects of different levels of alfalfa hay supplementation on non carcass traits of the yearling Arsi-Bale sheep are presented in Table 4. The liver with bile of sheep in treatment group which received 50% alfalfa hay was lower than the liver with bile values of sheep in other treatments. Kidney fat of the sheep in treatment 2 (25% alfalfa hay) was higher than the kidney fat values of sheep in other treatments. However, most of non carcass traits were did not show significant differences among the treatments. The report by Teklu (2016) also stated that the weights of offal components were not significantly affected by different feeding regimes.

**Table 4.** The effects of alfalfa hay supplementation on non-carcass components of the experimental sheep

Non carcass traits (kg)	Treatments					SME	SL
	T1	T2	T3	T4	T5		
Tongue	0.13	0.13	0.13	0.28	0.20	0.04	Ns
Heart	0.13	0.12	0.10	0.18	0.10	0.14	Ns
Kidney	0.13	0.13	0.1	0.08	0.1	0.01	Ns
Liver with Bile	0.47 <sup>a</sup>	0.48 <sup>a</sup>	0.40 <sup>b</sup>	0.52 <sup>a</sup>	0.45 <sup>a</sup>	0.02	**
Lung &Trachea	0.43	0.40	0.33	0.32	0.33	0.03	Ns
Kidney fat	0.07 <sup>b</sup>	0.13 <sup>a</sup>	0.07 <sup>b</sup>	0.08 <sup>b</sup>	0.07 <sup>b</sup>	0.01	**
Scrotal fat	0.08	0.13	0.12	0.13	0.10	0.01	Ns
Omental fat	0.25	0.23	0.10	0.18	0.17	0.03	Ns
Small Intestine	1.55	1.58	1.70	1.70	1.76	0.05	Ns
Large intestine	1.43 <sup>a</sup>	1.01 <sup>ab</sup>	1.35 <sup>a</sup>	0.92 <sup>ab</sup>	0.78 <sup>b</sup>	0.09	**
Tail	0.93	0.63	0.71	0.93	0.75	0.05	Ns
Head with skin	1.93	2.02	1.36	1.75	1.72	0.12	Ns
Skin	2.85	2.30	2.78	4.78	2.32	0.53	Ns
Feet with hooves	0.62	0.60	0.58	1.43	0.62	0.16	Ns

**Partial budget analysis**

The partial budget analysis of sheep fed different levels of alfalfa hay as a replacement for noug seed cake is presented in Table 5. The costs of feed ingredients used in the feeding trial were 14 and 25 ETB/kg for

wheat bran and noug seed cake, respectively. The production cost of alfalfa hay was 7.5 ETB/kg in dry mass. The gross return per yearling Arsi-Bale sheep fed noug seed cake and wheat bran was negative. However, the sheep fed higher proportion of alfalfa hay resulted in more profitability as compared to those fed lower proportion of alfalfa hay with values of Birr 188.20, 299.40 and 451.80 for treatment 3, 4 and 5, respectively. The profit variation among the experimental groups was mainly due to the expensive price of noug seed cake in the diets. Esubalew *et al.* (2020) reported that replacement of noug seed cake with cow pea at the proportion of 75 and 100% replacements appeared to be a better option for inclusion in sheep fattening ration.

Table 5. Partial budget analysis of sheep fed different alfalfa level as replace to noug seed cake

Items (Birr)	Treatment				
	T1	T2	T3	T4	T5
Purchasing price per sheep	1500	1500	1500	1500	1500
Total feeds cost per sheep	1353.2	1217.1	1051.8	940.6	788.2
Cost of transport per sheep	20	20	20	20	20
Labor cost per sheep	250	250	250	250	250
Veterinary cost per sheep	90	90	90	90	90
Total cost per sheep	3183.2	3077.12	2911.8	2800.6	2648.2
Final sell per sheep	3100	3100	3100	3100	3100
Gross return per sheep	-113.2	22.88	188.2	299.4	451.8
Gross margin per sheep	-679.1	137.29	1129.1	1796.6	2710.8

## Conclusion and Recommendation

The substitution of noug seed cake by alfalfa hay at 0, 25, 50, 75, and 100% had a similar positive effect on growth performance and carcass parameters of yearling Arsi-Bale sheep. The results suggest that, in feeding the yearling Arsi-Bale sheep, alfalfa hay could replace noug seed cake fully without impairing the growth and carcass traits of the sheep. The substitution of noug seed cake by alfalfa hay at 100% was more profitable than at 75% followed by 50%. Supplementation. Therefore, this alfalfa hay feeding as a substitute for protein source in supplementary feed of yearling Arsi-Bale sheep should be demonstration at on-farm level.

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# The feeding value of *Lablab purpurues* for Horro ewes managed under free-grazing condition

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## Abstract

*The study was conducted at Bako Agricultural Research Center to determine the feeding value of Lablab purpurues forage legume in terms of productivity and reproduction traits in Horro ewes. The dietary treatments comprised a mixture of concentrate (T1), as well as two varieties of Lablab purpurues namely, Gebis-17 (T2) and Beresa-55 (T3). Twenty-one Horro ewes of similar parity with a mean pre-mating weight of 26.3 + 2.1 (Mean + SD) were selected from the center's sheep breeding flock and placed into three groups of seven ewes each depending on pre-mating weight. The ewes were subsequently fed corresponding feed regimens over an 8-month period that included 5 months of gestation and 3 months of lamb growth until weaning. The results showed that supplementation enhanced lamb birth weight, ewe weight gain during pregnancy, and all reproductive features studied, except for abortion instances that occurred in ewes fed a diet in T2 and T3. The causes of abortion in T2 and T3 cannot be ascertained because the experimental ewes were grazing during the day. Despite the fact that the T1 diet improved ewe performance in a similar way to T3 and T2, the use of such supplements is usually limited due to their high cost and inaccessibility to smallholder farmers. This emphasizes the importance of seizing the chance to replace conventional protein supplements with low-cost, on-farm cultivated forage legumes like the one studied in this study. As a result, enhancing Horro ewes' performance with either T2 or T3 diets throughout their mating season is a promising strategy for maximizing their genetic potential. Future research should concentrate on the influence of forage legume supplements on post-weaning growth rate and survival in lambs.*

**Keywords:** Horro ewes, productive performance, reproductive performance, supplementation

## Introduction

Ethiopia has a diversified indigenous sheep population, with meat production being one of the most important aspects of the country's economy (Solomon et al., 2013). Their production is, however, characterized by low productivity in terms of growth rate, meat production, and reproductive performance (Solomon et al. 2013). Poor nutrition is the leading cause of livestock's failure to reproduce, and thus focused feeding is a strong tool for manipulating reproduction in farmed animals. Addah et al. (2007) examined the cost of undernutrition at each stage of pregnancy and determined that nutrition has a greater influence on intrauterine growth than genetics. Poor nutrition affects production and reproduction in sheep by influencing fetal losses, lamb birth weight, milk supply, and ewe mortality (Addah et al. 2007). Furthermore, due to the impacts on lamb birth weight, colostrums and milk supply, and ability to suckle, Dwyer et al. (2016) asserted that ewe nutrition was directly demonstrated as a primary driver for enhancing newborn survival.

Ewes require proper nutrition throughout the production cycle, beginning with weaning and continuing through gestation and lactation. Pre-and post-tupping nutrition has an impact on the number of fetuses formed, as well as oocyte/follicular maturation, ovulation, embryo development, and implantation. As the pregnancy advances, this extends to fetal survival, birth weight, vitality, and colostrum production (Robinson et al., 2005). Nutrition has long-term implications for the reproductive ability of the next generation, in addition to these short-term effects on the reproductive activity of the parents (Fowden et al., 2006). As a result, any animal production system will only be successful if flock nutrition is adequate throughout the reproductive cycle and both male and female requirements are met.

Legume forages are one of the supplements that have been employed in ruminant nutrition (Savon, 2005). Because of their easy availability on the farm, high nutritional content, and low feeding cost, forage supplements have significant promise for ruminant production in the tropics (Adugna, 2012). Jabbar et al. (1997) found that supplementation of *Leucaena* and *gliricidia* in late pregnancy and lactation boosted offspring survival to weaning from under 50% on a panicum diet to over 95% when given a *Leucaena/gliricidia* combo as 40% of total dry matter intake. This finding highlights the importance of seizing the chance to substitute conventional, high-cost protein supplements with low-cost, farm-grown forage legumes in smallholder farmers' situations.

Though a few studies on the Horro sheep breed's growth and carcass traits have been conducted at Bako and elsewhere in Ethiopia (Assefu, 2012; Mekonnen et al., 2016; Abuye et al., 2018; Gemeda et al., 2002a, 2007b), no comparative study on the ewes' reproduction and production characteristics of this sheep breed using available feed resources has yet been done. The lack of such data is predicted to stymie the establishment of a more effective sheep breed improvement program, both in the current study area and across the country. As a result, the purpose of this study was to see how supplementing two *Lablab purpureus* varieties, Beresa-55 and Gebis-17, with a concentrate mixture affected the reproductive and productive performance of Horro ewes managed under free-grazing conditions.

## **Materials and Methods**

### ***Study area***

The study was conducted at the Bako Agricultural Research Center (BARC). The location symbolizes western Ethiopia's mid-altitude sub-humid maize-growing agro-ecology. The center is located at a height of 1650 meters above sea level, with a latitude and longitude of 9° 06' N and 37° 09' E, respectively. The area receives 1316.7 mm of rain each month on average, with monthly minimum and maximum temperatures of 11.230 °C and 31.740 °C, respectively. BARC's soil is mostly reddish-brown and has a clay-loam texture (Wakene, 2001).

### ***Feed preparation***

Two varieties of *Lablab purpureus* (Beressa-55 and Gebisa-17) were planted on about one hectare of land at the Bako Agricultural Research Centre. During plantation, all prescribed agronomic techniques at BARC were properly followed for both varieties. Weeds were controlled by hand weeding and hoeing until the crops were ready to be harvested for fodder. It was harvested at 50% flowering, chopped to 3-5 cm in length with a chopping machine to make it more consistent and accessible to the animal, field-cured

for 2-3 days depending on the weather, then baled, and stored in a roofed hay barn. Wheat bran (WB) and Noug seed cake (NSC) thought to be sufficient for the full study period were acquired from nearby wheat and oil processing companies. A mixture of NSC 49.5% and WB 49.5% ratio with 1% salt was formulated and provided to one of the treatment groups considered as a positive control.

### ***Experimental ewes feeding, design, and treatment***

A total of 21 ewes of the same parity weighed a mean pre-mating weight of 26.3 + 2.1 (Mean + SD) were employed in the study. The experiment was conducted using a randomized complete block design with three treatments of seven ewes each. To limit variation among experimental ewes, ewes were grouped based on pre-mating weight, and just one ram was permitted to breed all ewes to eliminate the ram effect. Ewes were flushed with high-quality hay for 45 days before mating. The supplemental feed was then supplied to the ewes throughout their gestation periods and until the lambs were weaned, as per the treatment plan. Except for one hour every morning when each group was separated to receive their respective supplemental diet consisting of two legume forages varieties (Beres-55 and Gebis-17) and concentrate mixture with common salt, all ewes were kept together in the same pasture to graze for eight hours per day. The dietary treatments are: grazing + concentrate mixture (T1); grazing + Gebisa-17 (T2); grazing + Beresa-55 (T3).

The standards for Ethiopian indigenous sheep and goat breeds developed by Alemu Y. (2010) were used to calculate the level of supplementation at each stage of pregnancy. Based on this recommendation, one of the treatment groups (T1) received a mixed concentrate containing 49.5 percent wheat bran, 49.5 percent Noug seed cake, and 1% salt of 200 gm/head/day during the early stages of pregnancy, 300 gm/head/day during the mid-stage of pregnancy, and 450 gm/head/day during the late stages of pregnancy, in the hope that such levels of concentrate supplements would meet the nutrient requirements of pregnant ewes and/or does. As a result, diet in T2 and T3 that is being supplemented at each pregnancy stage was determined based on their crude protein (CP) content to make them iso-nitrogenous to the concentrate mixed supplements. Ewes were also given 200 gm/head/day of mixed concentrate during lactation, which is a general rule of thumb that should be supplemented for each offspring being fed and adjusted accordingly (Alemu Y., 2010).

### ***Reproductive parameters and live weight measurements***

At parturition, the ewe's weight at lambing, litter size, and lamb birth weight are all recorded. Reproductive parameters were computed according to Gatenby (1986) as:

Prolificacy = (number of lambs born/number of ewes lambing) x 100;

Fecundity rate = (number of lambs born alive/number of ewes mated) x 100;

Lambing rate = (number of lambing ewes/number of mated ewes) x 100;

Abortion rate = (number of ewes aborted/number of ewes mated) x 100;

Pregnancy rate = (number of ewes pregnant/ewes present to rams) x 100.

Gestation length for each ewe is the number of days ranging from the date of mating up to the date of parturition.



Pre-mating weight was taken to get the initial body weight. Then, before releasing the animals for grazing, a weight record was taken every fifteen days in the morning. Similarly, newly born offsprings were weighed right after birth to determine their initial birth weight and then every ten days until weaning age (3 months). The following calculation was used to calculate the average daily BW gain for each sheep and her offspring (Gulten et al., 2000).

**For ewes:**

$$\text{Average daily weight gain} = \frac{\text{final weight} - \text{pre-mating weight}}{\text{Number of gestation days}}$$

**For lambs:**

$$\text{Average daily weight gain} = \frac{\text{weaning weight} - \text{birth weight}}{\text{Weaning periods (3 months)}}$$

**Chemical analysis**

Dry matter (DM), crude protein (CP), and ash were analyzed according to the AOAC (2005) procedure. Crude protein (CP) was estimated by multiplying the N value by a factor of 6.25. Neutral detergent fiber (NDF), acid detergent fiber (ADF), and acid detergent lignin (ADL) were analyzed using the procedures of Van Soest et al. (1991). In *vitro*, organic matter digestibility was determined using the Tilley and Terry (1963) method.

**Statistical analysis**

Data collected from this experiment were analyzed by using the GLM procedure of SAS software (SAS, 2002, version 9.1.3) and means were separated by using the Least Significant Difference test at a 5% level of significance.

**Results and Discussion**

**Chemical composition of the feeds**

Table 1 shows the results of the chemical analysis of the experimental feeds used in this study. The dry matter (DM) composition of all experimental feeds was nearly the same, but there was variation in the remaining nutrient components. The DM, CP, ADF, and ADL levels in the Noug seed cake meal used in this study were higher than those reported by Abebe (2006) for the same nutrient classes. On the other hand, Ermias (2008) found a somewhat higher DM (93.9%) value than the one observed in the current study but a remarkably similar level of ADF (19.9%) content. On the other hand, these authors reported lower levels of ash (6.6%), CP (24.8%), and NDF (23.1%) than the results of the current investigation. The DOMD value achieved for Noug seed cake in this investigation is equivalent to the value of 69.20% reported by Tesfaye et al. (2016).

Table 1: Chemical composition of the experimental feeds

Feed samples	DM%	Nutrient composition (%DM)					
		Ash	CP	NDF	ADF	ADL	DOMD
Noug seed cake	92.33	7.02	29.43	34.56	19.2	7.17	71.04
Wheat bran	91.88	3.82	16.54	39.06	7.96	1.04	75.32
Gebis-17	91.85	10.08	22.92	48.34	32.8	5.57	62.28
Beres-a-55	91.99	13.46	23.46	44.3	30.31	5.64	63.88

*DM = dry matter; CP = crude protein; NDF = neutral detergent fiber; ADF = acid detergent fiber; ADL = acid detergent lignin; DOMD = digestible organic matter in dry matter*

The CP value of wheat bran used in this study is higher than that reported by Teklu (2016) and Abebe (2006), who reported 13 and 15.24%, respectively. However, it was lower than the levels reported by Dereje (2015), Hunegnaw and Birhan (2016), and Mekonnen et al. (2016), which were 17.9%, 17.77%, and 17.46%, respectively. On the other hand, the DM, ash, DOMD, and ADL contents of WB in the current study were in line with Mekonnen et al. (2016)'s corresponding values of 88.09, 4.89, 72.35, and 3.07%, but were lower for ADF and NDF contents.

The crude protein (CP) levels of the Beresa-55 and Gebis-17 varieties were 19.92 and 20.2%, respectively, which are comparable to the values reported by Worknesh (2014) and Hunegnaw and Birhan (2016). Most herbaceous legumes, according to Norton (1982), contain CP (greater than 15 percent), which is generally required for lactation and growth. As a result, the CP content of both the Beresa-55 and Gebis-17 varieties employed in this study coincides with this report and can thus be used as a supplement to Horro ewes based on a low-quality diet. Both Beresa-55 and Gebis-17 had greater ash values than the rest of the experimental diets. The current study's NDF and digestible organic matter in dry matter values were higher than those discovered by Abuye et al. (2018) for the same forage varieties, while the ADF and ADL values were lower

### ***Feed intake of pregnant Horro ewes***

Table 2 shows the average daily dry matter and nutrient intakes of Horro ewes estimated from their supplemental feeding. Except for total crude protein intake ( $P > 0.05$ ), the remaining intake characteristics were substantially different ( $P 0.001$ ) among the dietary regimens. Ewes fed a T3 diet consumed more total ash than ewes fed a T2 or T1 diet. The increased ash concentration in Beresa-55 (T3) compared to Gebis-17 (T2) and the concentrate mixture could be the cause of this difference (T1).

Table 2: Daily feed and nutrient intake of pregnant Horro ewes supplemented with Beresa-55, Gebis-17, and concentrate mixture

Intake (g/day)	Treatments			SEM	Sign.
	T1	T2	T3		
<b>Supplemental feed DMI</b>					
Beresa-55	-	-	280.45	-	-
Gebis-17	-	287.48	-	-	-
Concentrate mixture	286.93	-	-	-	-
<b>Nutrient intake</b>					
Ash	15.56 <sup>c1</sup>	28.98 <sup>b</sup>	37.75 <sup>a</sup>	0.08	***
CP	65.99	65.89	65.79	0.16	ns
NDF	105.61 <sup>c</sup>	138.97 <sup>a</sup>	124.24 <sup>b</sup>	0.29	***
ADF	38.99 <sup>c</sup>	94.29 <sup>a</sup>	85 <sup>b</sup>	0.2	***
ADL	11.79 <sup>c</sup>	16.01 <sup>a</sup>	15.82 <sup>b</sup>	0.04	***
DOMD	209.97 <sup>a</sup>	179.04 <sup>b</sup>	179.15 <sup>b</sup>	0.42	***
ME (MJ/day)	3.36 <sup>a</sup>	2.86 <sup>b</sup>	2.87 <sup>b</sup>	0.01	***

<sup>1</sup>Means within rows followed by different letters differ significantly; \*\*\*=( $P < 0.001$ );

ns= not significant; SEM= standard error of means; Sign.= significance; DMI= dry matter intake; CP= crude protein; NDF= neutral detergent fiber; ADF=acid detergent fiber; ADL= acid detergent lignin; DOMD= digestible organic matter in dry matter; ME= metabolizable energy; DM= dry matter; T1, T2 and T3= treatments.

Ewes in T2 consumed more fiber components (NDF, ADF, ADL) than those in T1 and T3. This discrepancy is most likely due to the supplemental feeds' fiber content, as the Gebis-17 had greater NDF, ADF, and ADL values than the Beresa-55 and concentrate mixture. Ewes fed the concentrate mixture had the highest total intake of digestible organic matter in dry matter (DOMD) and metabolizable energy (ME), followed by ewes fed Gebisa-17 and Beresa-55. Since the current study lacks enough information to estimate the intake from grazing, it is impossible to draw a precise conclusion on the daily total dry matter and nutrient intake parameters of the experimental ewes.

### **Live Weight gain of the ewes**

Live weight gain of the pregnant Horro ewes is indicated in Table 3. With the exception of pre-mating weight, all growth indicators showed significant ( $P < 0.05$ ) variations across the dietary regimens. The T1 diet resulted in the highest final body weight, which was similar to the T3 diet. On the other hand, T2 and T3 ewes' final body weights are statistically comparable. T2 ewes grew at a slower pace of 4.8 kg (37.1 vs. 32.34 kg) than T1 ewes. This appears to be related to T2 ewes' increased total fiber intake (Table 2), which may have reduced the intake of digestible nutrients available for absorption and metabolism.

Table 3: live weight gain of pregnant Horro ewes supplemented with Beresa-55, gebis-17, and concentrate mixture.

Parameters	Treatments			SEM	Sign.
	T1	T2	T3		
Premating body weight (kg)	25.85	26.71	26.29	0.93	ns
Final body weight (kg)	37.1 <sup>a1</sup>	32.34 <sup>b</sup>	34.23 <sup>ab</sup>	1.16	*
Body weight change (kg)	11.24 <sup>a</sup>	5.63 <sup>b</sup>	7.94 <sup>b</sup>	0.83	**
daily weight gain (g/day)	76.94 <sup>a</sup>	42.11 <sup>c</sup>	58.05 <sup>b</sup>	4.44	**

<sup>1</sup>Means within rows followed by different letters differ significantly; \* = ( $P < 0.05$ ); \*\* = ( $P < 0.01$ ); ns = not significant; SEM = standard error of means; Sign. = significance; ns = not significant; T1, T2 and T3 = treatments

The body weight change and daily weight increase characteristics found for ewes in the various feeding regimens did not follow the same pattern as the final body weight. Ewes fed the T1 diet had a greater change in body weight than those fed the T2 and T3 diets, but T2 and T3 are statistically at par. Daily weight gain, on the other hand, was higher for ewes in T1, T3, and T2, in that order. This disparity could have stemmed from variations in gestation time, in addition to differences in weight change among the dietary interventions. The live weight increase during pregnancy, which is the result of intrauterine growth and maternal body weight change, is commonly employed as a measure of nutritional sufficiency and is also used to compare the impact of various dietary treatments (Amoah et al. 1996). As a result, the final body weight gain achieved by ewes fed diets in T1 and T3 demonstrated that the diets had a higher nutritional content than the diet in T2. The final weight gain obtained in this study is substantially larger than Solomon et al. (2004) reported for Menze sheep, which ranged from 20.3 to 25.2 kg. This discrepancy could be explained by differences in sheep breeds and extra diets employed in the two investigations.

#### ***Trends in ewes` body weight change***

Figure 1 depicts the trend in the ewes' body weight changes throughout their gestation period. Even though there was no statistical difference ( $P > 0.05$ ) between ewes in T1 vs T3 and T2 vs T3, ewes in T1 and T3 had a faster growth rate than ewes in T2. Nonetheless, it can be concluded from the current study that all treatment diets increased the live bodyweight of the ewes during the gestation period. The nutritional value of the two *Lablab purpureus* varieties in general, and that of the Beresa-55 variety in particular, can be inferred to be comparable to the nutritional value of concentrate feed used to supplement Horro ewes during their gestation periods.

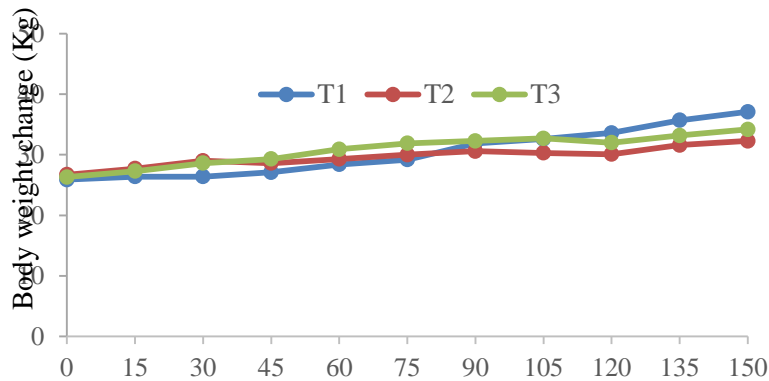


Figure 1: Trends in body weight of Horro ewes supplemented with Gebisa-17 and Beresa-55 varieties and concentrate feeds.

### Live weight gain of lambs

The number of male and female lambs born, as well as their birth weight (BW), weaning weight (WW), body weight change (BWC), and average daily gain (ADG) of ewes fed their respective supplemental diets, are shown in Table 4. A total of 18 lambs were produced from the 21 ewes used in the study. Due to the incidence of two abortion instances and one ewe that did not conceive, there are fewer lambs than expected. It was hypothesized that lambs born to ewes fed a concentrate combination would grow faster than those born to Gebis-17 and Beresa-55 varieties. Although there was no statistically significant data ( $P > 0.05$ ) in this study, it is interesting to note that lambs produced from ewes fed the three different nutritional diets performed similarly. According to Nsahlai and Umunna (1996), the nitrogen in *Lablab purpureus* is rapidly degradable in the rumen, which is beneficial in meeting the needs of rumen bacteria for efficient decomposition of low-quality diets, resulting in improved animal performance.

Table 4: live weight gain of offspring obtained from Horro ewes supplemented with Beresa-55, Gebis-17, and concentrate feeds.

Items	Treatments			SEM	Sign.
	T1	T2	T3		
Male (12)	5	3	4	-	-
Female (6)	1	3	2	-	-
Live weight change					
Birth weight (kg)	3.01	3.42	3.42	0.21	ns
Weaning weight (kg)	10.89	9.87	11.57	0.9	ns
Weight change (kg)	7.89	6.45	8.15	0.98	ns
Daily weight gain (g/day)	87.63	71.67	90.55	10.89	ns

ns= no significant; SEM= standard error of means; T1, T2 and T3= treatments

The lack of statistical difference in BW, WW, BWC, and ADG among lambs borne from ewes in the various dietary regimens indicated that the supplements were equal in their ability to provide nutrients to lactating ewes throughout lactation. The weaning weight recorded in this study is greater than that reported by Liliane et al. (2014) with values ranging from 9.03 to 9.15 kg but comparable to the value reported in a range from 9.8 to 10.1 kg (Amole et al. 2016), and the mean value of 11.81 kg (Solomon et

al 2000). The birth weight of lambs assessed in this study is higher than previously reported. For example, Solomon et al. (2000) recorded the birth weights of lambs varying from 2.63 to 2.77 kg for the same sheep breed. Mukasa-Mugerwa et al. (2000) reported a mean lamb birth weights of 2.4 kg for Horro sheep and 2.1 kg for Menz sheep. Furthermore, birth weights ranging from 2.1 to 2.97 kg for Ethiopian indigenous sheep breeds were reported also in the literature (Kassahun, 2000; Mukasa-Mugerwa et al. 2000; Surafel et al. 2012). The considerably higher lamb birth weight observed in the current study show that supplementing ewes with high-quality feed sources throughout the gestation period benefits lamb birth weight, as lambs with higher birth weights have a better chance of survival and future growth. EI-Hag et al. (1998) who evaluated the production performance of Sudan desert ewes under range circumstances reported lamb birth weights ranging from 2.93 to 4.42 kg which is consistent with the value recorded in this study.

### ***Trends in lambs` body weight changes***

Figure 2 depicts the changes in lamb body weight (kg) over the weaning periods. The weights of lambs in T1, T2, and T3 grew positively, with similar growth rates. This shows that the supplements were comparable in their ability to provide nutrients to lactating ewes throughout lactation to alleviate nutritional stress.

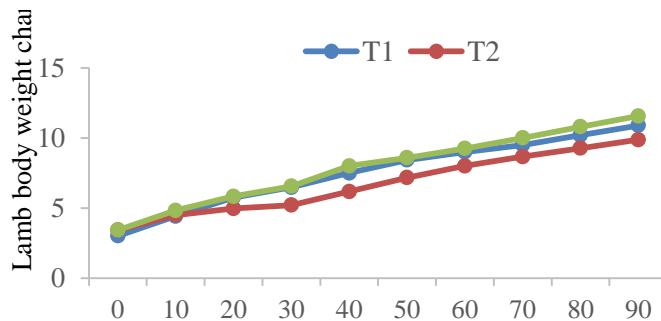


Figure 2: Trends in body weight of lambs borne from the ewes supplemented with Gebisa-17 and Beresa-55 varieties and concentrate feeds.

### ***Ewe reproductive performance***

Table 5 shows the reproductive performance of Horro ewes treated with two *Lablab pureurues* varieties (Gebis-17 and Beresa-55) and concentrate mixed diets. Only the rate of pregnancy and abortion rate was significantly different ( $P < 0.001$ ) in the ewes fed with the two *Lablab pureurues* varieties and concentrate mixed diets, but not the rest of the reproductive features studied ( $P > 0.05$ ). Neither of the dietary interventions resulted in the delivery of twins in the ewes. Ewes fed the T2 diet had a shorter gestation period (150.5 days), while ewes fed the T1 diet had a longer gestation period (151.8 days). This finding is consistent with reports in the literature (Solomon et al., 2000; Mukasa-Mugerwa et al., 2000; Solomon et al., 2004; Idris et al., 2010). When compared to ewes fed the T2 and T3 diets, ewes on the T1 diet had a lower pregnancy rate. This was due to the presence of one non-pregnant ewe in T1. In this study, two abortion cases were observed, one from each of the T2 and T3 groups of ewes. To determine the cause of the abortions, more research is needed to look into the existence of potentially hazardous substances in Beresa-55 (T3) and Gebis-17 (T2) that could have hampered embryonic and fetal development in ewes fed those feeds.

Table 5: Reproductive performance of Horro ewes supplemented with Beresa-55, Gebis-17, and concentrate feeds.

Parameters	Treatments			SEM	Sign.
	T1	T2	T3		
Gestation length (day)	151.8	150.5	151.2	1.18	ns
Pregnancy rate (%)	85.71 <sup>b</sup>	100 <sup>a</sup>	100 <sup>a</sup>	0	***
Prolificacy (%)	100	100	100	0	ns
Fecundity rate (%)	85.71	85.71	85.71	0	ns
Lambing rate (%)	85.71	85.71	85.71	0	ns
Abortion rate (%)	0 <sup>b</sup>	14.29 <sup>a</sup>	14.29 <sup>a</sup>	0	***
Weight at lambing (kg)	33.42 <sup>a</sup>	28.85 <sup>b</sup>	30.89 <sup>ab</sup>	1.14	*

<sup>1</sup>Means within rows followed by different letters differ significantly, \*= (P<0.05); \*\*\*= (P<0.001); SEM= standard error of means; Sign.= significance; ns= not significant; T1, T2 and T3= treatments

Ewes in T1 and T3 gained more weight at lambing than ewes in T2, despite the fact that T1 vs T3 and T2 vs T3 are statistically equal in this parameter. The weight of ewes at lambing followed the same pattern as their final weight before lambing, though to a lesser extent. The reduced weight of the ewes at lambing is most likely due to parturition stress. Weight at lambing for Sudanese desert sheep ranged from 31.3 to 36.8 kg was reported by EI-Hag et al. (1998), which was greater than the values seen in this study. Solomon et al. (2004), however, who evaluated the productive and reproductive performance of the Menth sheep breed found lower values ranging from 17.1-23.1 kg. The differences between the current study and the previous studies may be attributed to variations in sheep breed, feed type and quality, and the composition of the grazed natural pasture.

### Conclusion and Recommendation

The current study found that supplementing Horro ewes with high-quality feeds during their gestation phase helped them perform better in terms of productivity and reproduction. There were no significant differences among the dietary regimens studied for nearly all of the productive and reproductive variables evaluated. Even though the T1 (concentrate combo) diet improved the reproduction and production of sheep in a similar way to the Beresa-55 and Gebis-17 varieties, the use of concentrate supplements is usually limited due to its inaccessibility and high cost. As a result, it's critical to seize the chance to replace this conventional and expensive feed with low-cost, on-farm cultivated forage legumes like the ones used in this study. As a result, supplementing Horro ewes with either Beresa-55 or Gebis-17 varieties as an alternate protein source during their breeding season is a viable technique for maintaining their genetic potential. Supplementation's effect on lambs' post-weaning growth rates and survival will need to be studied more in the future.

### Acknowledgment

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# **Poultry Research Results**

# The effects of roasted pigeon pea meal on feed intake and growth performance of cockerel koekoek chickens

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## **Abstract:**

*One hundred twenty eight (128) cockerel koekoek chickens with four weeks of age were distributed according to a Complete Randomized Design of four treatment diets each containing 0%, 25%, 30% and 35% pigeon pea meals to evaluate the feed intake and growth performance of the chicken. Each treatment was replicated four times. Each replicate contained eight (8) cockerel koekoek chickens. Cockerels in each of the replicat group were reared in group feeding pens for 84 days. Feed offered and refusal were measured daily during the experimental period. Differences in daily feed intakes (in gram per bird) of birds in treatment one and two ( $74.89 \pm 12.78$  vs.  $73.09 \pm 13.4$ ) as well as those in treatment three and four ( $71.70 \pm 14.2$  vs  $69.16 \pm 14.43$ ), were not statistically significant ( $P > 0.05$ ). Similarly, total feed intake, in grams per bird, was not significantly ( $P > 0.05$ ) different between treatment one ( $6291 \pm 12.78$ ) and treatment two ( $6139.56 \pm 13.47$ ), as well as between treatment three ( $6022.8 \pm 14.2$ ) and treatment four ( $5848.92 \pm 12.8$ ) but there were significant differences ( $P < 0.05$ ) in total feed intakes per bird between treatment one and three, between treatment one and four, between treatment two and three and between treatment three and four. Significantly lower ( $P < 0.05$ ) daily weight gain in gram per bird was observed in treatment four ( $19.45 \pm 0.53$ ) compared to the other treatments but there were no significant ( $P > 0.05$ ) differences among treatment one ( $23.94 \pm 1.4$ ), treatment two ( $21.97 \pm 1.16$ ) and treatment three ( $21.6 \pm 0.6$ ) in gram per bird. Significantly ( $P < 0.05$ ) lower FCR was observed in treatment one ( $3.03 \pm 0.54$ ) compared to the rest treatments. Differences in FCR of treatment two ( $3.33 \pm 0.61$ ), treatment three ( $3.31 \pm 0.67$ ) and treatment four ( $3.56 \pm 0.74$ ) were not significant ( $P > 0.05$ ). The prices of cockerels koekoek chickens of treatment one (Birr  $271.67 \pm 5.77$ ), treatment two (Birr  $257.67 \pm 17.04$ ) and treatment three (Birr  $238.67 \pm 16.17$ ) did not differ ( $P > 0.05$ ) significantly. Similarly, the price of birds in treatment three (Birr  $238.67 \pm 16.17$ ) and treatment four (Birr  $217.67 \pm 10.78$ ) did not differ ( $P > 0.05$ ) significantly, but there was a significant difference between the price of birds in treatment one ( $271.67 \pm 5.77$ ) and in treatment four (Birr  $217.67 \pm 10.78$ ). From this study, net incomes of Birr 148.74, 138.25, 102.7 and 74.37 were obtained from T2, T1, T3 and T4, respectively. Generally, from the biological and economical point of view, the inclusion of 25% pigeon pea meal in cockerels' koekoek chicken diet is recommended in areas where the crop can be grown..*

**Key words:** pigeon pea meal, feed offered, feed intake, feed conversion, growth performance,

## **Introduction**

Poultry are unable to manufacture essential amino acids, the B vitamins and they cannot exist on high fiber diet. Their diet must contain the materials essential for maintenance, production and reproduction. Poultry diet containing high levels of protein are highly essential for maintenance, production and reproduction but expensive to purchase (Smith, 2001). The high cost and unavailability of commercial protein supplements is one of the main limitations to efficient animal production by small holder farmers

(Martens *et al.*, 2012). Feeding improperly at any stage can result in poor growth and poor egg production.

Introduction of fast growing and genetically improved strains has posed challenges to nutritionist in most developing countries. This is because the feed ingredients required to achieve optimum performance from these birds are getting increasingly expensive due to competition with man. This has prompted renewed effort at finding cheaper alternative to sustain production of poultry strain at a reasonable cost. The competition between food and feed is expected to further increase feed prices, forcing producer to look for alternate feeds and locally available protein source feeds for birds. Interests have naturally been on the oilseeds, the legumes in particular.

The feed constraint can be resolved by increasing the acreage dedicated to maize and soybean according to livestock master plan (LMP ,2015) but soybean cultivation is rare in mid rift valley area. Locally grown forages and grain legumes like pigeon pea offer ecological benefits for nitrogen fixation, soil improvement and erosion control which contribute to improve crop production. Besides these benefits its grain is used as an alternative protein source in the diet of poultry to reduce farmers` dependence on externally purchased protein concentrate. Pigeon pea can be used as protein source for heavier birds like koekoek chickens for the production of eggs and more muscles because these heavy breeds require plenty of high quality feeds.

The amino acid profile of pigeon pea is comparable with the conventional plant protein sources (Akande *et al.*, 2010). With regard to chemical composition, pigeon pea was found to contain 22-27% crude protein, 7.3-10% crude fiber, 61.2% NFE, 1.7-2.1% EE, 3.1-4.2 %ash, and lysine about 7.59 % (Girmand, 1988; Ahmed *et al.*, 2006). Pigeon pea is a good source of dietary minerals such as calcium, phosphorous, magnesium, iron, sulfur and potassium. Also pigeon pea (*cajanus cajan*) is a good source of soluble vitamins especially thiamin, riboflavin, niacin and choline (Singh, 1977). This makes it the best source of feed for livestock in addition to manufacture of the by product. Pigeon pea (*Cajanus cajan*) seeds are currently considered as a non-conventional feed stuff in poultry feeding and as a valuable protein feed resource (Amaefule *et al.*, 2000).

Protease inhibitor and cyanogens, the two anti-nutritional factors in pigeon pea seed, are heat labile and could be effectively removed by heat processing (Khadiga *et al.*, 2009). According to Yisa *et al.*, (2010), processing of pigeon pea seed at 100<sup>0</sup>c reduced hydrogen cyanide by 23.53%, Phytic acid by 6.07% tannin by 99.06% and trypsin inhibitors by 79.36%. Processing pigeon pea seeds also significantly improved its utilization and crude protein retention (Amaefule *et al.*, 2005). Khadiga *et al.*, (2009); Yisa *et al.*, (2010) found that the nutritional value of processed decorticated and roasted pigeon pea seeds was suitable to be used as chicken diet. The feed demand of heavier breed like koekoek chickens can be overcome through using non-conventional feed resources such as roasted pigeon pea seeds which are not used for human consumption. However, information is scarce on levels of pigeon pea meal utilization for cockerel koekoek chickens feeding. Therefore, this activity was proposed to address this issue with the following objectives:.

- To evaluate the growth performance of cockerel koekoek chickens fed different levels of pigeon pea seed meal.
- To know the economic advantage of pigeon pea seed meal using in chicken feed.

## Materials and Methods

### *Description of the study area*

The experiment was conducted at Adami tulu agricultural research center, located in the mid rift valley of Ethiopia at an altitude of 1650 m.a.s.l. and 7°9'N latitude and 38°7'E longitude. The average annual rain fall of the area was 700 mm and its mean maximum and minimum temperatures were 27 and 12.5°C, respectively (ATARC, 2020).

### *Feed management*

Pigeon pea seed was purchased from the nearby farmers. The seeds were roasted and then milled before it was used for the formulation of the experimental diets. Other feed ingredients like wheat bran, maize, noug seed cake, Limestone, grower premix, and salt were purchased from the local market and added in to the rations. Finally three poultry rations were formulated using the above feed ingredients (Table 1) and commercial grower chicken feed to be used as control feed was purchased from the feed processing company. The commercial grower chicken feed was composed of soybean, maize, noug seed cake, wheat bran, bone meal, grower premixes, salt and limestone.

Table 1: Composition (%) of the three experimental feeds

Ingredient	TR2	TR3	TR4
Pigeon pea	25.00	30.00	35.00
Noug cake	4.00	4.00	4.00
Maize	51.00	51.00	51.00
Wheat bran	12.00	9.00	9.00
Salt	2.00	2.00	2.00
Limestone	4.00	2.00	2.00
Premixes	2.00	2.00	2.00

From the commercial diet and from each of the formulated diet, 120 gram feed was daily provided per chicken in each respective treatment. Left over feeds were collected every next day morning and weighed before providing feed for the day. Water was provided *ad libitum* during experimental period.

### *Animal management*

A total of 128 cockerels koekoek chickens of four weeks of age were selected from chickens reared in Adami Tulu Agricultural Research Center poultry farm. The Cockerel koekoek chickens were assigned to the four treatment diets. Eight (8) cockerels were assigned for each treatment diet and each treatment was replicated four times. Deep litter housing system that was partitioned in to 16 equal size pens (4 m<sup>2</sup>) was used. The partitioning was made using wood and mesh wire. Before placing the experimental chickens in to the experimental pens, the whole units were cleaned and disinfected with Dizinon disinfectant and littered with properly dried tef (*Ergroscopic tef*) straw.

### ***Data collected and performance parameters considered***

Feeds offered to the chickens and refusals were measured every morning. The difference between the amount of offered feed and refusals were calculated as intake. The birds were weighed at the beginning of the experiment and subsequently on two weeks basis usually in the morning (8.00 - 9.00 am) hours before providing feed. Weight gain of the birds was calculated as the final live weight minus initial weight. Feed conversion ratio (FCR) was calculated as feed intake divided by weight gain. Mortality records and other observations were kept throughout the period of study.

### ***Statistical analysis***

Analysis of variance of feed intake and weight gain of cockerel chicken were done using the general linear model (GLM) procedure of the Statistical Analysis System (SAS, 2001). When the results were significant, mean comparisons were made using Turkey multiple range test procedure of the SAS package (2001)

$$\text{Models: } Y_{ik} = \mu + B_i + e_{ik}$$

Where:  $Y_{ik}$  = individual value of the dependent variables of chicken,  $\mu$  = Overall mean;  $B_i$  = the effect of the level of pigeon pea ( $i = 1$  to  $4$ ),  $e_{ik}$  = random error

### ***Economic analysis***

Variable costs collected include prices of dry matter feed intake per bird, vaccine, medicine and disinfectant costs used. Net return was obtained from the estimated price of cockerel chicken based up on local chicken market. The economic benefit was estimated by considering partial budget analysis, according to the formula developed by CIMMT (1988).

$$NI = TR - TVC$$

Where, NI = Net income, TR = Total return, TVC = Total variable cost.

## **Results and discussions**

### ***Chemical composition of the experimental feeds***

The chemical compositions of the formulated experimental diets were analyzed at Adami Tulu Agricultural Research Center Animal Nutrition laboratory using the method AOAC (1990) proximate principles and the nutritional composition of the control feed was taken from the feed processing company. Nutritional composition of the feed treatment is given in Table 2.

Table2. Nutritional composition (%) of experimental diets used in cockerels' koekoek chicken

TR	DM	Ash	OM	CF	CP	NDF	ADF	ME(kcal/kg DM
1	94.75	5.6	89.15	5.0	18.00	22.56	11.49	2950
2	94.77	5.5	89.27	6.5	18.50	23.08	11.46	2946
3	93.97	3.9	90.07	10.5	19.00	23.62	10.65	2943
4	94.72	7.4	87.32	11.0	19.50	25.87	10.61	2942

### ***Feed intake and live weight change of the birds***

The mean daily feed intake, total feed intake, daily weight gain, total weight gain, final body weight gain, feed conversion ratio and price of the cockerel koekoek chickens are summarized in Table 3. Total feed intake and daily feed intake in gram per bird were not statistically different in ( $P>0.05$ ) between treatment one and two, between treatment three and treatment four. But there is a significant ( $P<0.05$ ) difference in feed intake between treatment one and treatments three and four, as well as between treatment two and treatments three and four.

The energy content of the experimental diets which is the main determinant of feed intake (Muzim., *et al.*, 2017 and Melesse, 2007) was similar and the differences in feed intake were due to the increased crude fiber content of the experimental diets of treatments three and four because of higher level of pigeon pea seed meal inclusion. As the amount of roasted pigeon pea seed meal used increased the crude fiber content and protein content of the feeds increased and resulted in decreased feed intake and weight gain.

Increasing crude fiber affected the palatability of the diet and hence the feed intake. This is in agreement with Saeed *et al.*, (2007). The current finding also agree with Muzim *et al.*,(2017) who found lower diet intake by vanda chickens aged from 1 to 91 days because of the higher crude fiber content of the experimental diet used. Rajesh jha *et al.*, (2021) also reported that high fiber poultry diets decreased feed intake and body weight gain. The current finding is also in agreement with the findings of Krás *et al.*, (2013) who reported lower weight gain (WG) for birds fed higher dietary fiber feed of lower energy content. However, the current finding disagrees with the report of Salami *et al.*, (2017) who reported higher feed intake from using high level of crude fiber compared to using low level of crude fiber. This could probably be due to difference in the breed type used.

The current finding is in agreement with work of Etuk *et al.*,(2003) who found significantly ( $P<0.05$ ) lower feed intake and lower weight gain from using 40% cooked pigeon pea seed meal for cockerel chicken. Amaefule *et al.*, (2006), Mukhtar *et al.*, (2013), concluded and recommended that inclusion of 20% pigeon pea meal into the whole grower diet had no any adverse effect on growth performance of grower cockerels. On the other hand, Yisa *et al.*, (2010) recommended that processed pigeon pea seed meal can be included in cockerel diets up to 30% with out affecting the meat component of cockerels`. The current finding indicated the 25% roasted pigeon pea seed meal inclusion in whole cockerel diet did not adversely affect feed intake and body weight gain of cockerel koekoek chickens. In fact, it gave similar result with the commercial diet.

Significantly ( $P<0.05$ ) lower FCR was observed for treatment one compared to the other three treatments. This indicates the good quality of the diet as feed conversion ratio is the amount of feed required for a unit of weight gain. This is because of the lower crude fiber content, good palatability and essential amino acid content of the commercial diet, since feed processing companies prepare commercial diet with balanced amino acid for poultry.



Table 3 Feed intake and live weight change of the cockerel koekoek chicken fed the experimental diets (mean  $\pm$  s. deviation)

Parameters	Control (TR1)	TR2	TR3	TR4
Initial body weight (kg/bird)	0.35 $\pm$ 0.03	0.32 $\pm$ 0.16	0.33 $\pm$ 0.02	0.33 $\pm$ 0.03
Mean daily feed intake(g/bird)	74.89 $\pm$ 12.78a	73.09 $\pm$ 13.47a	71.7 $\pm$ 14.2b	69.16 $\pm$ 14.43 b
Mean total feed intake(g/bird)	6291 $\pm$ 12.78a	6139.56 $\pm$ 13.47a	6022.8 $\pm$ 14.2b	5848.92 $\pm$ 12.8b
Mean daily weight gain (g/bird)	23.94 $\pm$ 1.4a	21.97 $\pm$ 1.16a	21.6 $\pm$ 0.6a	19.45 $\pm$ 0.53b
Mean total weight gain (g/bird)	2350 $\pm$ 12.14a	2150 $\pm$ 16.11a	2110 $\pm$ 10.06a	1910 $\pm$ 14.05b
Final body weight (Kg/bird)	2.70 $\pm$ 0.11a	2.48 $\pm$ 0.11b	2.45 $\pm$ 0.40b	2.24 $\pm$ 0.02c
FCR (feed : gain)	3.03 $\pm$ 0.54 b	3.33 $\pm$ 0.61 a	3.31 $\pm$ 0.67 a	3.56 $\pm$ 0.74 a

### Changes in feed intake and weight gain

Changes in average weekly feed intake and average weight gain of the cockerel koekoek chicken during the study period are indicated figures 1 and 2, respectively. As age of the chicken advanced their feed intake increased (Fig.1) because of the fact that the development of different organs, muscles and feathers need high amount of quality feed.

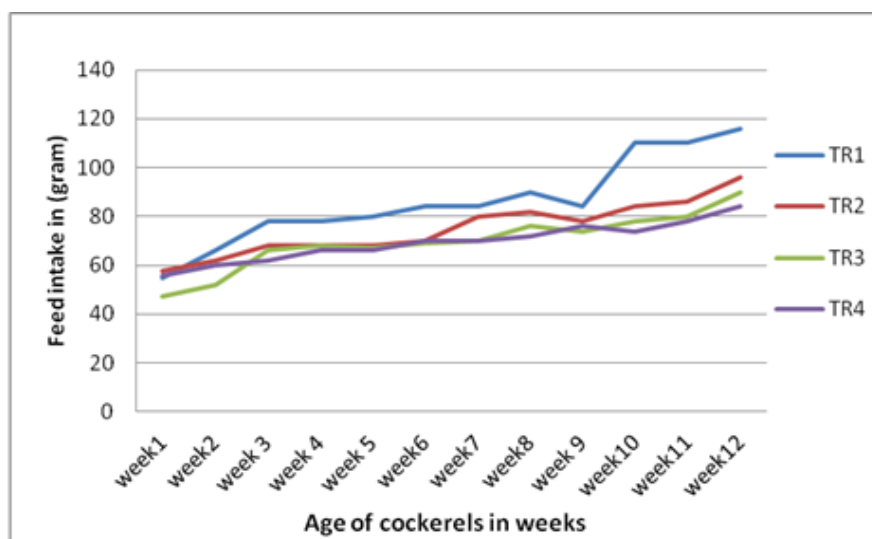


Figure1. Changes in average weekly feed intake of the cockerel koekoek chicken during the study period

The weekly weight gain of cockerel koekoek chickens (Figure 2) followed the same trend as the weekly feed intake. Especially after 9<sup>th</sup> week of age, the higher weight gain observed was directly related with the higher feed intake of the birds over the same period. The final weight that the experimental chicken attained in the current study, especially those in treatments one and two are similar with the previous values report (2.7 kg) by Tesfa *et al.*, (2018) for birds of five months of age.

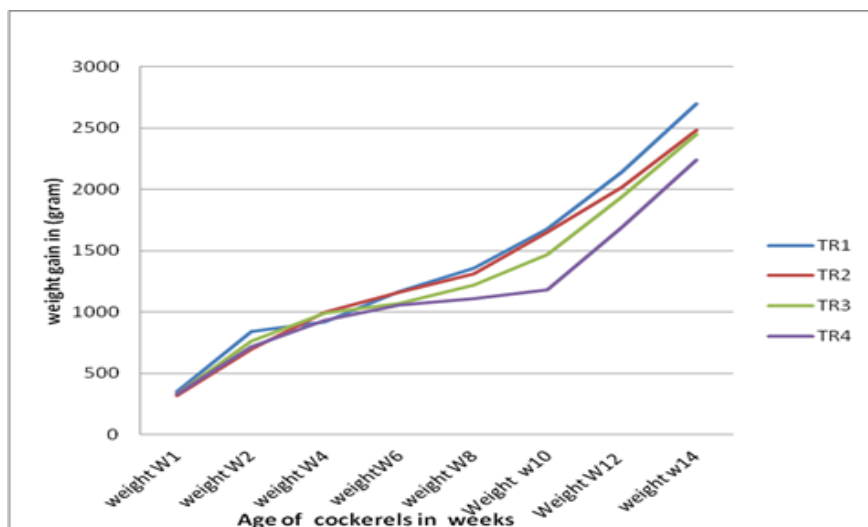


Figure 2. Average weight gain of cockerels koekoek chicken during the experimental period

### Partial budget analysis

The prices of cockerels of koekoek chickens in treatments one, two and three were not statistically different ( $P > 0.05$ ). The price of cockerel of koekoek chicken in treatments three and four were also not different ( $P > 0.05$ ) but there was a significant ( $P < 0.05$ ) difference between the price of koekoek cockerel in treatment one and four as well as between treatment two and four. All differences in prices are the results of weight gained from the feed consumed because in Ethiopia, chicken price estimation is highly dependent on the weight of chicken.

In this study the net income of Birr 148.74, 138.25, 102.7 and 74.37 was obtained from chickens in T2, T1, T3 and T4, respectively. Inclusion of pigeon pea more than 25% increased feed cost. This finding agrees with the findings of Amaefule *et al.*, (2005) who reported the inclusion of pigeon pea at lower percentage significantly reduced feed cost.

Table 4. Partial budget analysis are shown in

Partial budget cost	TR1(control)	TR 2	TR3	TR 4
Total feed consumed (kg/bird )	6.291	6.139	6.022	5.85
Day old chicken cost(EB)	10.00	10.00	10.00	10.00
Feed cost (EB)	74.42	49.93	76.97	84.3
Cost of Vaccine, Medicine and Disinfectant (ETB)	24.00	24.00	24.00	24.00
Cost of pen construction (ETB)	25.00	25.00	25.00	25.00
Total variable cost(TVC) (ETB)	133.42	108.93	135.97	143.3
Price of cockerel (GR)(ETB)	271.67±5.77a	257.67±17.04ab	238.67±16.17 abc	217.67±10.78c
NR (GR-TVC)	138.25	148.74	102.70	74.37

## Conclusions and recommendations

Using pigeon pea seed meal as a protein source in poultry ration formulation enable formulation of good poultry ration with a reasonable cost for those who cannot purchase afford purchasing the expensive commercial poultry feeds. The roasted pigeon pea seed meal can be included up to 25% as a protein source in whole cockerels koekoek chicken ration and this reduced age at which cockerels reach for market. In hot environmental areas where feed intake affected by heat stress and where more concentrated feed needed for poultry, inclusion of higher percentage (greater than 25%) of roasted pigeon pea in cockerels ration negatively affected feed intake and weight gain of chickens. Before adding in to the whole ration, processing/roasting and grinding is very important to decrease its fiber content and to improve its palatability and intake. From the biological and economical data obtained, the 25% roasted pigeon pea meal inclusion in cockerels' koekoek chicken as protein source is recommended in cockerel koekoek chicken diet. Since pigeon pea has many functions in addition to its forage value, during popularization and training farmers, attention should be given on creating awareness on the value of its seed meal for poultry rearing.

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# **Feed resources and Rangeland Research Results**

# Evaluation of Bracharia grass Cultivars for their Agronomic Performance in Midland Areas of East Guji Zone Southern Oromia, Ethiopia.

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## Abstract

The study was conducted with the objective to identify adaptability, high biomass and dry matter yielder of Bracharia grass. Four Bracharia grass *Brachiaria mutica* Dzf No 18659 (Dzf 483), *Brachiaria Decumbens* Dzf No 194, *Brachiaria mutica* 6964 (Dzf No 484) and *Bracharia mulato* were evaluated in randomized complete block design (RCBD) with three replications. The result revealed that plot cover, fresh biomass, dry matter yield and plant height were highly significantly ( $P < 0.01$ ) differ among the treatments. The highest value of plant height (170 cm) was measured from *Brachiaria mutica* 6964 Dzf No 484 cultivar followed by (160 cm) *Brachiaria mutica* Dzf No 18659 (Dzf 483) cultivar, while the shortest (90 cm) plant height was recorded from *Decumbens* Dzf No 194 cultivar. The highest dry matter yield (11.95t/ha) was obtained from *Brachiaria mutica* 6964 Dzf No 484 genotype, followed by (11.82t/ha) *Brachiaria mutica* Dzf No 18659 (Dzf 483) cultivar. The highest survive rate (95.5%) was measured from *Brachiaria mutica* Dzf No 18659 (Dzf 483) genotype, followed by *Brachiaria mutica* 6964 (Dzf No 484) cultivars (87%). The result implies that *Brachiaria mutica* Dzf No 18659 (Dzf 483) and *Brachiaria mutica* 6964 (Dzf No 484) were well performed in agronomic parameters. Thus it could be possible to conclude that the Bracharia grass should be recommended for improving the constraint of feed shortage in midland agro ecologies of Guji zone and similar areas.

**Keys words:** Bracharia, Midland, Cultivars, Agronomic performance

## 1. Introduction

Livestock production is an integral part of the farming systems in all parts of Ethiopia. The sector has a share of 12-16% of the total Gross Domestic Product (GDP) and 30-35% of agricultural GDP (Ayele *et al*, 2002). It contributes to the livelihoods of 60-70% of the Ethiopian population. Moreover, it ensures the availability of food, creates jobs, transportation and income to the farming community, serve as a source of agricultural inputs such as draft power and organic fertilizer as a direct contribution for crop production (Ayele *et al*, 2002).

One of the reasons for low productivity of the livestock sector in Ethiopia is shortage of feed and low quality of available feeds, particularly in the dry seasons. Low adoption and promotion of cultivated forages (Tolera *et al*, 2019). Bracharia grass is one of the most important tropical grasses distributed throughout the tropics especially in Africa (Renvoize *et al.*, 1996). Bracharia plays important roles in soil erosion control and ecological restoration. Brachiaria species have been important component of sown pastures in humid low lands and savannas of tropical America with current estimated acreage of 99 million hectare in Brazil alone (Jank *et al.*, 2014).

*Bracharia* as a forage grass has been used in crop pasture intergraded systems where the grass seed is over sown on maize crop planted earlier favoring the production of high quality forage in the off season (Maia *et al.*, 2014). The accompanying advantages include reduced degradation of pastures, improved chemical, biological and physical properties of the soil and yield potential of grain forage and silage (Silva *et al.*, 2010). It has high biomass production potential and produces nutritious herbage thus increase livestock productivity (Holmann *et al.*, 2004). *Bracharia* is adapted to drought and low fertility soils, sequesters carbon through its large roots system, enhance nitrogen use efficiency and subsequently minimize eutrophication and greenhouse gas emissions (Subbarao *et al.*, 2009; Arango *et al.*, 2014; Moreta *et al.*, 2014; Rao *et al.*, 2014).

*Bracharia* grasses are productive warm-season perennial grasses with superior nutritive value to other warm-season grasses (Vendramini *et al.*, 2014), and can be used for grazing (Inyang *et al.*, 2010a) or harvested and conserved for feeding when needed (Vendramini *et al.*, 2010). Adaptation of *Bracharia* grass is improve feed and nutrition security, income and livelihood of small holder farmers in region through improved livestock productivity.

In East Guji zone, low access to improved forage grasses, poor extension services on livestock forages and feed scarcity are the major constraints in livestock production. The farmers are used crop residues, local grasses, and natural pasture to feed their livestock in the area. To improve availability of livestock feed in terms of quantity and quality, it is better to cultivate *Bracharia* grass forage that have better dry matter yield and nutritional quality. Therefore, this study was aimed to evaluate performance of *Bracharia* grass cultivars and select best adaptable, higher herbage yield among four cultivars in study area

## **2. Materials and Methods**

### **2.1. Description of the study area**

The experiment was carried out at Adola sub-site of Bore Agricultural Research Center, Adola district, Guji Zone of Oromia. Adola district is located around at a distance of 470 km from Addis Ababa and 120 Km from the zonal capital city, Negele Borena. It is an area where mixed farming and Sami- nomadic economic activity takes place, which is the major livelihood of the local people. The total area of the district is 1254.56 km<sup>2</sup>. The district is situated at 5°44'10" - 6°12'38" N latitudes and 38°45'10" - 39°12'37" E longitudes. The district is characterized by three agro- climatic zones, namely highland 11%, mid-land 29% and low-land 60% respectively. The major soil type of the district is tsois (red basaltic soils) and orthic Acrisols.

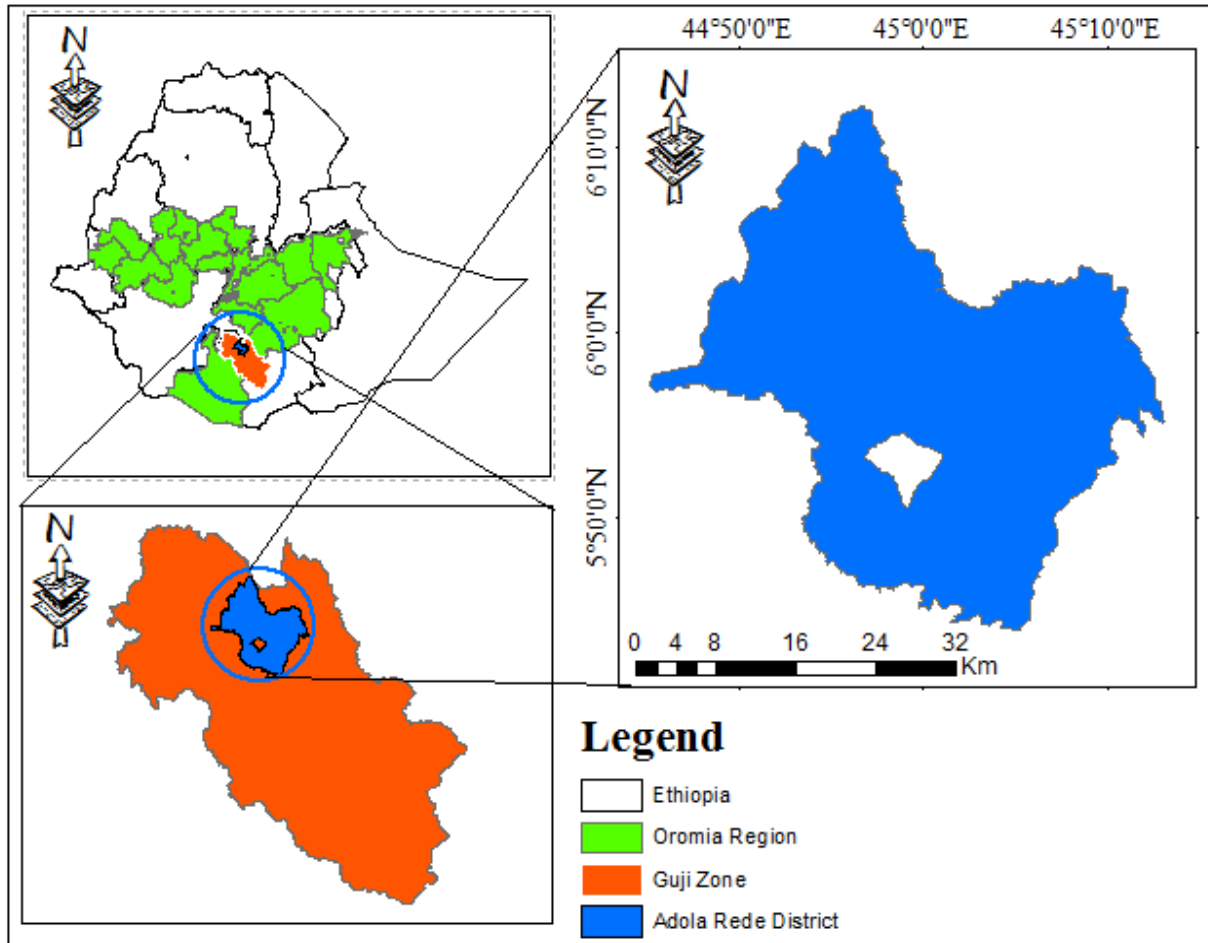


Figure 1. Map of study area., Source: Own computational GIS data.

## 2.2. Experimental treatments and design

The experiment was conducted at Bore Agricultural Research center during 2019 and 2020 cropping season. Four *Brachiaria* grass cultivars (*Brachiaria mutica* Dzf No 18659 (Dzf 483)), *Brachiaria decumbence* Dzf No 194, *Brachiaria mutica* 6964 Dzf No 484, and *Brachiaria mulato*) roots were brought from Ethiopian Institute of Agricultural Research, Debrezeite Agricultural Research Center (DZAC) and Oromia Agricultural Research, Mechara Agricultural Research Center (McARC) in randomized complete block design (RCBD) with three replications. The prepared experimental land was divided into three blocks which totally contain about 12 plots with each plot size area 7.5m<sup>2</sup>. The *Brachiaria* cultivar were spitted on plot size 2.5m length m x 3m width within space between rows and plants were 50 cm, 20cm and 1m between plots and replication respectively. Inorganic fertilizer of 100kg/ha of NPS and 50Kg/ha of urea were applied during the establishment.

## 2.3. Data collection

All data on morphological parameters and dry matter yield of forage were like plants height, fresh biomass, dry matter yield, leaf to steam ratio, survive rate, plot cover and vigor were recorded. Plant



survival rate was calculated as the ratio of the number of alive plants per plot to the total number of plants planted per plot and then multiplied by 100.

**Plant height:** Plant height was measured on the primary bud from the soil surface to the base of the top-most leaf using a meter designated by (Rayburn *et al.*, 2007). It was based on five plants randomly selected in each plot and measured using a steel tape from the ground level to the highest leaf. For determination of biomass yield, genotypes were cut at 5-10 cm from the ground level from two central rows.

**Dry matter yield (DMY):** After harvesting the middle four rows, the total biomass yield was determined using sensitive balance from each plot at each harvesting date. The dry matter yield (DMY) was determined at the end of every harvesting day. Total dry matter yields for each plot were calculated based on fresh biomass yield from the sample area of each plot. Thereafter it was converted to metric tons per hectare (Gelayenew *et al.*, 2019). The harvested fresh sample was measured right in field by sensitive weight balance and 300g subsample per plot was brought to Bore Agricultural Research Center and sampled sample was placed to oven dried for 72 hours at a temperature of 65c° for dry matter determination. Then dry matter yield (t/ha) was calculated by James *et al.*, 2008) formula.

$$\text{The dry matter yield (t/ha)} = \text{TFW} \times (\text{DWss} / \text{HA} \times \text{FWss}) \times 10$$

Where, TFW = total fresh weight kg/plot, DWss = dry weight of subsample in grams, FWss = fresh weight of subsample in grams, HA = Harvest plot area in square meters and 10 is a constant for conversion of yields in kg/m<sup>2</sup> to t/ha.

Leaf to stem ration, the morphological parts were separately weighed to know their sample fresh weight, oven dried for 72 hours at a temperature of 65oC and separately weighed to estimate the proportions of these morphological parts.

## 2.4. Statistical analysis.

All collected data were analyzed using the general linear model procedure of SAS (SAS 2002) version 9.1. Mean were separated using least significant difference (LSD) at 5% significant level. The statistical model for the analysis data was:

$$Y_{ijk} = \mu + A_j + B_i + e_{ijk}$$

Where;  $Y_{ijk}$  = response of variable under examination,  $\mu$  = overall mean,  $A_j$  = the  $j$ th factor effect of treatment/ cultivar,  $B_i$  = the  $i$ th factor effect of block/ replication and  $e_{ijk}$  = the random error

## Results & discussion

### 3.1. Performance of *Bracharia* grass genotypes

The performances of *Bracharia* grass cultivar were shown in Table 1. The result indicated that the tested cultivars were varied non-significantly ( $p > 0.05$ ) on survive rate percentages. The highest survive rate percentages were recorded from *Bracharia mutica* Dzf No 18659 (Dzf 483) (95.5%) followed by

Bracharia mulato (82.5%) cultivars. The lowest survive rate percentage was recorded from Brachiaria *Decumbens* Dzf No 194 (70.3%).

The result of plot cover was indicated highly significance difference ( $P < 0.01$ ) among Bracharia grass cultivars. This result indicated that the potential adaptability and productivity of cultivars were different. The rapidly and highly potential of plot cover were recorded from *Brachiaria mutica* 18659 Dzf No 483 (94.4%) and *Brachiaria mutica* 6964 Dzf No 484(87%) cultivars. This result was lower than the findings of Birmaduma Gadisa *et al.*(2020) who report the plot cover of 96-98%. for *Brachiaria gras*. This result is good indication for adaptability Bracharia grass with soil, water and environment of study area (Clara M (2013)) reported that ground cover is an important attribute of any vegetation, especially in relation to soil and water conservation support this study. It is also an important parameter in restoration of degraded areas, where moisture is the main limiting factor. The mean average of plants height at study area were show highly significant ( $P < 0.01$ ) different between treatments.

#### ***Plant height (cm)***

The analysis of variance for plant height in this study indicated highly significant difference ( $p < 0.01$ ) among cultivars. The highest plant height was recorded from Brachiaria Mutica 6964 Dzf No 484 (170cm) cultivar whereas, the lowest plant height was recorded from Brachiaria Decumbence Dzf No 194 (90cm) cultivars. This result is in agreement with Clara M (2013) who reported a 91 cm height for Bracharia hybrid (Mulat II) in Kenya and is higher than 50 -70 cm height reported in Northern Ethiopian (Wassie *et al.*, 2017).

#### ***Leaf to stem ratio***

The analysis of variance for leaf to stem ration in this study was not indicated statistically significance difference ( $p > 0.05$ ) among cultivars. However, the least square mean values of leaf to stem ratio was indicated numerically difference among cultivars. The highest leaf to stem ratio was recorded from *Brachiaria mutica* 18659 Dzf No 483 (0.82) cultivar and the lowest value was recorded from Brachiaria Mutica 6964 Dzf No 484 (0.4). However, this result is in agreement with the findings of Aldava *et al.*(2017) who reported the leaf to stem ratio of 0.4 for *Brachiaria brizantha* in Mexico.

#### ***Fresh biomass yield (t/ha)***

The fresh biomass yield (t/ha) result among cultivars were shown statistically highly significance difference ( $p < 0.01$ ). The highest fresh biomass yield value were recorded from *Brachiaria mutica* 18659 Dzf No 483 (33.52t/ha) which is followed by *Brachiaria mutica* 6964 Dzf No 484 (33.6 t/ha) cultivar. This finding was not comparable with the value of 45.8 t/ha reported for Bracharia mulato in West Hararghe Zone, Eastern Oromia, Ethiopia (Birmaduma Gadisa *et al.*, 2020). This amount of fresh biomass production indicates the potential of the genotype to be adaptable, drought tolerant and unsusceptible for diseases.

Table 1. Over all location mean agronomic performance of *Brachiaria* grass cultivars in Midland agro-ecologies of areas.

cultivars	SR%	Pc%	Vg%	FBM (t/ha)	LSR	PH (cm)	DMY t/ha
<i>Brachiaria mutica</i> Dzf No 18659 (Dzf No 483)	95.5	94.4 <sup>a</sup>	92.5	33.52 <sup>a</sup>	0.82	160 <sup>a</sup>	11.82 <sup>a</sup>
<i>Brachiaria Mutica</i> 6964 (Dzf No 484)	74.7	87 <sup>a</sup>	88.8	33.6 <sup>a</sup>	0.4	170 <sup>a</sup>	11.95 <sup>a</sup>
<i>Bracharia mulato</i>	82.5	62.9 <sup>b</sup>	57.1	28.13 <sup>b</sup>	0.53	92 <sup>b</sup>	10 <sup>a</sup>
<i>Brachiaria Decumbence</i> Dzf No 194	70.3	48.1 <sup>b</sup>	70.3	25.85 <sup>b</sup>	0.74	90 <sup>b</sup>	6.33 <sup>b</sup>
Mean	80.8	73.1	77.2	30.28	0.62	128.2	10.02
CV	14.8	12.3	31.9	6.6	43.8	12.7	12.2
LSD (5%)	ns	**	ns	**	ns	**	**

<sup>a,b,c</sup> Mean in a column within the same category having different superscripts differ significantly ( $p < 0.05$ )

PH (cm)=plant height in centimeter, Pc%=plot cover percentage, LSR=leaf to stem ratio, Vg%=vigor percentage, SR%=survive rate percentage, FBM t/ha= Fresh biomass tone per hectare, DMY t/ha =dry matter yield tone per hectare, CV=Coefficient of variation, LSD= Least significant difference, \*\*= highly significant, ns= None significant different.

### Dry matter yield (t/ha)

The dry matter yield (t/ha) result among cultivars were shown statistically highly significance difference ( $p < 0.01$ ). The highest dry matter yield value were recorded from *Brachiaria mutica* 6964 Dzf No 484 (11.95t/ha) followed by *Brachiaria mutica* 18659 Dzf No 483 (11.82t/ha) cultivars. This result is lower than the findings of Wassie *et al.*, (2017) who reported 37.75 t/ha from Eth. 13809 *Bracharia* cultivars in Northern Ethiopia and also lower than the result of compared with the 16.3 t/ha DM obtained by Hare *et al.*, (2007). Variations in dry matter yield production across the cultivars can be attributed to the differences in growth rate and growth habit, which are mediated through the genotypic and phenotypic differences.

### Conclusions and recommendations

The result implies that *Brachiaria mutica* Dzf No 18659 (Dzf 483) and *Brachiaria mutica* 6964 (Dzf No 484) were well adapted and being productive regarding the plant height, biomass yield and Dry matter yield which, is hopeful to fill the gap of low quality and quantity ruminant feed supply of the community.

The current study indicated that *Brachiaria mutica* Dzf No 18659 (Dzf 483) and *Brachiaria mutica* 6964 (Dzf No 484) cultivars were good agronomic performance at study area. Therefore, based on its adaptability, plant height, biomass yield and dry matter yield and survive rate *Brachiaria mutica* Dzf No 18659 (Dzf 483) and *Brachiaria mutica* 6964 (Dzf No 484) is recommended for further promotion in the midland of Guji and similar agro-ecologies.

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## Evaluation of Napier grass (*Pennisetum purpureum* L) Accessions for Agronomic and Biomass yield performance at Bore and Goro-dola Districts, Guji Zone, Oromia

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### Abstract

The study was conducted at Bore and Goro-dola districts representing the highland and lowland agro-ecologies of Guji zone respectively. The objective of the study was to identify adaptable and high biomass yield of Napier grass accessions for the study area. Accordingly, six Napier grass accessions (ILRI-16791, ILRI-16798, ILRI-16840, ILRI-16800, 16819, ILRI-15743) were arranged in randomized complete block design (RCBD) with three replications. All agronomic parameters and biomass yield of forage samples were collected and analyzed. The result from the trial conducted at Bore site revealed that plot cover, plant vigor, survival rate, dry matter yield were significantly ( $P < 0.05$ ) differ among the treatments. However, number of tiller per plant, plant height and leaf to stem ratio were not showed significantly difference ( $P > 0.05$ ) among the tested Napier grass accessions. The highest DM yield was recorded from ILRI-16791 (10.7 t/ha) followed by ILRI-16819 (8.7t/ha) Napier grass accession. The highest survival rate (62.2%) was recorded for accession ILRI-16819 followed by accession ILRI-16791 (48.8%). At Goro-dola site, the result indicated that survive rate ( $P < 0.01$ ), number of tiller per plant ( $P < 0.01$ ), and dry matter yield ( $P < 0.05$ ) were significantly differ among the treatments. The highest survival rate (88.8%) was recorded from ILRI-16791 followed by ILRL 16819 (75.5%). On the other hand the maximum value (12.11 t/ha) of DM yield was obtained from ILRI-16791 accession followed by ILRL-16840 (10.0t/ha) and ILRL-16819 (9.17 t/ha). Hence, ILRL-16819 and ILRI-16791 accessions performed well in most of the agronomic and biomass yield parameters at all sites. Thus it could be possible to conclude that these Napier grass accessions ILRI-16791 and 16819 should be recommended for improving the constraint of feed shortage in high and lowland agro-ecologies of Guji zone and similar areas.

**Key words:** Agronomic performance, Accessions, Biomass yield, Napier grass, *Pennisetum purpureum*,

### Introduction

Livestock production is an integral part of the farming systems in all parts of Ethiopia. The sector has a share of 12-16% of the total Gross Domestic Product (GDP) and 30-35% of agricultural GDP (Ayele *et al.*, 2002). It contributes to the livelihoods of 60-70% of the Ethiopian population. Moreover, it ensures the availability of food, creates jobs, transportation and income to the farming community, serve as a source of agricultural inputs such as draft power and organic fertilizer as a direct contribution for crop production (Ayele *et al.*, 2002).

Napier grass (*Pennisetum purpureum* (L.) Schumach) also known as Elephant grass is originated from sub-Saharan tropical Africa (Clayton *et al.*, 2013). It is a tall and deep-rooted perennial bunch grass well known for its high yielding capability and mainly used for cut-and-carry feeding systems (FAO, 2015) and fed in stalls, or it is made into silage or hay. It performs well in low, mid and highland areas of

Ethiopia (Seyoum *et al.*, 1998; Tessema, 2005). Napier grass can adapt to a wide range of soil types from sandy to clayey. It can also grow in soils in the pH range of highly acidic to alkaline (Centre for New Crops and Plant Products, 2002). It grows best at high temperatures but can tolerate low air temperatures under which the yield can be reduced and ceases to grow at a temperature below 10°C (Fekede *et al.*, 2005). The herbage can be killed by light frosts but the underground parts remain alive unless the soil is frozen and growth resumes rapidly when conditions become ideal (Fekede *et al.*, 2005). It is propagated vegetative by using stem cuttings, root splits or shoot tips which usually vary across agro-ecologies (Getnet and Gezahagn, 2012).

Napier grass can provide a continual supply of green forage throughout the year and best fits in all intensive small scale farming systems (Alemayehu, 1997). It is a fast growing and has a high annual productivity that depends on the climatic conditions, especially of temperature and rainfall and it can produce biomass yield of 20-30 t DM/ha/year with good agronomic and management practices (Farrell *et al.*, 2002). Napier grasses are principally used for cut-and carry fodder for animals and used as forage for Livestock to provide a continual supply of green forage throughout the year and it fits intensive small scale farming (Alemayehu Mengistu 1997). It is also sometimes cut for hay and to ferment into silage for dry season feeding, good nutritive value and mostly used for cut and carry system over the tropical and sub-tropical area of the world (Cook *et al.*, 2005; Duke, 1983; Wadi *et al.*, 2004).

To improve availability of livestock feed in terms of quantity and quality, it is better to cultivate Napier grass forage that have better biomass yield and nutritional quality. Therefore, the objective of the present study was to evaluate the adaptability potential good biomass yield, herbage dry matter yield of Napier grass accessions grown at high and lowland agro-ecology of Guji zone.

## **Materials and Methods**

### ***Description of the Study Area***

The study was conducted at Bore and Goro-dola districts representing respectively the highland and lowland agro-ecologies of Guji zone. Bore district is located at 385 km to the south Oromia from Finfinne and 220 km from the Guji Zone capital city (Negele) with geographical location of 557'23" to 626'52" N latitudes and 3825'51" to 3856'21" E longitudes, South-eastern Oromia. The annual rain fall is about 1400-1800mm and the annual temperatures of the district ranged from 10.1 to 20 C. The major soil types of the site is mostly black soil. Bore Agricultural Research station is located at 7 km from Bore town which is geographically located at 624'37" N latitude and 3834'76" E longitudes. Bore research site is situated at an altitude of 2736 m.a.s.l. receiving high rainfall characterized by bimodal distribution. The first rainy season extends from April to October and the second season starts late November and ends at the beginning of March.

Goro-dola district is located at distance of 565 km from Addis Ababa and 30 km from the Zonal capital city, Negele Borena. The district is situated at which is located between located between 4°56'15" - 5°48'00" northing latitudes and 39°43'00" - 39°6'30" easting longitudes, with an altitude of 1100 m.a.s.l. receiving high rainfall characterized by bimodal distribution. The annually temperature of the district range from 25c<sup>0</sup> and 27c<sup>0</sup> and annual rainfall depth varies in the range from 500 mm to 700 mm with mean value of 600 mm by two type of rainy season, namely season summer (locally name "Gana") which starts in early march up to June, and autumn (locally name" Hageya"), which starts late September up to reaches beginning of November. The total area of

the district is 2491.570 km<sup>2</sup>. The Goro-dola peasants association under study area where mixed farming economic activities take place, which is the major livelihood of the people and communities of the district depend up on pastoralist and some of them are agro pastoralist. The major soils of the district are Nitosols (red basaltic soils) and Orthc Acrosols. The two soils are found on the highland areas, and they are red brown and black brown in colors and on sloping terrain and their utilization are good under natural vegetation respectively. And also the major soil types of district sand, clay most of the times these soils expose for erosion; therefore it has little agricultural potentials (SAC profile of Goro dola district office, 2003)

### ***Experimental Treatments and Design***

A total of six Napier grass accessions; ILRI-16840, ILRI-16819, ILRI-16800, ILRI-16791, ILRI-15743 and ILRI-16798 were used for the experiment. Napier grass accessions were planted at the beginning of the main rainy season on 5 × 2.5 m plot using RCBD with three replications. The root splits were planted in rows with five rows per plot. A total of 25 root splits were planted per plot with the intra and inter row spacing of 0.5 m and 1 m respectively, giving a density of 20,000 plants/ha. There were 1.5 m width between blocks and 1m width between plots. The propagation of Napier grass is by root splits and stem cutting with three nodes planting upright by burying two basal nodes into soil. Nitrogen phosphate sulfate (NPS) at the rate of 100 kg/ha was uniformly applied to all plots. After every harvest, the plots were top dressed with 50 kg Urea/ha of which one-third applied at the first shower of rain and the remaining two third applied during the active growth stage of the plant. Other agronomic management practices was applied uniformly as per recommendation.

### ***Data Collection***

All agronomic and yield data including plant survival rate, plot coverage, Vigor, number of tillers per plant, plant height, leaf to stem ratio, forage DM yield were collected. Plant survival rate was calculated as the ratio of the number of alive plants per plot to the total number of plants planted per plot and then multiplied by 100. Plant height was based on five plants was randomly selected in each plot, measured using a steel tape from the ground level to the highest leaf. For determination of biomass yield, accessions were cutting at 5-10cm from the ground level from two central rows. In order to measure dry matter yield, the harvested fresh sample was measured right in field by sensitive weight balance and 300g subsample per plot was brought to Bore Agricultural Research Center and sampled sample was placed to oven dried for 72 hours at a temperature of 65c° for dry matter determination. Then dry matter yield (t/ha) was calculated by Mutegi *et al.*, (2008) formula.

$$\text{The dry matter yield (t/ha)} = \text{TFW} \times (\text{DWss} / \text{HA} \times \text{FWss}) \times 10$$

Where, TFW = total fresh weight kg/plot, DWss = dry weight of subsample in grams, FWss = fresh weight of subsample in grams, HA = Harvest plot area in square meters and 10 is a constant for conversion of yields in kg/m to t/ha.

The leaf to stem ratio was calculated by weighing the leaf and stem parts after oven dried for 72 hours at a temperature of 65°C.



## ***Statistical Analysis***

All collected data were analyzed using the general linear model procedure of SAS (SAS 2002) version 9.1. The statistical model for the analysis data was:

$$Y_{ijk} = \mu + A_j + B_i + e_{ijk}$$

Where;  $Y_{ijk}$  = response of variable under examination,  $\mu$  = overall mean,  $A_j$  = the  $j$ th factor effect of treatment/ cultivar,  $B_i$  = the  $i$ th factor effect of block/ replication,  $e_{ijk}$  = the random error.

## **Result and Discussion**

### ***Performances of Napier grass accessions at Bore (Highland) site***

The analysis result of agronomic and biomass yield of Napier grass accessions are presented in Table 1. There were a significant ( $P < 0.05$ ) variation in plot cover and survival rate among Napier grass accessions tested in the study area. The highest plot cover (89.5%) was recorded for accession ILRI-16791 while the least plot cover value (68.5%) was recorded from ILRI-16840 accession. Among the tested accessions, the highest plant survival rate (62.2%) was recorded for accession ILRI-16819 followed by accession ILRI-16791 (48.8%). On the other hand, accession ILRI-16840 showed the lowest (17.8%) plant survival rate. The better survival rate of Napier grass could be due to the suitability of the environment with regard to soil moisture and fertility condition of the study area. Other scholars also reported similar findings (Skerman and Riveros, 1990). The highest number of tillers is (104.6) over years was obtained from genotype ILRI-16791 followed by accession ILRI-16798 (68.1) while accession ILRI-16840 gave the lowest (46.6). The difference in tillers produced per plant among the genotype of Napier grass could be attributed to genetic variations among the accessions and their interactions to the environment. According to Tessema and Alemayehu (2010), Napier grass produces many tillers and dense vegetative growth as the pasture consolidates due to perennial nature of the grass. The number of tillers per plant of Napier grass increased with plant height at cutting (Tessema *et al.*, 2003).

The result showed that plant height was not significantly different ( $P > 0.05$ ) among the tested Napier grass accessions. Numerically, the highest plant height value were recorded from ILRI-16891 and ILRI-16798 (180 cm) followed by ILRI-15743 (178.3 cm). While the lowest plant height was recorded from ILRI-16840 (160.7 cm). The dry matter (DM) yield of Napier grass accessions showed significant ( $P < 0.05$ ) variation among the tested accessions. (Table 1). The DM yield value ranged from 10.6 to 5.7 t/ha with a mean of 7.8 t/ha. Accession ILRI-16791 gave the highest mean DM yield (10.7 t/ha) followed by 16819 (8.7 t/ha). On the other hand, the lowest DM yield was obtain from 16840 (5.7 t/ha). This result is lower than result reported by Deribe *et al*, 2017 (12.6). The variations in plant survival rate, tillering performance and plant height are the causes of difference in DM yield. According to Tessema (2005) and Ishii *et al.* (2005), the taller varieties showed higher DM yields than the shorter varieties. The analysis result for leaf to stem ratio indicated that there was not significance difference ( $P > 0.05$ ) among the treatments. The recorded values of leaf to stem ratio range from 0.4-0.68. This result different from range of leaf to stem ratio reported by Deribe *et al*, 2017 who reported 0.31 to 1.01 and Elkana *et al.* 2010 reported that 1.7 to 3.1.

Table 1. Mean value of agronomic and yield component of Napier grass at Bore (highland) site of Guji zone

Accessions	PC (%)	Vg (%)	SR (%)	NTPP	PH (cm)	LSR	DMY(t/ha)
ILRL-16840	68.5 <sup>b</sup>	62.9 <sup>b</sup>	17.8 <sup>c</sup>	46.6	160.7	0.4	5.7 <sup>c</sup>
ILRL-16791	89.5 <sup>a</sup>	83.3 <sup>a</sup>	48.8 <sup>ab</sup>	104.6	180	0.61	10.6 <sup>a</sup>
ILRL-16819	83.3 <sup>ab</sup>	77.7 <sup>ab</sup>	62.2 <sup>a</sup>	61.6	161.7	0.61	8.7 <sup>ab</sup>
ILRL-16800	82.7 <sup>ab</sup>	78.4 <sup>ab</sup>	24.4 <sup>c</sup>	50.67	175	0.6	8.4 <sup>ab</sup>
ILRL-16798	85.7 <sup>ab</sup>	81.4 <sup>ab</sup>	33.3 <sup>bc</sup>	68.1	180	0.68	6.9 <sup>bc</sup>
ILRL-15743	87.8 <sup>ab</sup>	83.3 <sup>a</sup>	33.3 <sup>bc</sup>	49.6	178.3	0.615	6.8 <sup>bc</sup>
Mean	83	72.9	36.7	63.6	172.6	0.6	7.8
CV	12.1	12.6	29.5	53.1	10.7	26.9	16.3
Sig.level	*	*	**	Ns	ns	ns	**

<sup>a,b,c</sup> Mean in a column within the same category having different superscripts differ significantly.

PH =plant height PC=Plot cover, LSR=leaf to steam ratio, Vg=vigor, SR=survive rate, NTPP=number of tiller per plants, DMY =Dry matter yield, CV=Coefficient of variation, \*\*= highly significant, \*=significant and ns= Non significant different

#### ***Performances of Napier grass accessions at Goro-dola (Lowland) site***

The agronomic and biomass yield performances of Napier grass accessions were shown in Table 2. The result indicated that the tested accessions were significantly differ on survive rate ( $p < 0.01$ ), number of tiller per plant ( $p < 0.01$ ) and dry matter yield. ( $p < 0.05$ ) The highest survive rate were recorded from *ILRL-19791* (88.8%) followed by *ILRL 16819* (75.5%) accession while the lowest survive rate percentage was recorded from *ILRL 16798* (62.8%). This variation is might be due to the differences in soil types and moisture with environmental factors. The mean average of plants height at study area were not significantly ( $P > 0.05$ ) differ among treatments. Numerically, the highest plant height was recorded from *ILRL-16819* (133.5 cm) accession whereas, the lowest plant height was recorded from *ILRL-16800* (126.2cm) accession. This result agreed with result reported by Mamaru, (2018).

The number of tiller per plant among the tested accessions were shown highly significance difference ( $p < 0.01$ ). The highest number of tiller per plant value recorded from *ILRL-16819* (19.6) followed by *ILRI-16791* (14.5) accession. The lowest number of tiller per plant was recorded from *ILRL-16800* (7.4) accession. This finding is in line with the result reported by values by Mamaru (2018) which ranged from 9-12 tillers per plant.

The analysis result for dry matter yield indicated that there was a significant difference ( $p < 0.05$ ) among the tested Napier grass accessions. The highest dry matter yield value were recorded from *ILRL-16791* (12.11t/ha) followed by *ILRL-16840* (10 t/ha) accessions. The lowest dry matter yield was recorded from *ILRL-16798* (7.7 t/ha). This result is lower than the other findings of which 41.05 t/ha was reported by (Ansay *et al.*, 2010). Similarly, Gemiyo *et al.*, (2017) reported biomass yield of 12-18 t/ha under rain fed condition. Variations in dry matter yield production across the accessions can be attributed to differences in growth rate and might be due to the proportional increment of dry matter yield with advance in age of cutting (Taye *et al.*, 2007).

Table 2. Mean agronomic and biomass yield performance of Napier grass accessions at Goro-dola (lowland) site of Guji zone

Accessions	SR (%)	PC (%)	Vig (%)	LSR	PH (cm)	NTPP	DMY (t/ha)
ILRL-16840	73.3 <sup>ab</sup>	86.9	87.9	0.67	131.9	14.4 <sup>abc</sup>	10.0 <sup>ab</sup>
ILRL-16791	88.8 <sup>a</sup>	85.1	86.9	0.72	126.6	14.5 <sup>ab</sup>	12.11 <sup>a</sup>
ILRL-16819	75.5 <sup>ab</sup>	87.8	86.05	0.66	133.5	19.6 <sup>a</sup>	9.17 <sup>ab</sup>
ILRL-16800	71 <sup>b</sup>	77.7	84.6	0.71	126.2	7.4 <sup>c</sup>	8.17 <sup>b</sup>
ILRL-16798	62.8 <sup>b</sup>	82.3	83.7	0.74	129	10.4 <sup>bc</sup>	7.7 <sup>b</sup>
ILRL-15743	68.8 <sup>b</sup>	91.6	93.4	0.71	128.2	11b <sup>c</sup>	8.39 <sup>b</sup>
<b>Mean</b>	<b>73.3</b>	<b>85.3</b>	<b>87.1</b>	<b>0.705</b>	<b>129.2</b>	<b>12.0</b>	<b>9.24</b>
<b>CV</b>	<b>12.4</b>	<b>13.6</b>	<b>12.6</b>	<b>32.8</b>	<b>19</b>	<b>29.3</b>	<b>28.4</b>
<b>Sig. level</b>	<b>**</b>	<b>Ns</b>	<b>ns</b>	<b>Ns</b>	<b>ns</b>	<b>**</b>	<b>*</b>

<sup>a,b,c</sup> Mean in a column within the same category having different superscripts differ significantly ( $p < 0.05$ ) PH = plant height PC=Plot cover, LSR=leaf to stem ratio, Vg=vigor, SR=survive rate, NTPP=number of tiller per plants, DMY =Dry matter yield, CV=Coefficient of variation, \*\*= highly significant, \*=significant and ns= None significant different

### Conclusion and Recommendations

The result indicated that most of agronomic parameters including plot cover, plant vigor, survival rate, dry matter yield were significantly ( $P < 0.05$ ) differ among the treatments at Bore site. The highest DM yield and survival rate were recorded from ILRI-16791 followed by ILRI-16819 Napier grass accession. At Goro-dola site, the result indicated that survive rate, number of tiller per plant ( $P < 0.01$ ), and dry matter yield ( $P < 0.05$ ) were significantly differ among the treatments. The highest survival rate was recorded from ILRI-16791 followed by ILRL 16819. Napier grass accessions ILRI-16791, ILRL-16840 and ILRL-16819 were among the best performance with regards to biomass yield at the lowland site. Hence, based on their performances such as survive rate, and dry matter yield, ILRL-16819 and ILRI-16791 Napier grass accessions are recommended for further promotion in the high and lowland agro-ecologies of Guji zone and similar areas.

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# Evaluation of Rhodes Grass (*Chloris gayana*) Cultivars in highland and midland areas of Guji Zone southern Oromia, Ethiopia.

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## Abstract

The study was conducted with the objectives to identify and select better adaptable, higher herbage yielding forage variety. Five Rhodes grass cultivars (Masaba, ILRI 6633, ILRL 7384, ILRL 13325 and Dz-253) were arranged in randomized complete block design (RCBD) with three replications. All agronomic parameters and biomass yield of forage samples were determined and collected data were examined using statistical analysis. The results indicated that dry matter yield was showed statistically significant variation ( $P < 0.05$ ) among Rhodes grass cultivars at tropical agro-ecologies and the results indicated that seed yield (qu/ha) was showed statistically significant variation ( $P < 0.05$ ) among the treatments at both agro-ecologies. The highest herbage dry matter yield and seed yield were recorded from ILRI 7384 (15.44 t/ha) followed ILRI 6633 (14.9 t/ha) Rhodes grass cultivar at tropical area and the mean values the highest seed yield at temperate agro-ecologies were recorded from ILRI 6633 (1 qu/ha) and Masaba (0.83 qu/ha) Rhodes grass cultivars. These cultivars are well adapted and suitable for use as animal feeds under the study areas. As a result, these three Rhodes grass ILRI 6633, ILRI 7384 and Masaba cultivars were recommended for livestock producers as feed resources to enhance animal production and productivity in the study areas and other with similar agro-ecologies.

**Key words:** *Rhodes Grass, cultivars, highland, and midland areas*

## Introduction

Rhodes grass (*Chloris gayana*) is now widespread in tropical and subtropical areas worldwide. Rhodes grass (*Chloris gayana*) is a perennial or annual tropical leafy grass 1-2 m in height, highly variable in habit. The culms are tufted or creeping, erect or decumbent, sometimes rooting from the nodes. The inflorescences are light greenish brown (rarely yellow) in color and turn darker brown as they mature (Cook *et al.*, 2005).

Rhodes grass thrives in places where annual temperatures range 25 to 30°C (day/night temperature). Optimal annual rainfall ranges between 600-750mm with a summer-rainfall period (Moore, 2006; Ecocrop, 2014). Rhodes grass grows better in areas where with an altitude ranges from 1400 – 2400 m.a.s.l (Ecocrop, 2014). Due to its deep roots, Rhodes grass can withstand long dry periods (over 6 months) and up to 15 days of flooding (Cook *et al.*, 2005; FAO, 2014).

Rhodes grass grows on a wide range of soils from poor sandy soils to heavy clayey alkaline and saline soils. Rhodes grass grows better on fertile, well-structured soils and it prefers soil pH between 5.5 and 7.5; even if, establishment on acidic soils is challenging. Rhodes grass survives on infertile soils although it is unproductive and may eventually die out particularly if grazed regularly. Rhodes grass is a full sunlight species, which does not grow well under shady environments (FAO, 2014; Eco crop, 2014).

Growth performance of Rhodes grass varies with type of cultivar, age of plant and other environmental factors (FAO, 2009; Akinola *et al.*, 1990).

Rhodes grass productivity generally ranges from 7 - 25 tons of DM/ha per year, depending on variety, soil fertility, environmental conditions and cutting frequency. Based on a study conducted on farmers' fields in the central highlands of Ethiopia, on average the herbage yield of Rhodes grass was from 8.74 to 9.1 tons DM/ha per year on rain-fed conditions (Cook *et al.*, 2005; CASCAPE, 2015; HARC, 2004).

To overcome this bottleneck problem of livestock sector introducing improved forages to livestock stakeholders should have to be an obligatory and persistent activity that is expected from responsible service providers. Therefore, the objectives of the study were to select/recommend adaptable and high biomass yielding Rhodes grass cultivars for the study area and other areas having similar agro-ecologies.

## **2. Materials and Methods**

### ***2.1. Description of the study area***

The experiment was carried out at Bore Agricultural Research Center, which is one of the recently established Research Centers of the Oromia Agricultural Research Institute (OARI) in Bore district, Guji Zone, Southern Oromia. Bore district is located at 385 km to the south Oromia from Finfinne and 220 km from the Guji Zone capital city (Negele) with geographical location of 557'23" to 626'52" N latitudes and 3825'51" to 3856'21" E longitudes, South-eastern Oromia. The annual rain fall is about 1400-1800mm and the annual temperatures of the district ranged from 10.1 to 20 C. The major soil types of the site is mostly black soil. Bore Agricultural Research station is located at 7 km from Bore district which is geographically located at 624'37" N latitude and 3834'76" E longitudes. The research site represents highlands of Guji Zone with an altitude of 2736 m.a.s.l. receiving high rainfall characterized by bimodal distribution. The first rainy season extends from April to October and the second season starts late November and ends at the beginning of March.

Adola District, where the Midland sub-site of Bore Agricultural Research Center is found, is located at a distance of 470 km from Addis Ababa and 120 km from the Zonal capital city, Negele Borena. It is an area where a mixed farming and semi-nomadic economic activities are undertaken as the major livelihood of the local people. The total area of the District is 1254.56 km<sup>2</sup>. The District is situated at 5°44'10" - 6°12'38" N Latitudes and 38°45'10" - 39°12'37" E Longitudes. The District is characterized by three agro-climatic zone, namely highland (11%), midland (29%) and lowland (60%). The major soil types of the district are nit sols (red basaltic soils) and orthicAcrosols (Yazachew E. and Kasahun D.2011).

### ***2.2. Experimental treatments and design***

The experiment was conducted using five Rhodes grass varieties Masaba, DZ-253, ILRI 6633 and ILRI 13325. The experiment was conducted in randomized complete block design (RCBD) with three replications. Seed yield were sowing in rows spaced 20cm between rows and 1m, 1.5m between plot and block respectively on plot size of 2m x 1m (2 m<sup>2</sup>). Fertilizer was applied and other agronomic crop protection practices was adopted uniformly as per recommendation for production.

### **3.3. Data Collection.**

All relevant data like days to emergence, Days to flowering, plant heights, and leaf to stem ratio, fresh biomass and dry matter yield value were recorded. Plant height was based on five plants was randomly selected in each plot, measured using a steel tape from the ground level to the highest leaf. For determination of biomass yield, genotypes were cutting at 5-10cm from the ground level from two central rows.

In order to measure dry matter yield, the harvested fresh sample was measured right in field by sensitive weight balance and 300g subsample per plot was brought to Bore Agricultural Research Center and sampled sample was placed to oven dried for 72 hours at a temperature of 65c° for dry matter determination. Then dry matter yield (t/ha) was calculated by James (2008) formula.

$$\text{The dry matter yield (t/ha)} = \text{TFW} \times (\text{DWss} / \text{HA} \times \text{FWss}) \times 10$$

Where TFW = total fresh weight kg/plot, DWss = dry weight of subsample in grams, FWss = fresh weight of subsample in grams, HA = Harvest plot area in square meters and 10 is a constant for conversion of yields in kg/m to t/ha.

Leaf to stem ration, the morphological parts were separately weighed to know their sample fresh weight, oven dried for 72 hours at a temperature of 65oC and separately weighed to estimate the proportions of these morphological parts.

### **Statistical analysis**

All collected data were analyzed using the general linear model procedure of SAS (SAS 2002) version 9.1. Mean were separated using least significant difference (LSD) at 5% significant level. The statistical model for the analysis data was:

$$Y_{ijk} = \mu + A_j + B_i + e_{ijk}$$

Where;  $Y_{ijk}$  = response of variable under examination,  $\mu$  = overall mean,  $A_j$  = the jth factor effect of treatment/ cultivar,  $B_i$  = the ith factor effect of block/ replication,  $e_{ijk}$  = the random error.

### **Result and Discussion**

#### **Land preparation**

The land should be cleared from weeds and trees before ploughing. The land should be ploughed two to three times to get a fine and leveled seedbed. As the Rhodes grass seed is very small it needs a well-prepared seedbed Fekede F (2000). The seedbed should be ploughed and prepared well. A well-prepared seedbed favours seed germination, seedling emergence and growth (CASCAPE, 2015).



### ***Planting time***

Planting should be done at the start of the main rainy season when the soil has received sufficient moisture to support germination and establishment. Sowing too deeply will result in failure or poor germination. Rhodes grass seed will germinate under slightly cooler conditions than most summer growing grasses [21].

### ***Seed planting methods***

Rhodes grass can be established vegetatively (root splits) or from seed. Seed rate varies depending on seed quality (germination and purity), sowing method, environmental conditions and land preparation. Generally, the seed rate should be 15 kg/ha considering the previous factors. Seed should be sown on the surface no deeper than 2cm (Cook et al., 2005). Rhodes grass can be row sown or broadcasted and with spacing between rows should be 20cm HARC (2004).

### ***Days to 50 % emergence***

The over year statistical data analysis result indicates that days to emergence is a significant difference ( $P < 0.05$ ) among the cultivars at both agro-ecologies. This study was Rhodes grass cultivars required a range of 8.33 – 10.4 days for first emergence which is almost similar with the study conducted at Deghabour district of Ethiopian Somalia region (Mohamed and Gebeyew, 2018) which was recorded 10.66 days. According to (Cook *et al.*, 2005) studied Rhodes grass germinates 7 days after planting. This difference might be attributed to soil moisture content, soil fertility and other environmental factors. ...

### ***Days to 50% flowering***

The analysis result of days to 50% flowering indicate that highly significant ( $P < 0.01$ ) among the cultivars at midland agro-ecologies. The shortest days to 50% flowering was recorded from ILRI 6633 cultivar (72.67). The longest days to 50% flowering was recorded from Masaba cultivars (82) at midland agro-ecologies. This difference in days to 50% flowering among treatments could be attributed to genetic variation among genotypes and their interaction with the environment.

### ***Plant height (cm)***

The analysis result of plant height indicate that non-significant ( $P > 0.05$ ) among the cultivars at both agro-ecologies. The mean highest plant height (PH) values of Rhodes grass (*Chloris gayana*) cultivars was (181.7 cm) obtain from DZ-253 cultivars. The average plant height values of Rhodes grass cultivars recorded in this study was almost different with the study conducted at Deghabour district of Ethiopian Somalia region (Mohamed and Gebeyew, 2018) which was 139.10cm and study conducted by Yesihak (2008) revealed that height of Rhodes plants grown sole on savannah regions of Ethiopia at 8 weeks after sowing varied from 100.7 to 121.0cm tall.

### Leaf to stem ratio

The analysis result of leaf to stem ratio indicate that non-significant ( $P>0.05$ ) among the cultivars at both agro-ecologies. The highest leaf to stem ratio value of Rhodes grass cultivar were (0.83) obtain from Masaba and ILRI 6633 cultivars at midland agro-ecologies.

Table 1. The mean value of yield and yield components of Rhodes grass cultivars over years and over locations at highland agro-ecologies.

Varieties	DE	DF	LSR	DMY t/ha	FBM t/ha	PH (cm)	SYI qt/ha
7384	20 <sup>b</sup>	130	0.75	7.9	27 <sup>ab</sup>	108.3	0.68 <sup>ab</sup>
Masaba	22 <sup>ab</sup>	128.7	0.83	9.6	25.83 <sup>ab</sup>	109	0.66 <sup>ab</sup>
13325	22.33 <sup>ab</sup>	129.3	0.6	8.4	28.8 <sup>ab</sup>	101.2	0.83 <sup>ab</sup>
DZ-253	23.33 <sup>a</sup>	128.3	0.7	10.2	23.67 <sup>b</sup>	97.1	0.5 <sup>b</sup>
6633	20.33 <sup>ab</sup>	132.7	0.83	8.6	31 <sup>a</sup>	113.2	1 <sup>a</sup>
Mean	21.6	129.8	0.74	8.94	27.2	106.8	0.74
CV	7.1	2.1	28.7	26.2	10.1	8.8	30
LSD (5%)	*	ns	ns	ns	*	ns	*

DE=days to emergence, DF=days to flower, DM=days to maturity, LSR=leaf to stem ratio, DMY=dry matter yield, PH=plant height, CV=coefficient variation, LSD=least significant different, ns=non-significant, \*=significant and SYI= Seed yield.

Table 2. The mean value of yield and yield components of Rhodes grass cultivars over years and over locations at midland agro-ecologies.

Varieties	DE	DF	LSR	DMY t/ha	FBM t/ha	PH (cm)	SYI qt/ha
7384	8.33 <sup>b</sup>	77 <sup>b</sup>	0.59	15.44 <sup>a</sup>	75	164.7	2.5 <sup>ab</sup>
Masaba	10.67 <sup>ab</sup>	82 <sup>a</sup>	0.56	13.8 <sup>ab</sup>	50.83	171	0.917 <sup>b</sup>
13325	10 <sup>ab</sup>	77.6 <sup>b</sup>	0.76	9.73 <sup>b</sup>	58.42	171.7	2.66 <sup>a</sup>
DZ-253	10.33 <sup>ab</sup>	78.3 <sup>ab</sup>	0.86	12.83 <sup>ab</sup>	58.33	181.7	1.83 <sup>ab</sup>
6633	12.67 <sup>a</sup>	72.67 <sup>c</sup>	0.8	14.9 <sup>a</sup>	52.92	171.7	2.66 <sup>a</sup>
Mean	10.4	77.5	0.66	13.34	59.1	172.1	2.12
CV	13.5	2.8	26	18.8	22	5.5	39.7
LSD (5%)	*	**	ns	*	ns	ns	**

DE=days to emergence, DF=days to flower, DM=days to maturity, LSR=leaf to stem ratio, DMY=dry matter yield, PH=plant height, CV=coefficient variation, LSD=least significant different, ns=non-significant, \*=significant, \*\* highly significant, and SYI= Seed yield.

### Dry matter Yield (t/ha)

The over year statistical data analysis result indicates that dry matter yield is a significant difference ( $P < 0.05$ ) among the cultivars at midland agro-ecology. The highest dry matter yield was recorded from ILRL 7384 (15.44 t/ha) cultivar flowed by ILRI 6633 (14.9 t/ha) cultivars. This result is higher than result studied conducted on farmers' fields in the central highlands of Ethiopia, on average the herbage yield of

Rhodes grass was from 8.74 to 9.1 tons DM/ha-1 per year on rain-fed conditions (Cook et al., 2005, CASCAPE, 2015 and HARC, 2004).

Additionally, a This result was higher than research conducted at Adami Tulu Agricultural Research Center (ATARC) and Negele Arsi Farmers Training Center (FTC) also indicated that the average herbage dry biomass yield of Rhodes grass was from 7.8 to 9.16 tons DM/ha-1 per year without manure application (Tesfaye *et al.*, 2020) which is almost similar with this study.

### ***Seed yield (qu/ha)***

The over year statistical data analysis result indicates that seed yield is highly significant difference ( $P < 0.01$ ) among the cultivars at midland and significant ( $p < 0.05$ ) among the cultivars at highland agro-ecologies. The highest seed yield was recorded from ILRI 6633 (2.66 qu/ha) and ILRI 13325 (2.66 qu/ha) cultivars at tropical area of agro-ecologies. This result was lower than result studied by (Tesfaye *et al.*, 2020 and Dawit *et al.*, 2020) which mean seed yield values recorded were 370.55 and 313.88 Kg/ha which were conducted at Adami Tulu Agricultural Research Center (ATARC) and Negele Arsi Farmers Training Center (FTC). This variation in seed yield productivity might be due the difference in soil fertility and soil moisture factors. On the other hand, climate and soil types or their interactions have effects on the performance of forage crops as indicated by Diriba *et al.*, (2014).

### **Conclusions and recommendation**

The result of this study indicated that ILRI 6633 and ILRI 7384 for tropical areas and ILRI 6633 and Masaba cultivars for highland agro-ecologies were well adapted and being productive regarding to dry matter yield and seed yield they are hopeful to fill the gap of low quantity ruminant feed supply of the community. Therefore; based up on its adaptability, Dry matter and seed yield ILRI 6633 and ILRI 7384 for tropical areas and ILRI 6633 and Masaba cultivars for highland agro-ecologies areas of Guji zone.

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## **Effect of Planting Time and Seeding Rate of *Dolichos Lablab* Intercropped with Maize on Grain Yield, Forage Yield and Quality at Bako, Ethiopia**

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### ***Abstract***

*The experiment was conducted at Bako Agricultural Research Center during 2019-2021 cropping season with the objective to determine the proper lablab planting time and seeding rate in maize-lablab intercropping for yield and yield attributes. The treatments contained four level of planting time (Simultaneous, 2 weeks, 4 weeks and 6 weeks) after maize plantation and three level of lablab seeding rates (25, 50 and 75)%. The experiment was laid out in randomized complete block design (RCBD) in factorial arrangement with three replications. Sole cropping of maize and *Dolichos lablab* was used as the control. Data collected from the experimental field were subjected to 'R' software (R, 2017, version 3.4.1). The analysis of variance indicated that maize grain yield was significantly affected ( $P < 0.05$ ) by lablab planting time and seeding rate interaction. The lower maize grain yield (5.15 t/ha) was recorded when Lablab was sown at 75% seed rate simultaneously with maize followed by 50% seed rate simultaneously with maize (6.11 t/ha). Sole cropped lablab had significantly higher dry matter (DM) yield (7.48 t/ha) than the intercropped system. Conversely, in lablab-maize mixture, better lablab DM yield (3.41 t/ha) was obtained when lablab was sown at 75% seed rate simultaneously with maize. However, maize grain yield was significantly reduced at 75% seed rate with simultaneously planting. The optimum DM yield (2.16t/ha) of lablab forage was obtained at 75% of lablab seed rate sown two weeks later than maize plantation. Although, the amount of herbage DM yield obtained from this combination is low, it is selected as best association due to the fact that this yield was attained without affecting maize grain and stover yields. The results also indicated that maize-lablab intercropping contributed to improve the nutritive values of maize stover. The highest crude protein (CP) content of maize stover (7.48%) was obtained from plots of 75% seed rate sown at two weeks after maize plantation. Similarly, higher in vitro dry matter digestibility (IVDMD) value was recorded from 75% lablab seed rate sown two weeks later than maize (55.41%). Higher cumulative total herbage DM yield was obtained from the mixture than both of the sole crops, resulted in total land equivalent ratio (LER) values greater than one and ranged 1.03-1.34. The higher LER value (1.34) obtained from the treatment with 75% lablab seeding rate sown at two weeks later than maize, while lower LER value (1.03) recorded from 25% seeding rate sown simultaneously with maize. Generally, the results of the current study indicated superiority of intercropping over sole cropping with intercropping treatments showing improved yields and yield attributes of fodder parameters. Therefore, it was concluded that maize-lablab intercropping at 75% of lablab seed rate sown two weeks later than maize plantation formed a better association with maize than the other combinations. Hence, this association is recommended as the most compatible combination to produce optimum forage biomass from maize-lablab intercropping without affecting maize grain yield in the study area.*

**Keywords:** *Dolichos lablab*, Herbage dry matter, intercropping, Maize stover, Seed rate

## **Introduction**

Feed shortage in terms of both quantity and quality is the leading problem affecting the livestock productivity in Ethiopia (Aduga, 2007; Fekede *et al.*, 2015). In the country, natural pasture and crop residues are the primary feed sources, which have low nutritive value (CSA 2018), resulting in poor animal performance unless concentrates or conserved hay is fed (Tolera 2008). One option to improve the quantity and quality of feed resource is to integrate improved forage legumes in to cropping system through food and forage crops intercropping strategy. (Tarekegn and Zelalem 2014). Intercropping is a type of mixed cropping and defined as the agricultural practice of cultivating two or more crops planted concurrently in the same space (Eskandari *et al.*, 2009; Lithourgidis. *et al.*, 2011). A major benefit of intercropping is to increase in production per unit area compared to sole cropping through the effective use of resources, including water, nutrients and solar energy (Nasri *et al.*, 2014). The practice is common among smallholder farmers (Seran & Brintha, 2010), in many areas of Africa as a part of traditional farming systems commonly implemented due to declining land sizes and food security problems (Waddington *et al.*, 2007; Tamiru, 2014). This common combination in intercropping systems mostly involves cereal legumes (Ijoyah & Fanen, 2012), particularly Maize-legumes (Matusso *et al.*, 2012). Inter-cropping legumes with cereal crops has multiple advantages in terms of improving biomass yield, nutritive value and land equivalent ratio (Beedy *et al.*, 2010; Jensen *et al.* 2020), but competition among mixtures and population densities of the component crops in cereal legume intercropping is the major attribute affecting yield as compared with sole cropping of cereals (Teshome *et al.*, 2016). Padhi *et al.* (2010) also suggested that one of the key factors successful for intercropping is proper plant density, which depends on the plant species as well as the specific varieties used.

Maize is widely grown in the project implementing areas and it is considered as a staple food besides its other uses such as animal feed. However, sole cropping of the maize crop is widely practiced though there is a shortage of arable land. In an investigation of the lablab varietal selection and planting density in maize intercropping system, improved yield parameters of maize and land equivalent ratio were recorded (Diriba *et al.*, 2003; Temesgen *et al.*, 2014). The same authors reported that in lablab maize intercropping, lower lablab plant density that sown after maize reached knee growing stage resulted in lablab stunted growth and yield reduction and the higher rate planted simultaneous with maize tended to reduce the maize yield components. Therefore, to utilize the resources efficiently and improve productivity, lablab planting time and planting density with maize intercropping is an important knowledge gap that requires investigation in the study area. Thus, this study was aimed with the objectives of determining the proper lablab planting time and to fix the appropriate lablab plant density in maize intercropping for yield and yield attributes.

## **Materials and Methods**

### ***Description of the Study Area***

The experiment was conducted at Bako Agricultural Research Center for three (2019-2021) consecutive cropping season. The experimental site is located in sub- humid areas of central western Ethiopia at a distance of 260 km to the west of Addis Ababa on the main road to Nekemte and represents mid-altitude agro-ecology of the country. The area lies at a latitude of 9°06'N; longitude of 37°09'E, and at an altitude of 1650 m above sea level. The area receives an annual rainfall of about 1200 mm, 90% of which falls

between June and September. Average temperature of the study area is about 27°C ranging from 22°C to 31°C. About 60% of the soil of BARC is reddish brown in color, and clay-loam in texture. Dominant soil type is Nitosols with fertile alluvial soils in valley bottom. The area is known for its mixed farming system and in which the system is predominantly maize- based mono- cropping with low soil fertility, which directly influences the production and productivity of the cultivated crops. Cattle and sheep are important livestock species found in the areas. Major animal feed resources are natural pasture, crops residues, improved forage grasses, herbaceous legumes and multipurpose trees.

### *Treatment Description and Experimental Design*

Newly released lablab variety (Beresia-55) and maize variety BH-661 were used for intercropping. Four level of planting time (Simultaneous, 2 weeks, 4 weeks and 6 weeks) after maize plantation and three level of lablab seeding rates (25, 50 and 75) % were used as experimental treatments. A randomized complete block design in factorial arrangement with three replications was used to conduct the experiment. A plot size of 3.7 m x 4 m (14.8 m<sup>2</sup>) having 6 rows of maize with intra-row spacing of 30 cm and 75 cm distance between rows with 1m distance between plots while lablab seed intra-row spacing of 40 cm between maize raw were used to apply the treatment. Fertilizer was applied to the experimental plots at the recommended rate for the main crop.

Table 1: Description of the experimental treatments

Treatments	Description	Treatments	Description
T1	Sole Maize	T8	50 % at 2 weeks after maize
T2	Sole Lablab	T9	50 % at 4 weeks after maize
T3	25 % with Simultaneous	T10	50 % at 6 weeks after maize
T4	25 % at 2 weeks after maize	T11	75 % with Simultaneous
T5	25 % at 4 weeks after maize	T12	75 % at 2 weeks after maize
T6	25 % at 6 weeks after maize	T13	75 % at 4 weeks after maize
T7	50 % with Simultaneous	T14	75 % at 6 weeks after maize

### *Collected Data*

Data on lablab herbage biomass, maize grain yields, stover yields and maize stover for nutritional quality analysis were quantified. The lablab herbage was harvested for biomass yield determination at 40-50% flowering and maize stover was harvested after maize grain was harvested.

### *Chemical Analyses*

Representative samples were collected from each treatments and dried in an oven at 65 °C for 72 hours. The oven-dried samples were ground to pass 1 mm sieve screen size stored at room temp until sub-sampled for laboratory analysis. The ground samples were kept in sealed plastic bags pending for chemical analysis. The nitrogen (N), dry matter (DM), organic matter (OM), and ash content were analyzed according to AOAC (1990). The CP content was calculated by multiplying N content with a factor of 6.25. Neutral detergent fibers (NDF), acid detergent fiber (ADF), and acid detergent lignin

(ADL) were analyzed based on the method of Van Soest and Robertson (1985). The *in vitro* dry matter digestibility was determined by using Tilley and Terry (1963) procedure.

### ***Land equivalent ratio (LER)***

Land equivalent ratio was used to assess the advantage of crop production in mixture. LER was defined as the relative area of mono-crop plant required for the same yield obtained from its mixture. The LER was calculated using the formula given below:

$$\mathbf{LER = Yim/Ysm + Yil/Ysl}$$

Where, Yim and Ysm are the yields of intercrop and sole cropping of maize, and Yil and Ysl are the yields of intercrop and sole cropping of lablab.

A LER of 1.0 indicates that the two intercropped species make alike demands on the same limiting resources. A LER more than 1.0 reveals an intercropping advantage or a demonstration that interspecific facilitation is higher than interspecific competition so that intercropping results in greater land-use efficiency. A LER under 1.0 reveals mutual antagonism in the intercropping system. As a result, a LER less than 1.0 has no intercropping advantage and indicates that interspecific competition is more than interspecific facilitation in the intercropping system (Fetene 2003; Wahla *et al.* 2009).

### ***Data Analysis***

All collected parameters were subjected to 'R' software (R, 2017, version 3.4.1). Whenever the effects of the treatments were found to be significant, the means were compared using Tukey's test at 5% level of significance.

### **Results and Discussion**

Analysis of variance showed that a significant effect of lablab seeding rate and planting time interaction on maize grain and stover yield ( $p < 0.05$ ) and lablab herbage dry matter yield ( $p < 0.001$ ) (Table 2). The main effects of planting time were highly significant ( $p < 0.001$ ) on all parameters were specified, whereas the main effects of lablab plant population was showed significant effect on maize grain and lablab dry matter yield, but non-significant effect was shown on maize stover ( $p > 0.05$ ). Experimental year and lablab plant population effects showed similar trend in this study.



Table 2: Analysis of variance for lablab herbage DM yield, maize grain and stover yields as influenced by lablab seed rate, planting time and the interaction effects

Source of variation	Mean squares			
	DF	MGY	MSY	LDMY
Replication	2	0.853	2.127	0.064
Planting time (PT)	3	7.403***	6.829***	13.530***
Seed rate (SR)	2	4.173**	0.219	15.796***
PT*SR	6	1.766*	1.599*	2.897***
Year	2	12.915***	1.101	0.603***
Residual	70	0.7564	0.667	0.047

Where: DF= degree of freedom, MGY= maize grain yield; MSY= maize stover yield, LDMY= lablab DM yield

### Maize Grain yield

The interaction effects of lablab plant population and planting time showed slightly significant ( $p < 0.05$ ) effects on maize grain yield. Intercropping of lablab at high population density simultaneous with maize significantly declined maize grain yield as compared to other treatments (Table 3). The treatment with intercropped 75% of lablab seed rate simultaneous with maize had significantly lower maize grain yields (5.15 t/ha) followed by 50% of lablab seed rate simultaneous with maize (6.11 t/ha), while supreme and statistically comparable maize grain yields were obtained from other maize in intercropped (6.11-7.46 t/ha) or sole-maize (6.69 t/ha). The maize grain yield reduction at this junction might be associated with interspecific competition between the intercrop components for growth resources and the depressive effects of lablab on maize at the early growth stage because both crops were planted simultaneously and might be caused lodging of some of the maize plants before the harvesting stage was reached. The results in the present study was in agreement with findings of Diriba *et al.* (2003), who reported lower maize grain yield (4.34 t/ha) from the plots where lablab was simultaneously planted with maize and highest maize grain yield (7.04 t/ha) from plots where lablab was intercropped at 6 weeks after maize plantation. Similar findings were made by Gbaraneh *et al.* (2004) and Bonginkosi *et al.* (2018), who noted a reduction in maize grain yield when lablab was sown less than two weeks later than maize.

Table 3. Interaction effect of Dolichos lablab seeding rate with planting time on maize grain yields (t/ha).

Planting Time	Seed Rate (%)		
	25	50	75
Simultaneous	7.05 <sup>a</sup>	6.11 <sup>ab</sup>	5.15 <sup>b</sup>
2 weeks	7.06 <sup>a</sup>	6.98 <sup>a</sup>	7.07 <sup>a</sup>
4 weeks	7.20 <sup>a</sup>	7.46 <sup>a</sup>	6.91 <sup>a</sup>
6 weeks	7.40 <sup>a</sup>	7.26 <sup>a</sup>	6.91 <sup>a</sup>
Sole maize	6.69		
CV%	12.6		
SEM	0.290		
<i>p</i> -value	0.041		

<sup>a,b</sup> Mean having different superscripts differ significantly ( $p < 0.05$ ), CV=Coefficient of variation, SEM= Standard error of mean

### Maize stover and Lablab herbage DM yields

The analysis result of maize stover yield and Dolichos lablab herbage yield grown in pure stand and in intercropping with different seeding rates at different planting time are indicated in table 4. The interaction effects of lablab seed rates and planting time in maize-lablab mixture significantly influence the maize stover yield ( $p < 0.017$ ) and Dolichos lablab herbage DM yield ( $p < 0.001$ ). The lower and statistically similar maize stover values for 75%, 50 % and 25% of lablab plant population were 5.95 t/ha, 6.29 t/ha and 6.2 t/ha, respectively was obtained from the plots where lablab was simultaneously planted with maize, while higher and statistically at par mean maize stover yields were obtained from plots of other maize in intercropped where lablab was plated at any time after maize plantation (6.11-7.46 t/ha) or sole maize (6.86 t/ha). In this study, the maize grain yield and stover yield showed the same trend.

Sole cropped lablab had significantly higher herbage DM yield (7.48 t/ha) than the intercropped system at varies lablab population density (0.19-3.41 t ha<sup>-1</sup>). In lablab-maize combination, better lablab DM yield was obtained when Lablab was sown at 75% seed rate simultaneously with maize (3.41 t/ha) followed by 75% seed rate two weeks later than maize plantation (2.16 t/ha). The herbage DM yield reduction of the intercropped lablab might be associated with interspecific competition between the intercrop components for growth resources and the shading effects of maize on lablab at the early growth. The optimum biomass yield (2.16t/ha) of lablab forage was obtained at 75% of lablab seed rate sown two weeks later than maize plantation. Although, the amount of herbage DM yield obtained from this combination is low, it is selected as best association due to the fact that this yield was attained without affecting maize grain and stover yields. These results were in conformity with Diriba *et al.* (2003) who noted that lower maize grain yield (4.34 t/ha) and higher lablab herbage DM yield (3.15 t/ha) was recorded in plots where Lablab was simultaneously planted with maize. Conversely, the same authors reported that highest maize grain yield (7.04 t/ha) and lower lablab herbage DM yield (1.63 t/ha) from plots where Lablab was intercropped at 6 weeks after maize planting, which was in agreement with the current finding.

Table 4. Interaction effect of lablab seeding rate with planting time on maize stover yield (t/ah) and Dolichs lablab DM yield (t/ha)

Planting Time	Seed Rate (%)					
	Maize stover yield			Lablab dry matter yield		
	25	50	75	25	50	75
Simultaneous	6.20 <sup>bc</sup>	6.29 <sup>bc</sup>	5.95 <sup>c</sup>	0.92 <sup>d</sup>	1.28 <sup>c</sup>	3.41 <sup>a</sup>
2 weeks	6.70 <sup>abc</sup>	7.59 <sup>a</sup>	7.22 <sup>ab</sup>	0.55 <sup>e</sup>	0.65 <sup>de</sup>	2.16 <sup>b</sup>
4 weeks	7.78 <sup>a</sup>	6.68 <sup>abc</sup>	7.19 <sup>ab</sup>	0.33 <sup>ef</sup>	0.46 <sup>ef</sup>	0.91 <sup>d</sup>
6 weeks	7.08 <sup>abc</sup>	7.15 <sup>abc</sup>	6.84 <sup>abc</sup>	0.20 <sup>f</sup>	0.19 <sup>f</sup>	0.37 <sup>ef</sup>
Sole maize	6.86			-		
Sole lablab	-			7.48		
CV%	11.0			22.9		
SEM	0.253			0.073		
<i>p-value</i>	0.017			<.001		

<sup>a,b,c,d,e,f</sup> = Mean having different superscripts differ significantly, CV = Coefficient of variation, SEM = Standard error of mean

Furthermore, intercropping maize with lablab reliably increased the total fodder yield (maize + lablab). The highest total fodder yield was observed from 75% lablab seed rate sown at two weeks after maize (9.38 t/ha) followed by 75% of lablab seed ratio sown at simultaneously with maize (9.36 t/ha). Improved total fodder (maize stover + lablab) DM yields could be attributed to higher proportion of lablab in the intercrop and efficient utilization of water resources and soil nutrients in maize-lablab mixture. The results are supported by Bonginkosi *et al.* (2018) who reported that an increase of total fodder yield when maize was intercropped with lablab (12.11 t/ha), compared to maize grown as mono crop (7.35 t/ha). A similar effect of intercropping on fodder yield was observed by Dahmardeh *et al.* (2009) and Reddy *et al.* (2009) when maize was intercropped with lablab or cowpea.

### ***Chemical Composition***

The effect of lablab plant population and planting time on the nutritive content of maize stover is illustrated in (Table 5). The data revealed that the interaction effect of lablab seed rate and planting time intercropped with maize had significant influence ( $p < 0.001$ ) on CP and IVDMD of maize stover. On the other hand, the main and interaction effects of lablab seed rate and planting time in lablab-maize intercropping was found to be non-significant ( $p > 0.05$ ) on DM% and NDF% of maize stover. The highest CP content of maize stover (7.48%) was obtained from plots of higher lablab seed rate (75%) sown at two weeks after maize plantation, followed by lablab intercropped with maize at 75% simultaneous with maize (7.30%), but they were statistically at par. Conversely, the least and statistically similar CP values were recorded from pure stand of maize (4.59%) and plots of 25% lablab seed rates of maize and lablab mixture at all described planting time (4.73-4.91%). The increased CP concentration in maize stover when inter-cropped with lablab is in agreement with the findings of Akhtar *et al.* (2013) and Rahel *et al.* (2021), who reported increases in CP concentration in sorghum when inter-cropped with lablab.

Similar to CP yield, significantly higher IVDMD value was recorded from 75% lablab seed rate sown at two weeks after maize (55.41%) followed by the same lablab seed rate (75%) sown simultaneously with maize (55.37%), although both of these means are statistically similar. Improved maize stover CP and IVDMD yields could be attributed to higher proportion of lablab in the intercrop and had better ability to extract nitrogen from soil than the other treatment combination, as it also offered higher total fodder (maize stover + lablab) yields. Interestingly, the lowest and significant ADF and ADL was recorded from 75% lablab seed rate sown at two weeks after maize plantation followed by same seed rate sown simultaneously with maize. Similar findings were made by Bonginkosi *et al.* (2018) and who reported increase of maize nutritive value with a decrease in ADF and ADL when intercropped with legumes.

Table 5. Interaction effect of maize with Lablab intercropping on chemical composition and in IVDMD of maize stover

Planting Time	Seed Rate (%)	DM %						
		DM %	Ash	CP	NDF	ADF	ADL	IVDMD
Simultaneous	25	94.89	7.90 <sup>ab</sup>	4.91 <sup>e</sup>	62.36	44.71 <sup>a</sup>	6.13 <sup>a</sup>	49.33 <sup>d</sup>
2 weeks	25	94.48	7.97 <sup>ab</sup>	4.88 <sup>e</sup>	62.05	43.69 <sup>ab</sup>	5.33 <sup>bc</sup>	49.68 <sup>cd</sup>
4 weeks	25	95.14	8.09 <sup>ab</sup>	4.78 <sup>e</sup>	62.04	45.46 <sup>a</sup>	5.64 <sup>b</sup>	49.43 <sup>cd</sup>
6 weeks	25	95.21	8.49 <sup>a</sup>	4.73 <sup>e</sup>	63.11	45.31 <sup>a</sup>	5.51 <sup>bc</sup>	48.78 <sup>d</sup>
Simultaneous	50	95.5	7.30 <sup>abc</sup>	6.58 <sup>b</sup>	59.93	43.54 <sup>abc</sup>	5.14 <sup>c</sup>	52.40 <sup>bc</sup>
2 weeks	50	95.2	7.49 <sup>abc</sup>	6.58 <sup>b</sup>	62.18	43.43 <sup>abc</sup>	5.22 <sup>bc</sup>	50.68 <sup>cd</sup>
4 weeks	50	95.03	8.03 <sup>ab</sup>	5.65 <sup>d</sup>	62.32	45.15 <sup>a</sup>	6.14 <sup>a</sup>	50.19 <sup>cd</sup>
6 weeks	50	95.07	8.11 <sup>ab</sup>	5.68 <sup>cd</sup>	63.08	45.23 <sup>a</sup>	5.18 <sup>bc</sup>	48.74 <sup>d</sup>
Simultaneous	75	95.24	6.99 <sup>bc</sup>	7.30 <sup>a</sup>	59.53	41.47 <sup>bc</sup>	5.04 <sup>c</sup>	55.37 <sup>ab</sup>
2 weeks	75	95.3	6.07 <sup>c</sup>	7.48 <sup>a</sup>	59.39	41.28 <sup>c</sup>	5.11 <sup>c</sup>	55.41 <sup>a</sup>
4 weeks	75	94.89	7.51 <sup>ab</sup>	6.17 <sup>bc</sup>	62.59	44.97 <sup>a</sup>	5.23 <sup>bc</sup>	50.47 <sup>cd</sup>
6 weeks	75	95.35	8.68 <sup>a</sup>	6.07 <sup>cd</sup>	62.22	45.58 <sup>a</sup>	5.44 <sup>bc</sup>	49.34 <sup>d</sup>
Sole maize		94.67	8.75	4.59	62.86	45.63	6.21	48.58
CV %		0.5	6.3	2.9	2.2	1.7	2.9	2.0
SEM		0.2926	0.2786	0.0977	0.774	0.44	0.092	0.584
<i>p</i> -value		0.508	<.023	<.001	0.179	0.008	<.001	<.001

<sup>a,b,c,d,e</sup> Mean in a column within the same category having different superscripts differ significantly, CV=Coefficient of variation, SEM= Standard error of mean DM=dry matter, CP=crude protein, NDF=neutral detergent fiber, ADF=acid detergent fiber, ADL=acid detergent lignin, IVDMD= *in-vitro* dry matter digestibility.

### Land Equivalent Ratio

In the current study, higher cumulative total herbage DM yield was obtained from the mixture than both of the sole crops, resulted in total LER values greater than one. Total LERs greater than one are an indication of an intercropping system's effectiveness on forage productivity, land usage and beneficial association between maize and lablab species. Values of LER below a unity indicates that the DM yield of crops in the mixture is below a pure stands of the respective crops. The acceptable total LER in this study ranged from 1.03 to 1.34 (table 5), this indicates that 0.03% to 34% more land area would be required by a mono cropping system for equal yield of intercropping system. The higher LER value (1.34) obtained from the treatment with 75% lablab seeding rate sown at two weeks later than maize, while lower LER value (1.03) recorded from treatment with 25% seeding rate sown simultaneously with maize.

Similar results were obtained by Amole *et al.* (2015), in which LER of grass-legume mixtures herbage DM yield recorded for Lablab intercropped with *Andropogon gayanus* greater than 1 and which ranged between 1.21 and 1.48. Conversely, the LER values obtained in this study were lower than the values noted by Rahel *et al.* (2021) which ranged from 1.54 to 1.87 in sorghum-lablab inter-cropping. Even though, all the tested treatment combination in intercropped have the potential to be used for maize-lablab

intercropping, treatment with 75 % lablab seeding rate sown at two weeks later than maize was found to be more compatible and high yielder so that it produce high total LER (1.34).

Table 5: Land equivalent ratio (LER) for dry matter yield of Lablab intercropped with maize

Planting time	Seed Rate (%)	MPLER	LPLER	TLER
Simultaneous	25	0.90	0.12	1.03
2 weeks	25	0.98	0.07	1.05
4 weeks	25	1.14	0.05	1.18
6 weeks	25	1.03	0.03	1.06
Simultaneous	50	0.92	0.17	1.09
2 weeks	50	1.11	0.09	1.19
4 weeks	50	0.97	0.06	1.04
6 weeks	50	1.04	0.03	1.07
Simultaneous	75	0.87	0.46	1.32
2 weeks	75	1.05	0.29	1.34
4 weeks	75	1.05	0.12	1.17
6 weeks	75	1.00	0.05	1.05
Mean		1.01	0.13	1.13

Where; MPLER= maize partial land equivalent ratio; LPLER= Lablab partial land equivalent ratio and TLER= Total land equivalent ratio

### Conclusion and Recommendations

The present study has revealed the beneficial effects of intercropping maize with lablab on sustainable fodder and maize grain yield production. The lower maize grain yield was recorded when Lablab was sown at 75% seed rate simultaneously with maize followed by 50% seed rate simultaneously with maize. Dolichos lablab sole stand had higher herbage DM yield than the intercropped system. Nevertheless, in lablab-maize mixture, better lablab DM yield was obtained when lablab was sown at 75% seed rate simultaneously with maize. It means that, in maize-lablab intercropping system, high rate of lablab plant population sown simultaneously with maize resulted in low maize grain yield and higher lablab herbage DM yields. The optimum biomass yield of lablab was obtained at 75% of lablab seed rate sown two weeks later than maize plantation. Although, the amount of herbage DM yield obtained from this combination is low, it is the best association due to the fact that this yield was attained without affecting maize grain and stover yields. The results indicated that maize-lablab intercropping contributed to improve the nutritive values (CP and IVDMD) of maize stover, but didn't affect DM%. Higher cumulative total herbage DM yield was obtained from the mixture than both of the sole crops, resulted in total LER values greater than one and ranged 1.03-1.34. In conclusion, this study has shown that Maize-lablab intercropping technology is a particular importance to resource poor crop-livestock farmers for it would provide improved fodder production to fill the feed gap during the dry season while improving maize grain yield from the same piece of land. Therefore, it was concluded that maize-lablab

intercropping at 75% of lablab seed rate sown two weeks later than maize plantation formed a better association with maize than the other combination, Hence, this association is recommended as the most compatible combination to produce optimum forage biomass from maize-lablab intercropping without affecting maize grain yield in the study area.

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# Effects of Fertilizer Level and Time of Application on Dry Matter Yield, Species Composition and Nutrient Content of Natural Grass at Bako, West Showa Zone of Oromia

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## Abstract

*The activity was conducted to assess the effects of fertilizer rates and time of application on herbage dry matter (DM) yield, species composition and chemical composition of natural grass at Bako areas during 2019-2021 cropping season. The treatments contained four rates of NPS and N fertilizers combinations (100:100, 75:75, 50:50 and 25:25 kg ha<sup>-1</sup>) and three times of application (1 week, 3 weeks, 5 weeks after the commencement of main rainy season, control). The experiment was laid out in randomized complete block design in factorial arrangement with three replications. Data on forage DM yield, species composition and chemical composition of natural grasses were collected and subjected to 'R' software. The analysis of variance indicated that herbage DM yield was significantly affected ( $P < 0.005$ ) by fertilizer rate and time of application interaction. Higher herbage DM yield (15.32 ton ha<sup>-1</sup>) was recorded from the fertilizer rate of 100 NPS: 100 N at third weeks' of application time, followed by 75 NPS: 75 N fertilizer rate treated plots (14.6 ton ha<sup>-1</sup>) at same time of application, but both are statistically at par. However, the lower DM yield was received from unfertilized treatments (6.87 ton ha<sup>-1</sup>). The crude protein (CP) and in-vitro dry matter digestibility (IVDMD) contents of the grasses were significantly ( $P < 0.001$ ) influenced by fertilizer rate and time of application. The highest CP (8.03%) and IVDMD (53.22%) were recorded from 75 NPS: 75 N kg/ha fertilizer rate at three weeks' time of application, while the lowest (6.15%) CP and (46.75%) IVDMD content was recorded from unfertilized plots. Results from the partial budget analysis also indicated that 75 NPS: 75 N kg/ha fertilizer rate applied at the three weeks' time of application had the better economic benefit (85,389 Birr·ha<sup>-1</sup>) with best MRR of 1413.3% followed by fertilizer rate of 100 NPS: 100 N kg ha<sup>-1</sup> at same time of application, which gave higher 88820.7 Birr·ha<sup>-1</sup> with a MRR of 1,171.5% than the other treatment factors. Therefore, it can be concluded that use of combined fertilizers at a rate of 75 NPS: 75 N during the 3 weeks' time of application optimize both biological and economic benefits and should be recommended for promising natural grass production.*

**Key words:** Fertilizer rate, Herbage dry matter, Natural grass, Nutrient content, Species composition

## Introduction

Livestock production is increasingly constrained by feed shortage both in quantity and quality in Sub-Saharan countries (Kindomihou *et al.*, 2014). Shortage of quality and quantity of livestock feed in Ethiopia is taking the lion share for the poor livestock productivity (Mengistu *et al.*, 2017). The major feed resources in the country are natural grazing land and crop residues (CSA, 2017). Grazing lands in the country play great role in livestock production. However, the productivity was extremely low due to serious grazing land degradation problem (Ulfina *et al.*, 2013). The poor production of pasturelands and large herd size on small grazing lands caused overgrazing of natural pasturelands and less productive due to its continuous heavy grazing and mismanagement practices (Abule, 2015). This results in serious land

degradation and decline in feed quality (dry matter yield and quantity), which in turn leads to invasion by unpalatable plant species and the critical shortage of animal feed, below the maintenance requirement for livestock production (Yadessa *et al.*, 2016). Lack of nutrients, inadequate management of pastures and inappropriate cultural practices are responsible for pasture degradation. Soil fertility status of the grazing lands is one of the main factors that could contribute to the low productivity, quality and botanical composition of a natural pasture (Kebede *et al.*, 2016; Głowacz and Niżnikowski 2018).

Proper nutrient management in natural pastures improves the vigor of the forages and quality, allows them to compete with undesirable plants (weeds) and improves overall yield (Howard 2010). With good soil fertility and fertilizer management, the productivity of hay and pasture fields can be greatly improved (Ross 2005). The application of nitrogen has proved to be effective in maximizing plant growth, the leaf area and the production of dry matter and nutritional status of grasses (Bonfim-Silva and Monteiro, 2006; Batista and Monteiro, 2008). Mekonnen *et al.* (2021), pointed out that application of fertilizer either sole urea or mixed with NPS gave promising herbage DM yield ranging from (8.9-12.9 t/ha) from pastureland intervention sites. Hence, application of fertilizer seems imperative to enhance plant growth and increase herbage biomass production and quality. Therefore, the aim of the current study was to determine the optimum fertilizer rate and to determine proper time of fertilizer application on natural grass land.

## **Materials and Methods**

### ***Description of the Study Area***

The experiment was conducted at Bako Agricultural Research Center for three (2019-2021) consecutive years. The experimental site is located in sub-humid areas of central western Ethiopia at a distance of 260 km to the west of Addis Ababa on the main road to Nekemte and represents mid-altitude agro-ecology of the country. The area lies at a latitude of 9°06'N; longitude of 37°09'E, and at an altitude of 1650 m above sea level. The area receives an annual rainfall of about 1200 mm, 90% of which falls between June and September. Average temperature of the study area is about 27°C ranging from 22°C to 31°C. About 60% of the soil of BARC is reddish brown in color, and clay-loam in texture. Dominant soil type is Nitosols with fertile alluvial soils in valley bottom. The area is known for its mixed farming system and in which the system is predominantly maize-based mono-cropping with low soil fertility, which directly influences the production and productivity of the cultivated crops. Cattle and sheep are important livestock species found in the areas. Major animal feed resources are natural pasture, crops residues, improved forage grasses, herbaceous legumes and multipurpose trees.

### ***Treatment Description and Experimental Design***

Four combined fertilizer levels; (NPS: N) (100:100, 75:75, 50:50 and 25:25) kg ha<sup>-1</sup> and three level of application time (1 week, 3 weeks and 5 weeks) after the commencement of main rainy season were used as experimental treatments. A randomized complete block design in factorial arrangement with three replications was used to conduct the experiment. A plot size of 5 m x 5 m (25 m<sup>2</sup>) with 2 m distance between plots was used to apply the treatment. Nitrogen was applied in the form of urea in two split applications (1/3 dose at the beginning and 2/3 dose after a month of the first application) and NPS was applied at the beginning together with the nitrogen applied at start.

Table 1: Treatments description

Treatments	Description	
	Fertilizer rate (NPS:N) kg ha <sup>-1</sup>	Time of application (AT)
1	100 : 100	<b>1 week</b>
2	100 : 100	<b>3 weeks</b>
3	100 : 100	<b>5 weeks</b>
4	75 : 75	<b>1 week</b>
5	75 : 75	<b>3 weeks</b>
6	75 : 75	<b>5 weeks</b>
7	50 : 50	<b>1 week</b>
8	50 : 50	<b>3 week</b>
9	50 : 50	<b>5 weeks</b>
10	25 : 25	<b>1 weeks</b>
11	25 : 25	<b>3 week</b>
12	25 : 25	<b>5 weeks</b>
13	Control	

### ***Data Collection and Measurement***

Herbage DM yield and species diversity were determined using 1m x 1m quadrats. The quadrats were placed purposely at five (four at the corner and one at the center) spots per 25 m<sup>2</sup> and the biomass was harvested using manual system by using sickle. The species compositions were classified from samples of pasture in each experimental plot by counting (Mannetje *et al.*, 1976). Identification of species was undertaken in the field together with experienced personnel on the basis of plant morphological, structural, and floristic characteristics of each botanical composition. In each of the sample plots, the herbaceous vegetation was harvested together with grass species at 50% flowering stage. The biomass was harvested twice a year (two-month interval). The cut biomass was measured just after mowing with suspended field balance. The five samples were thoroughly mixed and composite subsamples per treatment were taken from each replication and about 300 gms of sample was taken to the laboratory.

### ***Samples and Sample Preparation***

Representative samples were collected from each treatment and dried in an oven at 65 °C for 72 hours until a constant weight was obtained to determine the herbage DM yield. The oven-dried samples were ground to pass 1 mm sieve screen size stored at room temp until sub-sampled for laboratory analysis. The ground samples were kept in sealed plastic bags pending for chemical analysis.

### ***Chemical Analyses***

Representative samples were subjected to chemical analysis for determination of the forage chemical composition measurements. The nitrogen (N), Dry matter (DM), Organic matter (OM), and ash content were analyzed according to AOAC (1990). The crude protein (CP) content was calculated by multiplying N content with a factor of 6.25. Neutral detergent fibers (NDF), acid detergent fiber (ADF), and acid detergent lignin (ADL) were analyzed based on the method of Van Soest and Robertson (1985). The *in vitro* dry matter digestibility was determined by using Tilley and Terry (1963) procedure.

### ***Partial Budget Analysis***

The economically acceptable treatments were determined by partial budget analysis to estimate the gross value of the herbage DM yield by using the methodology described in CIMMYT (1988) in which market prices for inputs at sowing and for outputs at harvesting were used. Only total costs that varied (TCV) were used to compute costs. Natural grass, fertilizers, and application cost of fertilizers were considered as variable with their cost. Costs of field management, guarding, harvest and transportation were not included in the analysis as they were not variable. The ideas used in the partial budget analysis were the mean herbage yield of each treatment, the gross benefit ha<sup>-1</sup> (from mean yield for each treatment) and the field price of fertilizers (NPS, urea and the time of application costs). Marginal rate of return (MRR), which refers to net income (NI) obtained by incurring a unit cost of fertilizer and its application, was calculated by dividing the net increase in yield of herbage dry matter due to the application of each fertilizer's rate. Total variable cost (TVC) was calculated by summing up the costs that vary, including the cost of fertilizers; urea (1669.8 ETB·qt<sup>-1</sup>), NPS (2058 ETB·qt<sup>-1</sup>) and for each time of application cost (4 persons, 75 birr/day/ha) and herbage yield of natural grass was valued at an average open market price of 2.00 Birr/kg in November 2021 during harvesting time. Both the costs and benefits were converted to monetary values in Ethiopian Birr (ETB) and reported per hectare basis. Treatments net benefits and total variable costs (TVC) were compared using dominance analysis. Net return (NR) or net benefit was calculated as the amount of money left when total variable costs (TVC) are subtracted from total returns or gross field benefit (TR).

$$\mathbf{NR = TR - TVC}$$

The dominance analysis or the change in net income ( $\Delta NR$ ) was computed as the difference between the change in total return ( $\Delta TR$ ) and the change in total variable costs from the control ( $\Delta TVC$ ):

$$\mathbf{\Delta NR = \Delta TR - \Delta TVC}$$

All treatments which had NR less than or equal to treatment with lower TVC were marked with a letter “D” since they were dominated and eliminated from any further analysis. Undominated treatments were subjected to marginal rate of return (MRR) analysis as suggested by CIMMYT (1988) in a stepwise manner, moving from lower TVC to the next. The marginal rate of return (MRR), which measures the increase in net income ( $\Delta NI$ ) in relation with each additional unit of expenditure ( $\Delta TVC$ ) normally expressed as a percentage:

$$\mathbf{MRR (\%) = \Delta NR / \Delta TVC \times 100}$$

### ***Data Analysis***

All collected parameters were subjected to ‘R’ software (R, 2017, version 3.4.1). Whenever the effects of the treatments were found to be significant, the means were compared using Tukey’s test at 5% level of significance.

## **Result and Discussion**

### ***Analysis of Variance***

The analysis of variance shown that highly significant effects ( $p < 0.001$ ) of fertilizer rates and time of application on herbage DM yield of natural grassland. Similarly, experimental year significantly affected herbage DM yield performance of natural grassland ( $p < 0.001$ ). Fertilizer rate by time of application interaction highly affected ( $p < 0.005$ ) herbage DM yields. However, no significant interactions were observed between the experimental years and either of the treatment factors (table 2).

Table 2: Mean squares of ANOVA for Fertilizer Rate and time of application on herbage dry matter yields of the natural grass

Source of Variation	d.f	Mean Square	Variance	F Probability
Replication	2	3.5915	7.84	
Time of application (TA)	2	16.4548	35.92	<.001
Fertilizer rate (FR)	3	220.8565	482.14	<.001
Year (Yr)	2	36.6811	80.08	<.001
TA*FR	6	1.5936	3.48	0.005
TA*Yr	4	0.4084	0.89	0.474
FR*Yr	6	0.5839	1.27	0.28
TA*FR*Yr	12	0.6865	1.5	0.145
Residual	70	0.4581		
Total	107			

### ***Herbage Dry Matter Yield***

Pleasingly, the interaction effect of fertilizer rate and time of fertilizer application had higher significance on herbage dry matter yields of the natural pasture ( $p < 0.005$ ). Substantial herbage DM yield increases were recorded due to treatment factors which was ranged from 6.87-15.32) ton ha<sup>-1</sup>. The highest herbage DM yield (15.32 ton ha<sup>-1</sup>) was recorded from application of 100 NPS: 100 N fertilizer rate at the third week application time (three weeks after main rainy season started), followed by 75 NPS: 75 N fertilizer rates treated plots (14.60 ton ha<sup>-1</sup>) at same time of application, but both are statistically at par. However, the lower DM yield was received from lower fertilizer rates, NPS: N (50:50 and 25:25) resulted between the range of 7.95-10.3 ton ha<sup>-1</sup> and unfertilized treatment (6.87 ton ha<sup>-1</sup>), respectively (Table 3). The result showed that there was a yield declining trend from higher rates of combined fertilizer application to the lower rates. This implies that fertilizer application had played significant roles in improving grassland management through enhancing forage productivity in the study area. A study by Cahit *et al.* (2010) showed that it is possible to boost forage yield and quality through addition of nitrogen and phosphorous. This result is in agreement with that of Kebede *et al.*, (2016), who reported that an increased DM yield of natural pasture (11.33 ton ha<sup>-1</sup>) due to chemical fertilizer application at around Holeta, central highland of Ethiopia.

Likewise, an increasing in herbage DM yield following chemical fertilizer application was described by earlier study (Hanife *et al.*, 2010; Ahmed *et al.*, 2013; Yossif & Ibrahim, 2013; Tesfaye *et al.*, 2015). Therefore, it is realistically evidence by the use of chemical fertilizer application as a potential intervention option for grassland improvement.

Table 3. Interaction effect of fertilizing rate and time of application on herbage dry matter yield of natural grass

Fertilizer Rate (NPS : N) kg/ha	Herbage Dry matter yield (t/ha)		
	Time of Application		
	1 week	3 weeks	5 weeks
100 : 100	14.08 <sup>bc</sup>	15.32 <sup>a</sup>	13.18 <sup>cd</sup>
75 : 75	12.77 <sup>d</sup>	14.60 <sup>ab</sup>	13.22 <sup>cd</sup>
50 : 50	9.54 <sup>ef</sup>	10.30 <sup>e</sup>	9.67 <sup>ef</sup>
25 : 25	8.09 <sup>g</sup>	8.75 <sup>fg</sup>	7.95 <sup>g</sup>
Control	6.87		
CV %	5.9		
SEM	0.226		
P-value	0.005		

Where; SEM=Standard Error of Mean; CV= Coefficient of variation

### Species Composition

The main effect of fertilizer level and time of application on herbage dry matter yields and species composition (grass and legumes) are presented in (Table 4). Even though main effects of time of NPS and N application did not show significant variation to species composition (grass and legumes), there were highly significant ( $p < 0.003$ ) effects due to the different applications of fertilizer rates. In the present study, the result of total species composition was shown similar yield increment trend with the herbage dry matter yields from lower to higher fertilizer rates treated plots. The highest proportion of grass (5.24) was recorded when fertilizer was applied at a rate of 100 NPS: 100 N, while the lowest (4.35) was at a rate of 25 NPS: 25 N. Similarly the highest proportion of legumes (2.58) was observed at a rate of 75 NPS: 75 N, while the lowest result was from control treatment (2.13). This is specified that application of increased rates of fertilizer considerably enhanced grassland species composition as compared to the lower rates and the control treatment. The study was also in agreement with Zewdu *et al.* (2010) stated that herbage dry matter yields of the grass component increased with the application of nitrogen fertilizer in the north-western parts of Ethiopia.

Table 4. Main effects of fertilizing rate and time of application on species composition of natural grass

Variables	Parameters		
	TSCP (No.of spp)	Grass (No.)	Legume (No.)
Control	6.5	4.37	2.13
1 week	7.32	4.90	2.41
3 weeks	7.18	4.81	2.37
5 weeks	6.94	4.65	2.29
SEM	0.1	0.1	0.049
SL	Ns	Ns	Ns
Fertilizer rates (NPS:N in kg ha <sup>-1</sup> )			

100:100	7.83 <sup>a</sup>	5.24 <sup>a</sup>	2.58 <sup>a</sup>
75 :75	7.36 <sup>ab</sup>	4.93 <sup>ab</sup>	2.43 <sup>ab</sup>
50 : 50	6.89 <sup>bc</sup>	4.62 <sup>bc</sup>	2.27 <sup>bc</sup>
25 : 25	6.49 <sup>c</sup>	4.35 <sup>c</sup>	2.14 <sup>c</sup>
SEM	0.2	0.115	0.057
CV (%)	12.5	12.5	12.5
SL	***	***	**

Where; TSCP= Total species composition; SEM=Standard Error of Mean; SL= Significance Level; CV=Coefficient of variation

The current finding showed that application of fertilizer increased the legume species composition as compared to unfertilized plots. Generally, the current study resulted with higher botanical composition of grasses than other legume species which might be related to decreasing proportions of legumes in the pasture as a result of a suppression suffered from shading of grasses (Feyter *et al.* 2005).

### ***Nutrient Content***

The interaction effect among fertilizer rate and time of application on the chemical composition of natural grassland is presented in (Table 5). The result indicated that all chemical composition parameters were significantly affected ( $p < 0.001$ ) by fertilizer rates and time of application while dry matter content (DM %) of the natural grass was not influenced ( $p < 0.092$ ) by both factors.

The crude protein (CP) content of the grasses was significantly ( $p < 0.001$ ) influenced by fertilizer rate and time of application. The highest CP content of the natural grass (8.03%) was recorded from application 75 NPS: 75 N fertilizer rate at three weeks' time of application and statistically par with T2,T4,T6,T7 and T9, while the lowest (6.15%) CP content was recorded from unfertilized plots. The interaction of fertilizer rate and time of application significantly affected the in-vitro dry matter digestibility (IVDMD) at ( $P < 0.001$ ). The highest (53.22%) IVDMD was observed from 75 NPS: 75 N kg/ha at three weeks' time of application. The lowest (46.75%) IVDMD was recorded in control group. The mean IVDMD of tropical grasses was 57.8% (Barton, 1997), which is slightly higher than the result obtained in this study. This might be due to the dissimilarity in soil fertility and agro ecologies of the study areas.

The current finding indicated that there was an increase in total ash and NDF content of the natural pasture with decrease in the rate of combined fertilizer application. In other words, there were slight improvements of the fiber fraction contents of natural grass due to combined application of fertilizers.

Table 5. Effect of fertilizer rate and a time of application on the chemical composition of the natural pasture

Treatment combination		DM%						
FR (NPS:N)	TA (week)	DM	Ash	CP	NDF	ADF	ADL	IVDMD
100:100	1	91.87	6.73 <sup>de</sup>	6.81 <sup>de</sup>	68.19 <sup>d</sup>	39.92 <sup>bcd</sup>	5.17 <sup>ab</sup>	48.95 <sup>ef</sup>
100:100	3	91.68	6.73 <sup>de</sup>	7.41 <sup>a-d</sup>	68.88 <sup>bcd</sup>	40.57 <sup>abc</sup>	5.20 <sup>ab</sup>	50.46 <sup>cd</sup>
100:100	5	92.07	6.64 <sup>ef</sup>	6.97 <sup>cde</sup>	68.38 <sup>cd</sup>	40.77 <sup>abc</sup>	5.40 <sup>a</sup>	50.70 <sup>bc</sup>
75:75	1	91.93	6.01 <sup>fg</sup>	7.65 <sup>ab</sup>	68.12 <sup>d</sup>	40.26 <sup>abc</sup>	5.23 <sup>ab</sup>	50.64 <sup>bc</sup>
75:75	3	91.88	5.72 <sup>g</sup>	8.03 <sup>a</sup>	66.14 <sup>e</sup>	38.33 <sup>e</sup>	5.31 <sup>ab</sup>	53.22 <sup>a</sup>
75:75	5	91.63	7.32 <sup>bcd</sup>	7.70 <sup>ab</sup>	67.65 <sup>d</sup>	38.41 <sup>de</sup>	4.32 <sup>de</sup>	48.84 <sup>d</sup>
50:50	1	91.86	7.19 <sup>cde</sup>	7.42 <sup>a-d</sup>	68.66 <sup>bcd</sup>	39.25 <sup>cde</sup>	4.66 <sup>cde</sup>	51.61 <sup>b</sup>
50:50	3	91.59	7.28 <sup>bcd</sup>	7.23 <sup>b-e</sup>	65.98 <sup>e</sup>	37.90 <sup>e</sup>	4.29 <sup>e</sup>	49.11 <sup>d</sup>
50:50	5	91.58	7.32 <sup>bcd</sup>	7.48 <sup>abc</sup>	68.44 <sup>cd</sup>	39.38 <sup>cde</sup>	4.99 <sup>bc</sup>	48.55 <sup>b</sup>
25:25	1	91.81	7.45 <sup>abc</sup>	6.60 <sup>ef</sup>	69.70 <sup>abc</sup>	41.30 <sup>ab</sup>	5.14 <sup>ab</sup>	51.64 <sup>b</sup>
25:25	3	91.73	7.9 <sup>ab</sup>	6.87 <sup>cde</sup>	70.04 <sup>ab</sup>	41.18 <sup>ab</sup>	5.44 <sup>a</sup>	49.84 <sup>cd</sup>
25:25	5	91.66	7.48 <sup>abc</sup>	7.23 <sup>b-e</sup>	68.95 <sup>bcd</sup>	40.72 <sup>abc</sup>	4.69 <sup>cd</sup>	49.97 <sup>cd</sup>
Control		91.69	7.96 <sup>a</sup>	6.15 <sup>f</sup>	70.74 <sup>a</sup>	41.78 <sup>a</sup>	5.31 <sup>ab</sup>	46.75 <sup>e</sup>
CV %		0.4	5.3	5.4	1.3	2.3	4.5	1.4
SEM		0.655	0.63	0.653	1.486	1.541	0.382	1.202
<i>p-value</i>		0.092	<.001	<.001	<.001	<.001	<.001	<.001

Where; FR=fertilizer rate, TA= application time, DM=dry matter, CP=crude protein, NDF=neutral detergent fiber, ADF=acid detergent fiber, ADL=acid detergent lignin, IVDMD= *in-vitro* dry matter digestibility, SEM=Standard Error of Mean; CV= Coefficient of variation.

### ***Partial Budget Analysis***

The result of partial budget analysis revealed that application of 75 NPS: 75 N kg ha<sup>-1</sup> at three weeks' time of application gave the highest net benefit of 85,389 Birr·ha<sup>-1</sup> and followed by 1,171.5 Birr·ha<sup>-1</sup> gained from treatment that received 100 NPS: 100 N kg/ha at same time of application, while the lowest net benefits 41,636.4 Birr·ha<sup>-1</sup> was obtained from control treatment (Table 6). Likewise, the highest marginal rate of return (MRR) 1413.3 % was obtained when 75 NPS: 75 N kg ha<sup>-1</sup> applied in three weeks' application time. In other words, use of 75 NPS: 75 N kg ha<sup>-1</sup> gave 14.13 ETH birr of return per one birr investment. Evidently, application of urea is beneficial to farmers as the expected net return and the marginal rate of return were so attractive to invest more at the time of the dry season (Tesfaye *et al.*, 2015; Nebi, 2018).



Table 6: Partial budget analysis of the effects of fertilizer rate and time of application on natural grass

FR kg ha <sup>-1</sup> (NPS:N)	TA (week)	HDMY (t/ha)	GFB (ETB ha <sup>-1</sup> )	TVC (ETB ha <sup>-1</sup> )	GI (ETB ha <sup>-1</sup> )	NB (ETB ha <sup>-1</sup> )	MRR %
0	0	6.87	41636.36	0.0	41636.4	41636.4	0.0
25:25	1	8.09	49030.30	1232.0	49030.3	47798.4	500.2
25:25	3	8.75	53030.30	1232.0	53030.3	51798.4	824.9
25:25	5	7.95	48181.82	1232.0	48181.8	46949.9	D
50:50	1	9.54	57818.18	2163.9	57818.2	55654.3	647.8
50:50	3	10.30	62424.24	2163.9	62424.2	60260.3	860.7
50:50	5	9.67	58606.06	2163.9	58606.1	56442.2	D
75:75	1	12.77	77393.94	3095.8	77393.9	74298.1	1055.0
75:75	3	14.60	88484.85	3095.8	88484.8	85389.0	1413.3
75:75	5	13.22	80121.21	80121.21	80121.2	77025.4	D
100:100	1	14.09	85393.94	4027.8	85393.9	81366.1	D
100:100	3	15.32	92848.48	4027.8	92848.5	88820.7	1171.5
100:100	5	13.18	79878.79	4027.8	79878.8	75851.0	D

**Note:** FR=Fertilizer rate; TA=Time of application; HDMY = Herbage dry matter yield; GFB = Growth field benefit; TVC= Total Variable cost; GI= Growth income; NB= Net Benefit; NI= net income; CTVC= Change in total variable cost; MRR = marginal rate of return; Market price of Natural grass = 2 ETB·kg<sup>-1</sup>; cost of urea = 1669.8 ETB·qt<sup>-1</sup>; cost of NPS = 2058 ETB·qt<sup>-1</sup>; labor cost for fertilizer application = 2 persons ha<sup>-1</sup>, each 75 ETB·day<sup>-1</sup> for one time application; ETB = Ethiopian birr.

### Conclusion and Recommendations

Application of increased rates of fertilizers on natural pasture considerably enhanced herbage dry matter yields, nutritional quality and economic benefit. The maximum herbage DM yield was 15.32 t/ha obtained from 100 NPS: 100N kg ha<sup>-1</sup> combined fertilizer applied at the third weeks' application time and statistically similar yield was recorded (14.6 t/ha) from the combination of 75 NPS: 75 N rates at same application time. Moreover, application of inorganic fertilizer at different rates affected grass-legume species composition. The partial budget analysis revealed that combined applications of 75 NPS: 75 N kg ha<sup>-1</sup> during third weeks' of application time gave the paramount economic benefit of 85,389 Birr·ha<sup>-1</sup> with best MRR of 1413.3%, followed by fertilizer rate of 100 NPS: 100 N kg ha<sup>-1</sup> at same time of application, which gave higher 88820.7Birr·ha<sup>-1</sup> with a MRR of 1,171.5%. Therefore, it can be concluded that use of combined fertilizer at a rate of 75 NPS: 75 N during the third weeks' time of application after main rain fall optimize both biological and economic benefits than other treatments. So, it can be recommended for farmers for natural grass land improvement in the study sites and other areas with similar agro-ecological conditions.

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# Evaluation of Rhodes Grass (*Chloris gayana*) Accessions for Agronomic and Biomass Yield at Adola district, Guji Zone of Oromia

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## Abstract

*The study was conducted at Adola district of Guji zone with the objectives to identify adaptable and high yielding Rhodes grass accessions for the study area. Accordingly, five Rhodes grass accessions (Masaba, ILRI-6633, ILRL-7384, ILRL-13325 and Dz-253) were evaluated in randomized complete block design (RCBD) with three replications. All agronomic parameters and biomass yield of forage samples were collected and the data were examined using statistical analysis. The result indicated that dry matter and seed yield were showed statistically significant variation ( $P < 0.05$ ) among the Rhodes grass accessions. . The highest herbage dry matter yield was recorded from ILRI-7384 (15.44 t/ha) followed by ILRI 6633 (14.9 t/ha) accession. The maximum seed yield (2.66 qt/ha) were recorded from ILRI- 6633 as well as from ILRI-13325 followed by ILRI-7384 (2.5qt/ha) accession. Hence, due to better biomass and seed yield performances; ILRI-7384 and ILRI- 6633 accessions were recommended for the study area as livestock feed resources.*

**Key Words:** Rhodes Grass, Dry Matter Yield, Seed Yield, Masaba

## Introduction

Rhodes grass (*Chloris gayana*) is a perennial or annual tropical leafy grass 1-2 m in height, highly variable in habit. It is now widespread in tropical and subtropical areas worldwide. It grows better in areas where with an altitude ranges from 1400 – 2400 m.a.s.l (Ecocrop, 2014). It grows on a wide range of soils from poor sandy soils to heavy clayey alkaline and saline soils. Rhodes grass grows better on fertile, well-structured soils and it prefers soil pH between 5.5 and 7.5; even if, establishment on acidic soils is challenging. Rhodes grass thrives in places where annual temperatures range 25 to 30°C (day/night temperature). Optimal annual rainfall ranges between 600-750mm with a summer-rainfall period (Moore, 2006; Ecocrop, 2014). The culms are tufted or creeping, erect or decumbent, sometimes rooting from the nodes. The inflorescences are light greenish brown (rarely yellow) in color and turn darker brown as they mature (Cook *et al.*, 2005). Due to its deep roots, Rhodes grass can withstand long dry periods (over 6 months) and up to 15 days of flooding (Cook *et al.*, 2005; FAO, 2014).

Growth performance of Rhodes grass varies with type of accessions, age of plant and other environmental factors (Akinola *et al.*, 1990). Rhodes grass productivity generally ranges from 7 - 25 tons of DM/ha per year, depending on variety, soil fertility, environmental conditions and cutting frequency. Based on a study conducted on farmers' fields in the central highlands of Ethiopia, on average the herbage yield of Rhodes grass was from 8.74 to 9.1 tons DM/ha per year on rain-fed conditions (Cook *et al.*, 2005; CASCAPE, 2015; HARC, 2004). In spite of the importance of Rhodes grass as feed sources, there is limited information on Rhodes grass adaptation performance in the study area. Hence, the intention of

this study was to select and recommend adaptable and high biomass Rhodes grass accessions for the study area and similar agro-ecologies.

## **Materials and Methods**

### ***Description of the Study Area***

The experiment was carried out at Adola sub-site of Bore Agricultural Research Center, Adola district, Guji Zone of Oromia. Adola district is located at a distance of 470 km from Addis Ababa and 120 Km from the zonal capital city, Negele Borena. It is an area where mixed farming and semi-nomadic economic activity takes place, which is the major livelihood of the local people. The total area of the district is 1254.56 km<sup>2</sup>. The district is situated at 5°44'10" - 6°12'38" N latitudes and 38°45'10" - 39°12'37" E longitudes. The district is characterized by three agro-climatic zones, namely highland 11%, mid-land 29% and low-land 60% respectively. The major soil type of the district is tools (red basaltic soils) and orthic Acrisols.

### ***Experimental Treatments and Design***

The experiment was conducted using five Rhodes grass accessions; Masaba, ILRI-7384, DZ-253, ILRI-6633 and ILRI-13325. The experiment was conducted in RCBD with three replications. Splits of plant tiller were planted in rows plot size of 4m x 3m at 1m between plot and 1.5m between replication. Other agronomic practices were applied uniformly as per recommendation.

### ***Data Collection***

All relevant data including days to emergence, days to flowering, plant heights, leaf to stem ratio, biomass yield and seed yield were recorded. Plant height was recorded based on five plants selected randomly in each plot, measured using a steel tape from the ground level to the highest leaf. For determination of biomass yield, the grass were cutting at 5-10 cm from the ground level from two central rows. Consequently, fresh biomass yield was collected and calculated by selecting 3 middle lines and cutting at a stubble height of 5 cm and weighing to find out the weight of fresh biomass matter (Yvan and Tessema 2007). In order to measure dry matter yield, the harvested fresh sample was measured right in field by sensitive weight balance and 300g subsample per plot was brought to Bore Agricultural Research Center and the sample was placed to oven dried for 72 hours at a temperature of 65°C for dry matter determination. Then dry matter yield (t/ha) was calculated by James (2008) formula.

**The dry matter yield (t/ha) = TFW × (DWss /HA × FWss) ×10 where,**

Where, TFW = total fresh weight kg/plot, DWss = dry weight of subsample in grams, FWss = fresh weight of subsample in grams, HA = Harvest plot area in square meters and 10 is a constant for conversion of yields in kg/m to t/ha.

The leaf to stem ratio was calculated by weighing the leaf and stem parts after oven dried for 72 hours at a temperature of 65°C.

### ***Statistical Analysis***

All collected data were analyzed using the general linear model procedure of SAS (SAS 2002) version 9.1. The statistical model for the analysis data was:

$$Y_{ijk} = \mu + A_j + B_i + e_{ijk}$$

Where;  $Y_{ijk}$  = response of variable under examination,  $\mu$  = overall mean,  $A_j$  = the  $j$ th factor effect of treatment/ accessions,  $B_i$  = the  $i$ th factor effect of block/ replication,  $e_{ijk}$  = the random error.

### **Result and Discussions**

The analysis result indicates that days to emergence is significantly differ ( $P < 0.05$ ) among the accessions. The tested Rhodes grass accessions required about 8.33 to 10.4 days for first emergence which is almost similar with the study conducted by Mohamed and Gebeyew (2018) who recorded 10.66 days for the first days of emergence. According to Cook *et al.* (2005) report, Rhodes grass germinates at 7.0 days after planting. This difference might be attributed to soil moisture content, soil fertility and other environmental factors.

The days to 50% flowering was significantly ( $P < 0.01$ ) varied among the tested accessions. The shortest (72.67) days to 50% flowering was recorded from ILRI-6633 accessions. The longest (82.0) days to 50% flowering was recorded from Masaba accession. The difference in days to 50% flowering among treatments/accessions could be attributed to genetic variation among accessions and their interaction with the environment.

The analysis data revealed that dry matter yield was significantly differ ( $P < 0.05$ ) among the tested accessions. Accordingly, the highest dry matter yield was recorded from ILRL-7384 (15.44 t/ha) accession followed by ILRI- 6633 (14.9 t/ha) accession. This result is higher than the finding of other scholars (Cook *et al.*, 2005, CASCAPE, 2015 and HARC, 2004) who reported the dry matter yield of 8.74 to 9.1 t/ha from Rhodes grass at rain-fed conditions. The biomass yield obtained in this study is also comparable with the yield (7.8 to 9.16 tons DM/ha-1 per year without manure application) obtained from study conducted at Adami Tulu Agricultural Research Center (ATARC) and Negele Arsi Farmers Training Center (FTC) by Tesfaye *et al.* (2020).

Mean seed yield recorded was also significantly varied ( $P < 0.01$ ) among the tested accessions. The maximum (2.66 qt/ha) seed yield value was recorded from ILRI-6633 and ILRI-13325 accessions (Table 1). This result was lower than the value reported by (Tesfaye *et al.*, 2020) in which mean seed yield values of 370.55 and 313.88 Kg/ha recorded. This variation in seed yield might be due the difference in soil fertility and soil moisture factors. On the other hand, climate and soil types or their interactions have effects on the performance of forage crops as indicated by Diriba *et al.* (2014).

Table 1. Mean agronomic, biomass and seed yield of Rhodes grass accessions tested at Adola site of Guji zone

Varieties	DE	DF	PH (cm)	LSR	DMY (t/ha)	SY qt/ha
ILRI-7384	8.33 <sup>b</sup>	77.0 <sup>b</sup>	164.7	0.59	15.44 <sup>a</sup>	2.50 <sup>ab</sup>
Masaba	10.67 <sup>ab</sup>	82.0 <sup>a</sup>	171.0	0.56	13.80 <sup>ab</sup>	0.92 <sup>b</sup>
ILRI-13325	10.0 <sup>ab</sup>	77.6 <sup>b</sup>	171.7	0.76	9.73 <sup>b</sup>	2.66 <sup>a</sup>
DZ-253	10.33 <sup>ab</sup>	78.3 <sup>ab</sup>	181.7	0.86	12.83 <sup>ab</sup>	1.83 <sup>ab</sup>
ILRI-6633	12.67 <sup>a</sup>	72.67 <sup>c</sup>	171.7	0.80	14.90 <sup>a</sup>	2.66 <sup>a</sup>
Mean	10.4	77.5	172.1	0.66	13.34	2.12
CV	13.5	2.8	5.5	26	18.8	39.7
Sig. level	*	**	Ns	ns	*	**

DE=days to emergence, DF=days to flower, PH=plant height, LSR=leaf to stem ratio, DMY=dry matter yield, SY=Seed yield, CV=coefficient variation, ns=non-significant, \*=significant, \*\* highly significant

### Conclusion and Recommendations

The result of this study indicated the highest herbage dry matter yield was recorded from Rhodes grass with accession ILRI-7384 followed by ILRI 6633. Similarly the maximum seed yield were recorded from ILRI- 6633 as well as from ILRI-13325 followed by ILRI-7384 accessions. Hence, due to better biomass and seed yield performances; ILRI-7384 and ILRI- 6633 accessions, were recommended for the study area as livestock feed resources.

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# Effect of Seeding Ratios of Alfalfa (*Medicago sativa*) and Rhodes Grass (*Chloris gayana*) Mixtures on Dry Matter Yield and Nutritive Quality of the Fodder

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## Abstract

*The study was conducted at Adami Tulu on-station and Shashemene (FTC) sites for two (2019-2020) consecutive years with the objective to determine the appropriate proportions of seeding ratio that could optimize the dry matter yield and quality of Alfalfa and Rhodes grass mixture. The treatment was laid out in a randomized complete block design (RCBD) with three replications. Different seeding ratios of Alfalfa and Rhodes grass were arranged according to the following treatments: T1; 100% Alfalfa + 0 % Rhodes, T2; 0 % Alfalfa +100 % Rhodes, T3; 50% Alfalfa + 50% Rhodes, T4; 75 % Alfalfa + 25% Rhodes and T5; 25% Alfalfa + 75% Rhodes. The results showed that number of tiller per plant of Rhodes grass and leaf to stem ratio of alfalfa, CP% content, NDF% and ADF% were showed a significance ( $p<0.05$ ) differences among the seeding ratios. The result indicated that seeding ratio has significant ( $p<0.05$ ) effect on the total dry matter yield of the mixture of alfalfa and Rhodes at both sites. The highest dry matter yield ( $8.47 \text{ t ha}^{-1}$ ) was obtained from seeding ratio of 25:75, followed by seeding ratio of 50:50 ( $7.84 \text{ t ha}^{-1}$ ). The land equivalent ratios for the mixture were more than one showing that mixture of alfalfa with Rhodes is more advantageous than pure stand of forage. The highest CP% content (21.2%) was recorded from pure stand of alfalfa followed by seeding ratio 75:25 (18.5%), 50: 50 (16.8%) and 25:75 (14.6%) while pure stand of Rhodes grass produced the least (11.1%) CP% value. The highest values of NDF% (34.48%) and ADF% (21.25%) were obtained from pure stands of Rhodes grass while the least values were recorded from pure Alfalfa. Thus, it can be concluded that seeding ratio combination of 50:50 could be recommended for use in the study areas and similar agro-ecologies due to its high dry matter yield, good quality and more balanced mixture of forage.*

**Key words:** Seeding ratio, Rhodes, Alfalfa, Mixture

## Introduction

Livestock production is an integral part of the agriculture in Ethiopia. It serve as sources of food, traction, manure, raw materials, investment, cash income, security, foreign exchange earnings and social and cultural identity. Despite enormous contribution of livestock to the livelihood of farmers, they are faced with multifaceted problems in the production system, among which the major one is the quantitative and qualitative inadequacy of feed supply (Manaye *et al.*, 2009).

In most areas of the country, livestock feed is normally in short supply and is also of poor quality mainly during the latter part of the dry season. According to Gelayenew *et al.* (2016) and Ulfina *et al.* (2013), seasonal feed shortage and inefficient utilization practices have been identified as the major problems affecting livestock production and productivity. Nowadays the available feed from natural pasture is very low due to overgrazing, erosion and overall land degradation. Moreover, in most areas there is an ever-increasing human population with subsequent increase in land area put under crop production resulting in

reduction of existing grazing areas (Tolera *et al.*, 2012). This suggests the need for improving the quantity and quality of the feed resource. Accordingly many grass and legume species have been tested and recommended for different agro-ecological zones in the country. To bring further improvement in yield and quality of forage species and land use efficiency, identifying appropriate forage production strategies are very crucial especially at smallholder farmers' level where land for forage production is very scarce.

Grass legume mixture forage production has many advantages including maximize yield, improve growth, produce palatability, supply the soil with nitrogen by legumes, make a better soil coverage and keep it from erosion, compete weeds, attained a balanced and highly nutritive feeding to animals and decrease animals bloats (Karadau, 2003, Aslaug *et al.*, 2018). The acceptance of these forage mixtures is based on the apparent advantages offered by the association of different species that influence the performance temporal stability of yield and forage quality. Species that have different physiological and/or morphological characters may complement better and make better use of the nutritional resources. Among the grass species, Rhodes grass (*Chloris gayana*) recommended for low to high elevations. Similarly, Alfalfa is one of the recommended forage legume with high potential as fodder crop. Alfalfa can grow in mixture with most of the grasses, but a balance of the alfalfa with the grass is best maintained if only an appropriate agronomic management is practiced. It also indicated that the establishment of Rhodes-Alfalfa mixture showed positive effect and compatible on their growth characters with each other's (Atif *et al.*, 2012).

However, management practices such as proportions of seeding rate influences the yield and quality of these species and their compatibility when grown in mixtures (Atis *et al.*, 2012). The benefits from mixed forage species can be efficiently exploited only if proper management strategies such as optimum proportions of seed rates are used. However, there is limited information on the proportions of seeding rates that influence yield and quality of most of the grass-legume mixture including Rhodes grass and Alfalfa under rift valley condition of Oromia. Hence, this study was intended to determine the appropriate proportions of seeding rates of Rhodes and Alfalfa that could optimize dry matter yield and quality of the mixtures.

## **Materials and Methods**

### ***Description of the study sites***

The study was conducted at Adami-tulu Agricultural Research Center (on-station) and Shashemene (FTC) under rain fed conditions. Adami Tulu Agricultural Research Center is found in Adami Tulu Jido Kombolcha district that located in the middle rift valley of Ethiopia, at 167 kilometers from the capital city of the country, in south eastern part of Oromia between 38°20' and 38.5°5' E and 7°35' and 8°05' N. It lies at altitudinal range from 1500 to 2000 masl. It has an average annual rain fall of 760mm. It has a bimodal rain fall from March to April (short rain) and July to September (long rains) with a dry period in May to June which separates short rains from long rains. The average annual minimum and maximum temperature of the area at the study year were 11.8°C and 28.3°C (metrology station of Adami Tulu Agricultural Research Center). The soil is loam with sand, silt and clay in proportion of 44%, 34% and 22% respectively and the PH of the soil is 7.88 (Teshome *et al.*, 2012). Shashamane district is found in West Arsi Zone, Oromia Region at about 240km south of Addis Ababa lying on the main way road to Hawasa. Shashamane town is the capital town and administrative center of west Arsi zone.

Geographically, the area is located at 7° 11' 33" N altitudes and 38° 35' 33" E longitudes. The area has an annual average temperature ranging from 12°C to 28°C. The rainfall ranges from 1500-2000ml (WAZADO, 2014).

### ***Treatments and Experimental Design***

Adapted cultivars of Rhodes grass (*Chloris gayana*) and Alfalfa (*Medicago sativa*) were used. The experiment was undertaken in a randomized complete block design with three replications. Different seeding ratios were arranged according to the following treatments: T1; 100% Alfalfa + 0% Rhodes, T2; 0% Alfalfa +100 % Rhodes, T3; 50% Alfalfa + 50% Rhodes, T4; 75% Alfalfa + 25% Rhodes and T5; 25% Alfalfa + 75% Rhodes. The seed proportions were calculated based on the recommended sole seed rates of 15 and 10 kg per hectare for Rhodes and Alfalfa, respectively. The plot size was 2.5 m x 2 m (5 m<sup>2</sup>). A total of 10 rows per plot with row spacing of 25 cm apart (Atif *et al.*, 2012) from each other was used. The spacing between replications and plots were 1.5 and 1.0 m, respectively. Germination test was done for both forages before sowing in order to adjust the seeding rates. The seeds was established in rows on a well-prepared seedbed and covered with soil. All other cultural practices including weeding was kept normal and uniform for all treatments.

### ***Data collection***

Data on yield and yield related traits such as: plot cover (%), plant height (cm), leaf to stem ratio, number of tiller per plant and biomass yield were collected from each treatments. The forage were harvested for biomass yield determination at 10-50% flowering stage. Then consecutive cuts was conducted after the re-growth while it reach an appropriate harvesting stage. Land equivalent ratio (LER) was used to assess the advantage of forage production in mixture. LER was defined as the relative area of mono-crop plant required for the same yield obtained from its mixture. The LER was calculated using the formula given below (Ta and Faris, 1987):

$$\text{LER} = \frac{\text{Yield of alfalfa in mixture}}{\text{Yield of alfalfa alone}} + \frac{\text{Yield of grass in mixture}}{\text{Yield of grass alone}}$$

Where, LER is greater than 1, the mixed growing favors the growth and yield of the mixture species. In contrast when LER is lower than 1, the mixed growing negatively affects the growth and yield of plants grown in mixture (Caballero *et al.*, 1995; Dhima *et al.*, 2007).

The feed sample was taken from each treatment and dried in an oven at 60°C for 72 hours to a constant weight and ground in a Willey mill to pass through a 1mm sieve. The ground samples were kept in airtight plastic bags prior to analysis for chemical composition. Crude protein (CP) was determined according to AOAC (2000) methods. The dry matter (DM) was determined by an oven drying at 105° overnight and ash content was determined by igniting the dry samples in a muffle furnace at 550°C for 6 hours to burn off all the organic material. The inorganic material which does not volatilized at that temperature is ash. The difference between sample DM and ash gives the organic matter (OM). Neutral detergent fiber (NDF) and acid detergent fiber (ADF) were analyzed according to Vansoest and

Robertson (1985). For nitrogen (N) analysis, the Kjeldhal method was used and crude protein (CP) content was estimated from the N content by use of a multiplier of 6.25. Collected data was organized, summarized and analyzed by using SAS. LSD test at 0.05 probability levels was used to compare the treatment means (Steel and Torrie, 1984).

## Results and Discussion

Analysis of variance showed that different seeding ratio of alfalfa and Rhodes grasses were significantly ( $p < 0.0016$ ) affected the total dry matter of the mixture. Experimental sites were also significantly ( $p < 0.0001$ ) influenced the total biomass yield of the mixed alfalfa and Rhodes biomass production. On the other hands, the result showed that interaction effect of site and the treatments were didn't significantly ( $p > 0.1298$ ) affected the total dry matter production of the mixture.

Table 1: Mean squares of ANOVA for total dry matter yield of Alfalfa and Rhodes mixture under different seeding ratios

Source of Variation	DF	Mean squares	F-value	p-value
Replication	2	1.771	0.97	0.3868
Treatment	3	9.391	5.14	0.0016
Site	1	48.533	26.56	0.0001
Treatment*Site	3	3.429	1.88	0.1298
Error	38			
Total	47			

Where: DF= degree of freedom,

### *Agronomic parameters*

The combined analysis result for plot cover, plant height, number of tiller per plant and leaf to stem ratio were indicated in table 1. The results showed that non-significant ( $p > 0.05$ ) effect was observed on plot cover and plant heights for both fodder among the tested seeding ratios. However, numerically the maximum plot cover (87.5%) was recorded from 50:50 seeding ratio followed by sole Rhodes (85.2%) treatment. In this trial the plant height of alfalfa grown alone was statistically similar ( $p > 0.05$ ) to those alfalfa grown in a mixture. Similarly, plant height of Rhodes grass were not significantly differ among sole and mixture treatments. The non-significance ( $p > 0.05$ ) differences observed for alfalfa and Rhodes plant height illustrate the mutual benefits of the forage crops from mixed establishment.

The analysis also showed non-significant ( $p > 0.05$ ) difference on number of tiller pre plant of alfalfa in the tested seeding ratios. On the other hands, the number of tiller per plant of Rhodes grass was showed a significance ( $p < 0.05$ ) differences among the seeding ratios. Accordingly the highest number of tiller per plant of Rhodes grass was recorded for sole Rhodes grass (22.44) followed by 25:75 (19.15) seeding ratio. As seeding ratio of alfalfa increases in the mixture, number of tiller of Rhodes grass were significantly decreases. This might be due to the slight competition of alfalfa and Rhodes for light and nutrients. The result can be relatively supported by other authors (Ahmed A, 2010). The result also showed the values of leaf to stem ratio (LSR) of alfalfa was significantly ( $p < 0.05$ ) differ among the tested seeding ratios. The highest LSR value (2.49) was recorded for sole alfalfa treatment followed by 50:50 (2.23) and 75:25 (2.23) seeding ratios. The highest value obtained for sole alfalfa could be due to the

better vegetative growth as it had free from competition with other companion crops. The leaf to stem ratio values were higher than one shows that the leaf component was higher in proportion than the stem, which is should be taken as reference for the quality of alfalfa as fodder crops (Rojas *et al.*, 2019). On the other hands, the leaf to stem ratio for Rhodes grass was not significantly ( $p>0.05$ ) differ among the treatments.

Table 1. Agronomic performance of Rhodes and Alfalfa produced from different seeding rate proportions (combined)

Seeding ratio Alfalfa: Rhodes	Plot cover (%)	Plant height (cm)		No of tiller/plant		Leaf to stem ratio	
		Alfalfa	Rhodes	Alfalfa	Rhodes	Alfalfa	Rhodes
100:0	83.9	68.16	-	15.36	-	2.49 <sup>a</sup>	-
0:100	85.2	-	103.75	-	22.44 <sup>a</sup>	-	2.51
50:50	87.5	66.18	103.0	14.44	17.49 <sup>b</sup>	2.23 <sup>ab</sup>	2.33
75:25	83.8	65.13	100.22	14.88	17.58 <sup>b</sup>	2.23 <sup>ab</sup>	2.22
25:75	84.1	63.78	105.88	13.94	19.15 <sup>ab</sup>	2.06 <sup>b</sup>	2.42
Mean	84.9	65.82	103.2	14.66	19.2	2.25	2.37
CV (%)	10.7	15.76	24.3	13.82	24.8	19.0	20.67
LSD (0.05)	NS	NS	NS	NS	3.91	0.35	NS
<i>p-value</i>	0.8457	0.767	0.9575	0.260	0.049	0.0462	0.523

<sup>1</sup> CV=Coefficient of variation, LSD=Least significant difference, NS= Non significant,

<sup>2</sup> Figure having the same letters with in column are not significantly differ, while values followed by different letter (s) are significantly differ

### ***Dry matter yield***

The analysis result of Rhodes grass and Alfalfa biomass yield grown in pure stand and in mixture with different seeding ratios are indicated in table 2. The result shown that seeding ratio has significant ( $p<0.05$ ) effect on the total dry matter yield of pure stand and the mixture of alfalfa and Rhodes at both sites. The highest total forage biomass yield (10.2 t/ha) was recorded from seeding ratio of 25:75 at Adami Tulu site where as 50:50 seeding ratio gives the highest biomass yield (6.85t/ha) at Shashemene site. The lowest dry matter yield was recorded from sole alfalfa treatment at all sites. The combined analysis result also showed that significantly ( $p<0.05$ ) the highest dry matter yield (8.47t/ha) was obtained from seeding ratio of 25:75 and followed by seeding ratio of 50:50 (7.84t/ha). The higher biomass yield obtained from these seeding ratios could be due to the fact that the proportion of Rhodes grass produced much more forage yield through increased number of tillers production than the other mixtures.

The total dry matter yield of sole alfalfa treatment (36.17 t/ha) was significantly lower than the total biomass yield production from seeding ratio of 50:50 (7.84 t/ha) and 25:75 (8.47 t/ha). As the result shown, the biomass of the mixtures were superior and advantageous compared to their pure stands. In line with this study, scholars emphasized on the advantages of grass legume mixture (Kechero Yisehak, 2008; Albayrak and Mevlüt 2011 and Atif *et al.*, 2012). Lüscher *et al.* (2014) also reported that total dry matter

yield of mixed grass and legume pasture was greater than grass-based pasture, and overall feed value was better maintained throughout the grazing season when pastures include legumes.

On the other hands, the lowest performances (6.87 t/ha) of seeding ratio of 75:25 (alfalfa: Rhodes) as compared to the other mixtures could be due to the fact that alfalfa dominate and suppressed the growth of Rhodes grass in terms of light and nutrients utilization. These findings was supported by Atif *et al.* (2012) who reported seeding ratio of 50:50 for alfalfa and Rhodes produced the highest biomass yield. Moreover, the beneficial effects of mixing alfalfa and Rhodes may result from differences in their growth pattern in which Alfalfa is erected, whereas Rhodes grass is semi erected/ prostrate growth characters. This can lead to more efficient use of resources such as light when grown together than when grown separately (Atis *et al.*, 2012). Other findings also indicated that mixtures gave higher green forage yields than the pure stands (Karadau, 2003). This finding is in line with Miguel *et al.* (2012) who reported that higher mean biomass yield was obtained from the mixture as compared to their components grown in monoculture. Generally, the result shows that seeding ratio of 50:50 and 25:75 for alfalfa and Rhodes grass were showed the highest total biomass yields for the Rhodes grass and alfalfa in the intercropping system.

As indicated in the table 2, all alfalfa and Rhodes grass mixtures produced LER values greater than 1.0, indicating that these mixtures produced more DM yield as mixture compared to the yield of pure stands. The higher LER (1.21) was recorded for seeding ratio of 25:75 alfalfa to Rhodes grass followed by 50:50 (1.14). Reasons of the higher LER values for seeding ratio of 25:75 could be due to the better benefiting of the grasses from the fixed nitrogen through the alfalfa. Erkovan (2005) also reported that nitrogen transfer from the fixed nitrogen by legumes to the grasses in the legume-grass mixtures.

On the other hands, the lower LER (1.01) for 75:25 seeding ratio could be due to the fact that alfalfa created competition effect on Rhodes grass and thus influence the growth and yield than the other combinations. Intercropping systems that constantly give LERs greater than one are considered to be more efficient systems from a land use point of view than mono-crops. In addition, legume-grass mixtures generally provide more consistent and greater forage yields across a range of environments than grass or legume monocultures (Papadopoulos *et al.*, 2012). Hence, the value of LER for seeding ratios showing that mixture of the alfalfa and Rhodes are more advantageous than sole cropping.

Table 2. Biomass yield (t/ha) and land equivalent ratio of Alfalfa and Rhodes mixture under different seeding ratios

Seeding ratio Alfalfa: Rhodes	Adami Tulu			Shashemene			Combined			LER
	Alfalfa	Rhodes	Total	Alfalfa	Rhodes	Total	Alfalfa	Rhodes	Total	
100:0	6.46 <sup>a</sup>	-	6.46 <sup>c</sup>	5.88 <sup>a</sup>	-	5.88 <sup>b</sup>	6.17 <sup>a</sup>	-	6.17 <sup>c</sup>	-
0:100	-	8.4 <sup>a</sup>	8.4 <sup>abc</sup>	-	6.5 <sup>a</sup>	6.50 <sup>ab</sup>	-	7.45 <sup>a</sup>	7.45 <sup>abc</sup>	-
50:50	3.26 <sup>b</sup>	5.57 <sup>bc</sup>	8.84 <sup>ab</sup>	2.43 <sup>c</sup>	4.4 <sup>b</sup>	6.85 <sup>a</sup>	2.85 <sup>bc</sup>	4.99 <sup>b</sup>	7.84 <sup>ab</sup>	1.14
75:25	3.56 <sup>b</sup>	3.87 <sup>c</sup>	7.44 <sup>bc</sup>	3.1 <sup>b</sup>	3.2 <sup>c</sup>	6.31 <sup>ab</sup>	3.32 <sup>b</sup>	3.55 <sup>c</sup>	6.87 <sup>bc</sup>	1.01
25:75	2.95 <sup>b</sup>	7.22 <sup>ab</sup>	10.2 <sup>a</sup>	2.2 <sup>c</sup>	4.55 <sup>b</sup>	6.77 <sup>a</sup>	2.58 <sup>c</sup>	5.88 <sup>b</sup>	8.47 <sup>a</sup>	1.21
Mean	4.06	6.26	8.26	3.41	4.67	6.46	3.73	5.47	7.36	-
CV (%)	17.9	26.39	21.7	14.83	10.0	9.97	18.52	27.1	22.66	-
LSD (0.05)	0.87	2.0	2.13	0.61	0.56	0.77	0.56	1.21	1.36	
<i>p</i> -value	0.0001	0.0007	0.0172	0.0001	0.0001	0.0214	0.0001	0.0001	0.0156	

<sup>1</sup> LER=Land equivalent ratio, CV=Coefficient of variation, LSD=Least significant difference,

<sup>2</sup> Figure having the same letters with in column are not significantly differ, while values followed by different letter (s) are significantly differ

### ***Chemical Composition***

The analysis result of nutrient contents of Alfalfa and Rhodes with different seeding ratios combinations are indicated in table 3. The result indicated that the combinations of different seeding ratios didn't significantly ( $p>0.05$ ) influenced the percent of dry matter and ash content of the feeds. The DM content of sole Rhodes grass found in the present study was lower than the findings of Adugna (2008), who reported DM percent of 92.3%. On the other hand, the different seeding ratios of Alfalfa and Rhodes mixture had showed a significant ( $p<0.05$ ) variation among the values of CP%, NDF% and ADF%. The highest CP content (21.2%) was recorded from pure stand of alfalfa followed by seeding ratio 75:25 (18.5%), 50: 50 (16.8%) and 25:75 (14.6%) while pure stand of Rhodes grass produced the least (11.1%) CP value. The average values of CP content were directly related to the Alfalfa seeding ratios in the mixture. The CP content of the Rhodes-Alfalfa produced in mixture is much improved due to alfalfa component in the mixture. Studies also indicated that production of grass- legumes mixture increased fresh fodder yield and protein contents as well as enhanced fodder palatability (Karadau, 2003 and Ahmed A., 2010). Even though the value of CP content in 75:25 seeding ratio was higher than other seeding ratios in the mixtures, the high proportion of alfalfa in this ratio is undesirable since these normally have a low dry matter yield.

Of the tested seeding ratios, pure stand of Rhodes grass had the highest NDF (34.48%) and ADF (21.25%) content followed by the seeding ratios of 25:75 which produced 34.28% and 17.24% of NDF and ADF, respectively. The lowest value of NDF (23.8%) and ADF (13.2%) content were recorded from pure stand of alfalfa. NDF and ADF concentrations in forage were directly proportional to the grass percentage in the mixture. As the seeding ratios of the Rhodes grass increased in the mixture, the content of NDF and ADF were increased. Bosworth and Cannella (2007) showed significant positive correlations between legume inclusion in pastures and forage quality traits like crude protein and negative correlations with NDF and ADF.

Table 3. Chemical composition of Alfalfa and Rhodes grass feeds under different seeding ratios

Seeding ratio Alfalfa: Rhodes	DM%	Ash%	CP%	NDF%	ADF%
100:0	88.98	14.76	21.2a	23.8b	13.2c
0:100	89.9	14.23	11.1d	34.48a	21.25a
50:50	89.6	14.47	16.8b	32.93a	16.6b
75:25	89.3	14.83	18.5b	28.59a	14.55bc
25:75	89.78	14.50	14.6c	34.28a	17.24b
Mean	89.5	14.56	16.4	30.88	16.57
CV (%)	0.38	3.81	10.3	21.3	15.86
LSD (0.05)	NS	NS	2.0	7.82	3.12
<i>p-value</i>	0.0864	0.3578	0.0001	0.0387	0.0002

<sup>1</sup>CV=Coefficient of variation, LSD=Least significant difference, NS= Non significant,

<sup>2</sup>Figure having the same letters with in column are not significantly differ, while values followed by different letter (s) are significantly differ

### Conclusions and Recommendation

The study indicated seeding ratios significantly influenced the biomass yield of alfalfa and Rhodes mixture. The land equivalent ratio for the mixture was more than 1 showing that mixture of alfalfa with Rhodes is more advantageous than pure stand of the forage. Seeding ratio of 50:50 and 25:75 for alfalfa and Rhodes grass had produced better total biomass yields of the mixture. The highest CP content was recorded from pure stand of alfalfa followed by seeding ratio of 75:25, 50:50 and 25:75 for alfalfa and Rhodes. Even though the association of 25:75 seeding ratio accounted for the highest dry matter yield, it produced lower CP and high NDF and ADF contents as compared to the other mixture. On the other hands, the treatments with sole alfalfa and seeding ratios of 75:25 were showed the highest CP contents which are undesirable due to low total dry matter yields the treatments. Among the seeding ratios, 50:50 combinations produced the maximum dry matter yield with optimum nutritive quality of alfalfa-Rhodes mixture. Thus, it can be concluded that seeding ratio combination of 50:50 could be recommended for use in the study areas and similar agro-ecologies due to its high dry matter yield, good quality and more balanced mixture of forage.

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# Determination of Optimum Sowing Dates of Dolichos lablab Intercropping with Sorghum in West Hararghe, Oromia, Ethiopia

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## Abstract

*In Hararghe as whole, intercropping is the main and indigenous activity of the farmers due to land shortage that most farmers are practicing intercropping of different crops for different reasons like to minimize total crop failure and efficient land utilization. An experiment was conducted to evaluate the influence of sowing dates of Dolichos lablab intercropping with sorghum on sorghum yield and yield related and to identify the optimum sowing date of Dolichos lablab intercropping with sorghum in West Hararghe Zone, Oromia, Ethiopia. Six treatments, namely: sole lablab, sole sorghum, Sorghum + lablab simultaneous, Sorghum + lablab after two weeks, Sorghum + lablab after four weeks and Sorghum + lablab after six weeks were used with 3 replications in a randomized complete block design. Significant ( $p < 0.05$ ) variation were observed between 50% flowering, plant height, stand count at harvest, sorghum yield, and plot cover but has no significant ( $p > 0.05$ ) in terms of sorghum maturity and diseases incidence. The highest mean sorghum seed yield (20.61qt/ha) was obtained from sole sorghum followed by sorghum + lablab after four weeks intercropped while the lowest seed yield (10.82qt/ha) was recorded from Sorghum + lablab simultaneous intercropped. The study showed that sorghum-lablab intercropped at different sowing date has influence on grain yield of sorghum that grain yield of sorghum increases as sowing date of lablab increases. The ideal acceptable LER was produced from sorghum + lablab intercropped after four weeks. The highest mean grain yield of lablab yield (19.9 qt/ha) were recorded from sorghum + lablab simultaneous intercropping whereas the lowest mean grain yield (4.2 qt/ha) was recorded from sorghum + lablab after six weeks intercropped. The largest lablab dry matter yield (2.31 t/ha) was recorded from sorghum + lablab after four weeks whereas the lowest (2.15 t/ha) was recorded from sorghum + lablab after six weeks intercropped. Therefore, it recommended that four week after sown of sorghum is the appropriate date of sorghum-lablab intercropping.*

**Key words:** - grain yield, influence, intercropping, lablab, sowing date and sorghum

## 1. Introduction

In Ethiopia, the dominant farming system is a crop-livestock system (Assefa *et al.*, 2016), in which both crop and livestock production are economically important. In the country, natural pasture is the primary feed source, which has low biomass yield and nutritional value because of mismanagement (CSA, 2018).

Intercropping legumes into grass pastures have proven to be a viable means to mitigate the decline in quantity and quality of grass forages (Ibrahim, 1996). It has been reported that intercropping of grasses with legumes increased yield, improved growth, enhanced palatability and nutritive quality feeds for

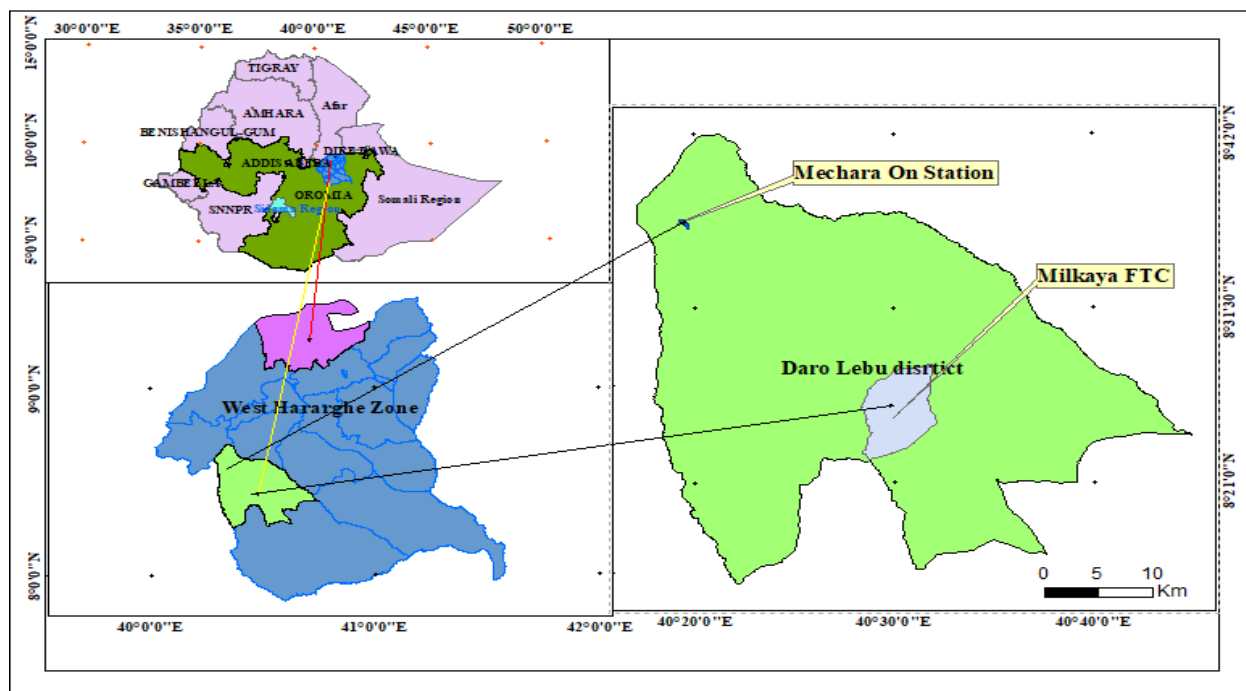
animals (Ibrahim, 2005). The advantages of intercropping forage legumes into farming system include the transfer of nitrogen to the component cereal crops thereby, increasing the crude protein content of the grasses (Tanko, 2004). Lablab (*Lablab purpureus*) is a creeping legume which produces high nutritive quality of conserved feed in form of hay or silage (Amodu *et al.*, 2003). Cereals –legumes cropping system is the most used by small scale farmers in Sub Saharan Africa because of their compatibility. Intercropping systems help farmers to exploit the principle of diversity, they are helpful to avoid reliance on a single crop and result in a variety of products of a different nature such as forages, oil and pulses. Another key advantage associated with intercropping is its potential to increase the land productivity per unit area and the efficient utilization of farm resources (Mucheru-Muna *et al.*, 2010).

In Hararghe as whole, intercropping is the main and indigenous activity of the farmers due to land shortage. Most of the farmers are practicing intercropping of different crops for different reasons like to minimize total crop failure and efficient land utilization (Birmaduma *et al.*, 2018). Therefore, this experiment was initiated with to evaluate the influence of sowing dates of *Dolichos lablab* intercropping with sorghum on sorghum yield and yield related and to identify the optimum sowing date of *Dolichos lablab* intercropping with sorghum in West Hararghe Zone

## **MATERIALS AND METHODS**

### ***Description of Study Area***

The study was conduct at Mecahara Agriculture Research Center on station and Milkaye FTC, Daro labu district, West Hararghe Zone during 2019/2020 to 2020/2021. Experimental sites are located at a distance of 434 km and 474 km to the east of capital city, Addis Ababa and from Chiro (Zonal Capital city) is 111km and 151km respectively. Daro Labu district is located at latitude of 40°19.114 north and longitude of 08°35.589 east. The district has altitude ranges from 1350 to 2450 m.a.s.l. The nature of rainfall is very erratic and unpredictable causing dangerous erosion. The area has a bimodal rain fall type ranging from 900 to 1300mm average of 1094mm. The ambient average temperature was 20 °C and the major soil type of the area is sandy clay loam which is reddish in color. The predominant production system in the district is mixed livestock-crop production system. The crops that growths in study area are cereals such as maize, sorghum, haricot bean, *teff* to large tree fruits like mango, banana, and Avocado especially coffee is the brand crop of the study area known as Hararghe coffee spatiality



Map of the study area, Source of the map: Ethio-GIS shape file, 2016

### ***Treatments and Experimental Design***

The experiment was conducted for two consecutive years with six treatments using a randomized complete block design (RCBD) with three replications. The treatments arrangements were sole lablab, sole sorghum, Sorghum + lablab simultaneous, Sorghum + lablab after two weeks, Sorghum + lablab after four weeks and Sorghum + lablab after six weeks. One sorghum variety (Dagim) and one Dolichus lablab (Dole-1) variety were used with plot size of 3m\*2.75m and the space between block, plot and rows were 1m, 1m and 0.55m respectively for sorghum. Dolichus lablab was sown between the rows of sorghum with 22.50cm distance from the sorghum for all sowing times and each plot has five rows for sorghum and four rows for lablab. All sorghum treatments were sown on the same date of sowing and on the other hand, sole lablab was sown on the second weeks of the treatments. Sorghum was sown at the seed rate of 15kg/ha whereas lablab was sown at the seed rate 10kg/ha through drilling techniques and thinned to 25cm and 10cm space between plants for sorghum and lablab respectively. Fertilizer application was done at the time of sowing for all treatments with the rate of 100kg/ha and 50kg/ha NBS and UREA respectively. Land preparation was done manually and every management was applied for all treatments uniformly.

### ***Data Collection Methods***

Regarding sorghum, data of 50% flowering and number of days to maturity were collected as days after emergency and data of plot cover, stand vigor, plant height, stand count at harvesting and grain yield was collected for all plots. The yield was adjusted and weighted at 12.5% of moisture content to calculate actual seed yield per hectare.

For legumes, major data like days to 50% flowering, maturity date, green biomass yield, leaf stem ratio, plot cover, stand vigor, plant height, disease incidence, grain yield and land equivalent ratio was collected.

### ***Land equivalent ratio (LER)***

Yield advantages was calculated though land equivalent ratio. Intercropping determined by advantage that indicated the amount of interspecific competition or facilitation in an intercropping system (Fetene 2003)

$$\mathbf{LER} = \frac{y_{is}}{y_{ss}} + \frac{y_{il}}{y_{sl}}$$

Where,  $Y_{is}$  and  $Y_{ss}$  are the yields of intercrop and sole crop of sorghum and  $Y_{il}$  and  $Y_{sl}$  are the yields of intercrop and sole crop of legumes.

A LER more than 1.0 reveals an intercropping has an advantage or a demonstration that interspecific facilitation is higher than interspecific competition so that intercropping results in greater land-use efficiency. LER less than 1.0 reveals mutual antagonism in the intercropping system. As a result, a LER less than 1.0 has no intercropping advantage (Wahla *et al.*, 2009).

### ***Statistical Analysis***

Data were subjected to analysis of variance (ANOVA) using (SAS version 9.1.3). Significance differences between treatments means were separated using Least Significance Difference (LSD) test at 5% probability level.

## **RESULTS AND DISCUSSIONS**

### **Influence of Lablab on Sorghum yield and yield components**

#### ***Days of 50% flowering and Maturity***

The combined analysis of 50% heading and maturity date of sorghum were presented in (Table 3). There were statistical ( $p < 0.05$ ) variation between sorghum in 50% heading date but has not variation in terms of maturity due to intercropping date of lablab. The longest date to 50% heading and maturity was recorded from (86.2 day). Sorghum intercropping with lablab in different sowing date does not influence maturity date of sorghum (Dagim variety).

#### ***Plant height and Stand count at Harvest***

The combined analysis of sorghum plant height and stand count at harvest were presented in (Table 3). There were statistical ( $p < 0.05$ ) variation between sorghum plant height and stand count at harvest due to intercropping date of lablab. The longest plant height of 160.3 cm was recorded from sorghum + lablab simultaneous whereas the shortest plant height 152.3 cm was recorded from sorghum + lablab after four weeks. The longest plant height for sorghum + lablab simultaneous may be due to growth competition of sorghum rather than gave grain yield. (Abebe *et al.* (2020) reported that plant height for dagim variety

118cm which was shorter than the present results and similar results reported by Ishiaku *et al.* (2016) from 162 to 167cm. This variation may be due to intercropping of lablab helps plant height increment.

Plant population count at harvest has direct effect on grain yield and yield components of most crops. The highest plant count at harvest was recorded from sorghum + lablab after four weeks (73.2) followed by sole sorghum (67.5). The list plant population count at harvest was recorded from sorghum + lablab simultaneous (41.5). Intercropping sorghum-lablab simultaneous causes reduction of plant population that leads grain and grain yield related reduction.

### ***Sorghum grain yield and Yield components***

The combined result of grain yield of sorghum intercropped with lablab in different sowing date for two years were presented (table 3). Sorghum yield was showed significance ( $p < 0.05$ ) variation among the treatments. The highest mean grain yield of sorghum was recorded from sole sorghum (20.61 qt/ha) followed by sorghum + lablab after four weeks (20.32 qt/ha) that indicated the higher yield under sole cropped condition is directly related with lack of competition for nutrient and moisture (Gebremichael *et al.* (2019) whereas the lowest grain yield (10.82 qt/ha) was recorded from Sorghum + lablab simultaneous. The mean of sorghum grain yield variation was due to main effect of planting date that similar result reported by Habtamu and Tadele (2021) there was a significantly difference on grain yield of sorghum due to intercropping legumes in different planting pattern. Shehu *et al.* (1999) also reported that inter-cropping of lablab with sorghum caused a marked reduction in yields of sorghum stem, leaves and grain, while Isaacs *et al.* (2016) found a reduction in height of maize following inter-cropping with lablab. The present findings for sorghum grain yield (Dagim variety) is higher than the findings of Abebe *et al.* (2020) that 13qt/ha who reported that without intercropping. In the current study, the grain yield difference might be due to legumes can fix nitrogen that similar result reported by Karuma *et al.* (2011) Dolichos can fix about 20 kg N/ha under drought conditions. Different scholars reported different sorghum varieties intercropping with lablab on grain yield of sorghum that Rahel *et al.* (2021) 52.5 to 59.5 qt/ha), Habtamu and Tadele (2021) from 31.19 to 36.99) qt/ha.

### ***Plot Cover and Disease Incidence***

Significance ( $p < 0.05$ ) variation was recorded for plot cover (table 3). The highest plot cover (81.5%) was recorded from sole sorghum followed by sorghum + lablab intercropped after four weeks (80.8%) while the lowest plot cover (69.2%) was from Sorghum + lablab simultaneous intercropped. Intercropping lablab with sorghum can affect plot cover of sorghum when it intercropped inappropriate date of sowing.

The diseases severity was the most important criteria during the data collection. Sole sorghum (1.17) was relatively affected by diseases than the other treatments whereas the lowest (0.92) disease incidence was recorded from sorghum + lablab after six weeks. Disease is the most factor reduce grain yield Sanchez-Martin *et al.* (2014) reported that the most affected by diseases, the lowest in yield but intercropping favors the growth and yield of the crops reducing weed distribution, paste and disease Gebremichael *et al.* (2019) also reported similar findings

Table 1: Mean phenology, growth, yield and yield related characters of sorghum as affected by sowing date grown in sole and intercropped with lablab in 2019/20

Treatments	50F	MD	PH	DI	SCH	PC	SV	GY(qt/ha)
S+Lb6w	87.7	138.5	176.3 <sup>c</sup>	0.34 <sup>c</sup>	84.4 <sup>a</sup>	88.3 <sup>a</sup>	1.17 <sup>c</sup>	21.08 <sup>b</sup>
S+Lb4w	87.7	138.5	179.1 <sup>bc</sup>	0.67 <sup>ab</sup>	82.8 <sup>a</sup>	89.2 <sup>a</sup>	1.17 <sup>c</sup>	<b>26.71<sup>a</sup></b>
S+LbSi	87.7	138.5	181.9 <sup>ab</sup>	0.67 <sup>ab</sup>	50.8 <sup>b</sup>	57.5 <sup>b</sup>	3.5 <sup>a</sup>	15.58 <sup>c</sup>
S+Lb2w	87.7	138.5	182.8 <sup>a</sup>	0.84 <sup>a</sup>	58.5 <sup>b</sup>	66.7 <sup>b</sup>	2.5 <sup>b</sup>	20.34 <sup>b</sup>
Sole sorghum	87.7	138.5	178.8 <sup>bc</sup>	0.5 <sup>bc</sup>	83.0 <sup>a</sup>	91.1 <sup>a</sup>	1.34 <sup>c</sup>	25.71 <sup>a</sup>
Mean	87.7	138.5	179.8	0.6	71.9	78.6	1.93	21.88
CV(%)	0	0	1.57	33.02	19.77	9.78	28.41	11.48
LSD (5%)	0	0	3.39	0.24	17.02	9.21	0.66	3
p-value	NS	NS	***	***	***	**	**	***

S+Lb6w= Sorghum + lablab after six weeks, S+Lb4w= Sorghum + lablab after four weeks, S+Lbsi= Sorghum + lablab simultaneous, S+Lb2w= Sorghum + lablab after two weeks, PH =Plant height, MD = maturity date, DI = diseases incidence, PC = plot cover, SV =Stand vigor, SY(qt/ha) = Seed yield quintal per hectare, DMY = dry matter yield, GBth = green biomass yield

Table 2: Mean phenology, growth, yield and yield related characters of sorghum as affected by sowing date grown in sole and intercropped with lablab in 2020/21

Treatments	50F	MD	PH	DI	SCH	PC	SV	GY(qt/ha)
S+Lb6w	87 <sup>a</sup>	139	136 <sup>a</sup>	1	62 <sup>a</sup>	72 <sup>a</sup>	1.7	16.4 <sup>a</sup>
S+Lb4w	86 <sup>ab</sup>	139	126 <sup>b</sup>	1	51.1 <sup>a</sup>	73 <sup>a</sup>	1.8	13.9 <sup>a</sup>
S+LbSi	86 <sup>a</sup>	139	137 <sup>a</sup>	0.7	32.2 <sup>b</sup>	51 <sup>b</sup>	2.3	6.1 <sup>b</sup>
S+Lb2w	85 <sup>b</sup>	139	138 <sup>a</sup>	1	53.8 <sup>a</sup>	72 <sup>a</sup>	1.7	12.9 <sup>ab</sup>
Sole Sorghum	85 <sup>b</sup>	139	135 <sup>a</sup>	1.3	52 <sup>a</sup>	72 <sup>a</sup>	2	15.5 <sup>a</sup>
Mean	86	139	134	1	50.3	68	1.9	13
CV (%)	1.5	0	4.1	57	20.6	20.3	48.	47.5
LSD (5%)	1.57	0	6.6	0.7	12.6	16.6	1.1	7.5
p-value	**	NS	**	**	***	*	NS	**

S+Lb6w= Sorghum + lablab after six weeks, S+Lb4w= Sorghum + lablab after four weeks, S+Lbsi= **Sorghum** + lablab simultaneous, S+Lb2w= Sorghum + lablab after two weeks, PH =Plant height, MD = maturity date, DI = diseases incidence, PC = plot cover, SV =Stand vigor, SY(qt/ha) = Seed yield quintal per hectare, DMY = dry matter yield, GBth = green biomass yield



Table: 3. Combined mean of phenology, growth and yield and yield related characters of sorghum as affected by sowing date grown in sole and intercropped with lablab of two years (2019/20 - 2020/21)

Treatment	50%F	MD	ph	DI	SCH	PC	SV	GY(qt/ha)
S+Lb6w	85.3 <sup>ab</sup>	138.5	156 <sup>b</sup>	0.92	73.2 <sup>a</sup>	80.0 <sup>a</sup>	1.4 <sup>c</sup>	18.74 <sup>a</sup>
S+Lb4w	84 <sup>b</sup>	138.5	152.3 <sup>c</sup>	1.08	67 <sup>a</sup>	80.8 <sup>a</sup>	1.5 <sup>c</sup>	20.32 <sup>a</sup>
S+LbSi	86.2 <sup>a</sup>	138.5	159.8 <sup>a</sup>	1.08	41.5 <sup>c</sup>	54.2 <sup>c</sup>	2.09 <sup>a</sup>	10.82 <sup>b</sup>
S+Lb2w	81.5 <sup>c</sup>	138.5	160.3 <sup>a</sup>	1.0	56.2 <sup>b</sup>	69.2 <sup>b</sup>	2.92 <sup>a</sup>	16.60 <sup>ab</sup>
Sole Sorghum	82.2 <sup>c</sup>	138.5	156.9 <sup>ab</sup>	1.17	67.5 <sup>a</sup>	81.5 <sup>a</sup>	1.67 <sup>bc</sup>	20.61 <sup>a</sup>
Mean	83.8	138.5	157	1.05	61.1	73.1	1.92	17.42
CV (%)	2.61	0	2.83	22.16	18.58	9.7	29.4	40.57
LSD (5%)	1.8	0	3.64	0.91	9.30	5.82	0.46	5.79
p-value	***	NS	***	NS	**	**	**	***

S+Lb6w= Sorghum + lablab after six weeks, S+Lb4w= Sorghum + lablab after four weeks, S+Lbsi= Sorghum + lablab simultaneous, S+Lb2w= Sorghum + lablab after two weeks, PH =Plant height, MD = maturity date, DI = diseases incidence, PC = plot cover, SV =Stand vigor, SY(qt/ha) = Seed yield quintal per hectare, DMY = dry matter yield, GBth = green biomass yield

### Land Equivalent Ratio

Land equivalent ratio was used to evaluate the intercrop efficiency in yield relative to the monocropped condition. The total land equivalent ratios (TLER) were obtained by summing up of the partial land equivalent ratio of sorghum and legume crops (Lablab). The combined result of sorghum intercropping lablab total land equivalent ratio were significantly ( $P < 0.05$ ) difference due to the main effect of lablab intercropping date. The higher total land equivalent ratio (1.89) was obtained from sorghum + lablab simultaneous followed by sorghum + lablab intercropped after four weeks (1.88), indicating that yield advantage over sole crops (table 4). Even though sorghum + lablab simultaneously intercropped produce the highest total LER, the individual LER of sorghum significantly affected by lablab intercropped. LER for sorghum + lablab intercropped after four weeks was produced the ideal acceptable LER for sorghum. The combatable LER value obtained with sorghum + lablab intercropping after four weeks is further evidence of the benefits of planting sorghum and lablab intercropping system in this environment. The same land equivalent ratio of 1.54 to 1.87 was reported by Rahel *et al.* (2021) and 1.88 to 1.98 was reported by Ishiaku *et al.* (2016) for sorghum intercropped with lablab

Table 4: Land equivalent ratio (LER) for biomass yield of intercropping sorghum variety with lablab

Treatments	SPLER	LPLER	TLER
S+Lb6w	0.96 <sup>a</sup>	0.29 <sup>c</sup>	1.25 <sup>b</sup>
S+Lb4w	1.01 <sup>a</sup>	0.87 <sup>c</sup>	1.88 <sup>a</sup>
S+LbSi	0.48 <sup>c</sup>	1.39 <sup>a</sup>	1.87 <sup>a</sup>
S+Lb2w	0.78 <sup>b</sup>	0.67 <sup>d</sup>	1.45 <sup>b</sup>
So Lablab	1.0 <sup>a</sup>	1 <sup>b</sup>	1.00 <sup>c</sup>
Mean	0.84	0.84	1.41
CV (%)	21.65	13.49	11.68
LSD (5%)	0.15	0.09	0.13
p-value	***	*	***

S+Lb6w= Sorghum + lablab after six weeks, S+Lb4w= Sorghum + lablab after four weeks, S+Lbsi= Sorghum + lablab simultaneous, S+Lb2w= Sorghum + lablab after two weeks, SPLER= Sorghum Partial land equivalent ratio, LPLER= Lablab partial land equivalent ratio, TLER = Total land equivalent ratio

### Influences of maize on forage legumes

#### *Lablab yield and yield components*

Analysis of variance showed that, yield of lablab intercropped with sorghum had highly significant ( $p < 0.05$ ) difference due to the effect of date of planting. The highest mean value of lablab yield (19.9 qt/ha) were recorded from sorghum + lablab simultaneously intercropping followed by sole lablab ((14.6 qt/ha) whereas the lowest lablab mean grain yield (4.2 qt/ha) was recorded from sorghum + lablab after six weeks intercropped. The grain yield obtained from intercropped lablab with sorghum in different sowing date was the same with potential productivity of lablab. Similar findings reported by Bagegnehu *et al.* (2021) reported lower (7.25 qt ha<sup>-1</sup>) lablab grain yield when lablab was intercropped with maize as compared with the values obtained in this study.

#### *Fresh biomass and dry matter yield*

The combined result of fresh biomass yield and dry matter yield of lablab intercropped with sorghum were presented in table 7. The combined fresh biomass yield where significantly ( $p < 0.05$ ) difference due to lablab intercropped with sorghum. The highest mean fresh biomass was recorded from sorghum + lablab simultaneous intercropping (58.9 t/ha) followed by sole lablab (52.4 t/ha) whereas the lowest (20.7 t/ha) was recorded from sorghum + lablab after six weeks intercropped (table 7). Fresh biomass yield of lablab was decreasing as sowing date increasing that was due to suppressing and high sun light competition of sorghum over lablab.

There was no statistical ( $p > 0.05$ ) variation observed among the treatments on dry matter yield of lablab due to intercropped with sorghum. Numerically the highest dry matter yield (2.31 t/ha) was recorded from sorghum + lablab after four weeks intercropped whereas the lowest dry matter yield (2.15 t/ha) was recorded from sorghum + lablab after six weeks intercropped (table 5). This result is higher than the

findings of Daniel *et al.* (2020) 1.05 t/ha intercropped with maize whereas Rahel *et al.* (2021) reported higher results from 3.47 to 5.65 t/ha intercropped with sorghum

### Leaf to stem ratio and Plot Cover

The combined mean of leaf to stem ratio and plot cover were presented in table 7. Statistical ( $p < 0.05$ ) variation observed for both leaf to stem ratio and plot cover. Higher leaf to stem ratio was recorded from sorghum + lablab after two weeks (0.68) followed by sorghum + lablab after six weeks intercropped (0.67), while Sorghum + lablab after four weeks, Sorghum + lablab simultaneous and sole lablab was produce the lowest. The highest plot cover of lablab was recorded from sole sorghum (95%) followed by sorghum + lablab simultaneous (92.2%) whereas the lowest was from sorghum + lablab after four weeks (67.5%) and sorghum + lablab after six weeks (68%) intercropped.

Table 5: The phenology, growth, yield and yield related characters of Lablab as affected by sowing date grown in sole and intercropped with sorghum in 2019/20

Treatments	MD	GBth	DMYtha	LSR	SV	PH(cm)	DI	PC(%)	SY(qt/ha)
S+Lb6w	174.3 <sup>ab</sup>	17.1 <sup>c</sup>	2.08 <sup>c</sup>	0.75 <sup>a</sup>	3 <sup>a</sup>	120.5 <sup>c</sup>	1 <sup>b</sup>	68 <sup>c</sup>	3.07 <sup>e</sup>
S+Lb4w	170.9 <sup>b</sup>	25.2 <sup>c</sup>	2.31 <sup>ab</sup>	0.52 <sup>b</sup>	2.5 <sup>b</sup>	132.9 <sup>c</sup>	1.2 <sup>b</sup>	67.5 <sup>c</sup>	11.1 <sup>c</sup>
S+LbSi	170.3 <sup>ab</sup>	71.6 <sup>a</sup>	2.2 <sup>abc</sup>	0.71 <sup>a</sup>	1.3 <sup>c</sup>	176.9 <sup>a</sup>	1.8 <sup>a</sup>	92.2 <sup>ab</sup>	20.1 <sup>a</sup>
S+Lb2w	180.8 <sup>b</sup>	42.1 <sup>b</sup>	2.38 <sup>a</sup>	0.56 <sup>b</sup>	1.3 <sup>c</sup>	161.2 <sup>b</sup>	2 <sup>a</sup>	88.3 <sup>b</sup>	5.6 <sup>d</sup>
Sole Lablab	176.8 <sup>a</sup>	61.7 <sup>a</sup>	2.18 <sup>bc</sup>	0.52 <sup>b</sup>	0.7 <sup>d</sup>	135.5 <sup>c</sup>	1.7 <sup>a</sup>	95 <sup>a</sup>	14.6 <sup>b</sup>
<b>Mean</b>	173.2	43.5	2.23	0.62	1.8	145.4	1.5	82.2	10.9
<b>CV%</b>	1.96	27.64	7.14	13.2	4.3	6.78	22	6.13	20.9
<b>LSD 5%</b>	4.06	6.04	0.91	0.09	14	11.8	0.4	0.09	3.2
<b>p-value</b>	***	***	*	***	***	**	**	***	***

S+Lb6w= Sorghum + lablab after six weeks, S+Lb4w= Sorghum + lablab after four weeks, S+LbSi= Sorghum + lablab simultaneous, S+Lb2w= Sorghum + lablab after two weeks, PH =Plant height, MD = maturity date, DI = diseases incidence, PC = plot cover, SV =Stand vigor, SY(qt/ha) = Seed yield quintal per hectare, DMY = dry matter yield, GBth = green biomass yield

Table 6: The phenology, growth, yield and yield related characters of Lablab as affected by sowing date grown in sole and intercropped with sorghum in 2020/21.

Treatments	MD	GBth	DMYtha	LSR	SV	PH(cm)	DI	PC(%)	SY(qt/ha)
<b>S+Lb6w</b>	195 <sup>a</sup>	24.3 <sup>b</sup>	2.12	37 <sup>ab</sup>	1.5 <sup>a</sup>	81 <sup>c</sup>	4.1 <sup>ab</sup>	84 <sup>bc</sup>	5.4 <sup>c</sup>
<b>S+Lb4w</b>	181 <sup>b</sup>	30 <sup>b</sup>	2.32	35 <sup>ab</sup>	1.5 <sup>a</sup>	83 <sup>bc</sup>	3.3 <sup>b</sup>	80 <sup>c</sup>	14.3 <sup>b</sup>
<b>S+LbSi</b>	192 <sup>a</sup>	46.4 <sup>a</sup>	2.13	39 <sup>a</sup>	1 <sup>b</sup>	111 <sup>a</sup>	4.43 <sup>a</sup>	89 <sup>ab</sup>	19.7 <sup>a</sup>
<b>S+Lb2w</b>	190 <sup>a</sup>	24.6 <sup>b</sup>	2.21	33 <sup>b</sup>	1.5 <sup>a</sup>	97 <sup>ab</sup>	3.67 <sup>ab</sup>	83 <sup>c</sup>	13.3 <sup>b</sup>
<b>Sole lablab</b>	182 <sup>b</sup>	43 <sup>ab</sup>	2.51	35 <sup>ab</sup>	1.2 <sup>ab</sup>	97 <sup>ab</sup>	4.3 <sup>ab</sup>	91 <sup>a</sup>	14.7 <sup>b</sup>
<b>Mean</b>	188	33.67	2.25	35.5	1.33	94	3.96	86	11.6
<b>CV%</b>	3	43.13	14.4	15.2	29.93	14.1	22.11	5.5	24.7
<b>LSD 5%</b>	6.9	15.63	0.4	6.5	0.48	16.1	1.06	5.71	3.5
<b>p-value</b>	**	*	NS	**	***	***	***	***	**

S+Lb6w= Sorghum + lablab after six weeks, S +Lb4w= Sorghum + lablab after four weeks, S+Lbsi= Sorghum + lablab simultaneous, S+Lb2w= Sorghum + lablab after two weeks, PH =Plant height, MD = maturity date, DI = diseases incidence, PC = plot cover, SV =Stand vigor, SY(qt/ha) = Seed yield quintal per hectare, DMY = dry matter yield, GBth = green biomass yield

Table 7. Combined mean of phenology, growth, yield and yield related characters of Lablab as affected by sowing date grown in sole and intercropped with sorghum of two years (2019/20 - 2020/21)

Treatments	MD	GBth	DMYt/ha	LSR	SV	PH(cm)	PC	SY(qt/ha)
S+Lb6w	184.7 <sup>a</sup>	20.7 <sup>c</sup>	2.15	0.67 <sup>a</sup>	2.25 <sup>a</sup>	100.8 <sup>d</sup>	76.0 <sup>c</sup>	4.2 <sup>e</sup>
S+Lb4w	175.9 <sup>c</sup>	27.6 <sup>cb</sup>	2.31	0.53 <sup>b</sup>	2 <sup>a</sup>	17.9 <sup>d</sup>	73.8 <sup>c</sup>	12.7 <sup>c</sup>
S+LbSi	180.4 <sup>b</sup>	58.9 <sup>a</sup>	2.16	0.52 <sup>b</sup>	1.42 <sup>b</sup>	129.1 <sup>ab</sup>	90.6 <sup>ab</sup>	19.9 <sup>a</sup>
S+Lb2w	182.7 <sup>ab</sup>	33.4 <sup>b</sup>	2.29	0.68 <sup>a</sup>	1.17 <sup>bc</sup>	143.9 <sup>a</sup>	85.7 <sup>b</sup>	9.43 <sup>d</sup>
Sole Lablab	179.4 <sup>b</sup>	52.4 <sup>a</sup>	2.29	0.54 <sup>b</sup>	0.93 <sup>c</sup>	116.3 <sup>c</sup>	93.0 <sup>a</sup>	14.6 <sup>b</sup>
<b>Mean</b>	180.6	38.6	2.24	0.59	1.55	119.61	83.8	12.9
<b>CV%</b>	2.2	27.96	10.5	10.86	26.42	7.66	7.17	13.17
<b>LSD 5%</b>	3.29	8.85	0.19	0.05	0.34	7.51	4.93	1.31
<b>p-value</b>	***	***	NS	***	***	**	**	**

S+Lb6w= Sorghum + lablab after six weeks, S +Lb4w= Sorghum + lablab after four weeks, S+Lbsi= Sorghum + lablab simultaneous, S+Lb2w= Sorghum + lablab after two weeks, PH =Plant height, MD = maturity date, DI = diseases incidence, PC = plot cover, SV =Stand vigor, SY(qt/ha) = Seed yield quintal per hectare, DMY = dry matter yield, GBth = green biomass yield

## CONCLUSIONS AND RECCOMENTATIONS

Intercropping of lablab with sorghum has influenced on yield and yield components of sorghum. So, it is important to develop optimum sowing date of sorghum-lablab intercropping. The highest yield of sorghum grain yield was recorded from sole sorghum followed by sorghum + lablab intercropping after four weeks whereas the lowest was from Sorghum + lablab simultaneous intercropped. The highest grain

yield of lablab recorded from Sorghum + lablab simultaneous intercropped whereas the lowest yield was recorded from sorghum + lablab after six weeks intercropped. The grain yield of intercropped lablab with sorghum in different sowing date was the same with potential productivity of lablab that intercropping lablab with sorghum has no influence on grain yield of lablab. In conclusion, this study has revealed that, lablab sown after four weeks later than sorghum is the appropriate sowing date in sorghum-lablab intercropping system. Therefore, it is recommended as the most compatible association to improve yields and yield related traits in the study area and other areas with similar agro-ecologies. Further studies should be conducted to test fodder nutritional quality and changes in soil macro and micro minerals.

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# Evaluation of Bracharia Grass for Forage Yield and Nutritive Quality at Adami Tulu, Oromia, Ethiopia

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## Abstract

*The study was conducted at Adami Tulu Agricultural Research Center for two consecutive years during 2019 and 2020 rainy season to identify Bracharia cultivars for herbage dry matter (DM) yield and nutritive quality. The tested cultivars were ILRI 11553, Bracharia hybrid-ICARDA, ILRI 12974, Bracharia Mulato, ILRI 13165, Bracharia decumbens and ILRI 687. The cultivars were planted in randomized complete block design with three replications. All recommended agronomic managements were applied uniformly. A plot size of 1.5 m × 2 m (3 m<sup>2</sup>) with spacing between rows, plots and blocks were 50 cm, 1.5 m and 1.5 m, respectively, and used to establish the experiment. Forage dry matter yield was determined by harvesting the two middle rows for each plot and representative sample collected from each treatment and sub-sampled for DM and nutritional parameter analysis. The collected data were subjected to SAS software version 9.1. Significant effect was observed among the Bracharia cultivars on plant height ( $p < 0.001$ ), tiller number ( $p < 0.002$ ), plot cover ( $p < 0.015$ ) and vigority ( $p < 0.004$ ). Similarly, forage dry matter yield (DM) showed significant ( $p < 0.004$ ) differences among the cultivars. Higher herbage dry matter yield was recorded from ILRI Acc. 12974 (9.32 t/ha) followed by Bracharia decumbens (9.27 t/ha), but both are statistically the same, while the lower DM yield (5.49 t/ha) was received from ILRI Acc. 687. The highest leaf to stem ratio (LSR) (10.78) was recorded for ILRI Acc. 12974, while the lower LSR (3.59) was recorded for ILRI Acc. 13165. Concentration of CP varied significantly ( $p < 0.042$ ) among the treatments ranging from 6.2% for Bracharia hybrid-ICARDA to 7.73% for ILRI Acc. 687. Ash content was significantly ( $p < 0.002$ ) higher for Bracharia hybrid-ICARDA (15.45%), while lower value was recorded for ILRI Acc. 11553 (10.95%). Therefore, Bracharia ILRI Acc. 12974 and Bracharia decumbens produced optimum forage biomass and nutrient quality and hence recommended for promising forage biomass production at the study areas and other areas with similar agro ecologies.*

**Keywords:** Biomass, Nutritive value, crude protein, dry matter and yield

## Introduction

Ethiopia has enormous livestock resources and diverse agro-ecologies suitable for different kinds of livestock production. Livestock resources have significant economic and social importance at household level and makes significant contributions to the national economy and foreign currency earnings of the country through export of live animals, meat as well as hides, skins and leather products (Adunga *et al.*, 2012). However, its production has been mainly constrained due to different factors. Inadequate supply and poor qualities of available feed resources (Gelayenew, 2016; EIAR, 2017 and Wubetie *et al.*, 2018) have been identified to be one of the major factor limiting the production and productivity of livestock. In the rift valley area, livestock feed is based on natural pastures, fallow land, stubble grazing and crop residues which are poor in quantity and quality (Abebe *et al.*, 2015). Thus, the existing feed resources do

not meet the nutrient requirements for growth and reproductions of animals. Therefore, One approach for alleviating and tackling feed shortage problem is identification and development of adaptable improved forages which have high herbage yields and suitable for the existing climatic condition. However, the productivity of the different grass species could be distinctly different and is also influenced by area of origin, including temperature, light intensity, total rain fall, soil type, fertilization level, and by stage of maturity (Huhtanen *et al.*, 2006; Jančík *et al.*, 2009).

Bracharia is one of the improved forages, a tropical and warm season (Ashebir *et al.*, 2019), perennial forage grasses that have high forage yield potential and adapted to different agro-ecologies. It is native to Africa (Duncan *et al.*, 2020, Labarta *et al.*, 2017) and produces more dry matter than most tropical grasses during the dry season and has been widely implemented as quality pastures for animal production as cultivated forage grasses and grazing land improvement. The grasses is very important, because of its high productivity under intensive use, its tolerance of low fertility, relative freedom from pests and disease (Helder *et al.*, 2011) and remain green long into the dry season. It is also a valuable grass for erosion control as it covers the ground well, it withstands heavy grazing and establishes on poor and rocky soils. It also shows excellent response to fertilizer, improves soil quality and is persistent (Labarta *et al.*, 2017). The animals that consume the bracharia grass produce higher yields of milk and their manure emits smaller amounts of nitrous oxide (Asheber *et al.*, 2019). Data on nutritive value indicate that, forage from bracharia is highly palatable, leading to high intake, whether fed fresh or grazed (Ndikumana and Leeuw de, 1996). Yields range between 5 and 36 tDMha<sup>-1</sup> year depending on soil fertility, moisture and fertilizer application (Bogdan, 1977). The deep-rooted, productive bracharia grasses can capture atmospheric carbon on a scale similar to that of tropical forests, and could be a further plus for climate change mitigation (Labarta *et al.*, 2017). Regardless of the importance, it is widely adaptable and high yielding.

However, the forage yield, nutritive value and adaption potential of bracharia cultivar have not been identified in the study areas. Therefore, this study was designed with the objective to identify the adaptable and high yielding bracharia cultivars for forage production in the study area.

## **Materials and Methods**

### ***Description of the study area***

Adami Tulu Agricultural Research Center is found in Adami Tulu Jido Kombolcha district that located in the mid-rift valley of Ethiopia, at 167 kilometers from the capital city of the country, in south eastern part of Oromia between 38°20' and 38.5°5' E and 7°35' and 8°05' N. It lies at altitudinal range from 1500 to 2000 masl (Assefa *et al.*, 2013). The agro-ecology of ATJK district is semi-arid and sub-humid in which 90% of the area is lowland while the remaining 10% is intermediate. The minimum and maximum temperatures are 22 to 28°C, respectively. It has an average annual rainfall of 760 mm. It has a bimodal rainfall from March to April (short rain season) and July to September (long rain season) with a dry period in May to June, which separates short rains from long rain (Teshome *et al.*, 2012).



### ***Experimental Design and Treatments***

The experiment was conducted for two consecutive years during 2019 and 2020 rainy season. Both Bracharia varieties and accessions were used to conduct the experiment due to shortage of Bracharia cultivars for comparisons. The experimental design was RCBD and comprised seven (7) treatments with three replications. The experimental land was well ploughed and prepared for establishment of the bracharia grasses. Plot size was (3m<sup>2</sup>) 1.5m\*2m and spacing of 50cm, 1.5m and 1.5m between rows, plots and block were used respectively. The plant was established by splitting of bracharia cultivars. Fertilizer and other agronomic management practice were applied as per recommendation.

### ***Sampling procedures***

Plant attributes (Plant height, Leaf length and tiller numbers) recorded randomly and plot cover and vigourity were estimated visually. Forage yield evaluation took place by harvesting of the two rows of each plot. The fresh biomass was weighted and sub-sample also taken for further nutritional analysis. After collecting samples from the two rows, the whole plot was cut to allow the homogenous regrowth for the next cut. The sub sample was oven dried for 48hr at 60°C to determine the dry mater and sample ground to pass through 1 mm sieve at ATARC animal feed laboratory. In addition, the ash was determined by burning with carbolite oven at 550°C for 3hr. Laboratory chemical analyses for some of the major parameters have been done at ATARC. The feed sample was analyzed for crude protein using Kjeldahl method.

### ***Statistical Analysis***

The collected data on agronomic parameters and dry matter yields of forage samples were organized and interred into Microsoft excel for statistical analysis. The organized data were subjected to ANOVA based on the model designed for a RCBD. Therefore, analysis was undertaken by SAS software version 9.1, with One-way analysis of variance (ANOVA) procedures and Tukey's Studentized Range (HSD) Test used to separate the mean values for each parameter at P<0.05.

## Results and Discussion

The analysis of variance showed that a significant differences of *Bracharia* cultivars on plant height ( $p<0.001$ ), tiller number ( $p<0.002$ ), plot cover (PC) ( $p<0.015$ ) and vigourity ( $p<0.004$ ) (table 1).

Table 1. Mean of agronomic parameters and leaf to steam ration of bracharia cultivars at Adami Tulu

Treatments	Agronomical parameter of bracharia trial				
	PH	Tiller	Vigor	PC	LSR
ILRI 11553	101.25 <sup>bcd</sup>	94.84 <sup>b</sup>	50.81 <sup>b</sup>	58.75 <sup>b</sup>	5.645 <sup>bc</sup>
Bracharia hybrib-ICARDA	78.25 <sup>d</sup>	252.91 <sup>a</sup>	53.75 <sup>b</sup>	69.00 <sup>ab</sup>	8.202 <sup>ab</sup>
ILRI 12974	105.39 <sup>abc</sup>	259.45 <sup>a</sup>	80.67 <sup>ab</sup>	77.17 <sup>ab</sup>	10.783 <sup>a</sup>
Mulato	88.72 <sup>cd</sup>	207.00 <sup>ab</sup>	80.50 <sup>ab</sup>	76.17 <sup>ab</sup>	7.354 <sup>abc</sup>
ILRI 13165	104.72 <sup>abc</sup>	240.75 <sup>a</sup>	94.50 <sup>a</sup>	90.67 <sup>a</sup>	3.593 <sup>c</sup>
<i>Bracharia decumpens</i>	126.55 <sup>a</sup>	197.65 <sup>ab</sup>	86.83 <sup>a</sup>	85.00 <sup>ab</sup>	6.960 <sup>abc</sup>
ILRI 687	116.92 <sup>ab</sup>	237.17 <sup>a</sup>	67.50 <sup>ab</sup>	67.92 <sup>ab</sup>	6.871 <sup>abc</sup>
Overall mean	103.11	212.82	73.51	74.95	7.06
<b>LSD</b>	23.83	19.81	31.17	27.10	16.12
CV	12.80	21.19	23.50	20.11	21.68
P-value	<.0001	0.0021	0.0004	0.0156	0.0023

Means with the same letter across the column are not significantly different.

PH=plant height, CV=Coefficient variation, PH=plant height, PC=plot cover

*Bracharia decumpens* had higher plant height (126.55 cm) followed by ILRI 687 (116.92 cm). In addition, ILRI 13165 had higher plot cover (90.67%) and vigourity (94.5%) followed by *Bracharia decumpens* (86.83%) and (85%), respectively. In the study, Brachiaria species performed better in plant spread, plant cover and tiller recruitment. The tillering and spreading ability of bracharia is an added advantage for soil cover and protection as well as being an animal feed (Nguku, *et al.*, 2016).

Table 2. Nutritional parameters of bracharia cultivars at ATARC.

Treatments	Nutritional parameters					
	DM t/ha	Ash	OM	CP	NDF	ADF
ILRI 11553	7.79 <sup>ab</sup>	10.95 <sup>b</sup>	79.35	7.43 <sup>ab</sup>	21.76	36.84
Bracharia hybrib-ICARDA	5.72 <sup>b</sup>	15.45 <sup>a</sup>	76.05	6.2 <sup>b</sup>	21.65	39.22
ILRI 12974	9.32 <sup>a</sup>	14.60 <sup>a</sup>	76.73	7.16 <sup>ab</sup>	23.45	38.40
Mulato	6.58 <sup>ab</sup>	16.15 <sup>a</sup>	75.28	6.97 <sup>ab</sup>	23.84	33.33
ILRI 13165	6.97 <sup>ab</sup>	14.12 <sup>ab</sup>	76.95	6.97 <sup>ab</sup>	23.51	37.25
<i>Bracharia decumpens</i>	9.27 <sup>a</sup>	13.98 <sup>ab</sup>	76.77	6.81 <sup>ab</sup>	22.19	39.00
ILRI 687	5.49 <sup>b</sup>	15.13 <sup>a</sup>	76.25	7.73 <sup>a</sup>	25.17	40.64
Overall mean	7.31	14.34	76.77	7.04	23.08	37.81
LSD	3.47	3.60	4.67	1.33	7.98	11.49
CV	26.30	13.90	3.37	10.45	19.15	16.84
P-value	0.004	0.0026	0.2258	0.0424	0.8016	0.5650

Means with the same letter across the column are not significantly different. CV=Coefficient variations, LSD=least significance differences, LSR=leaf steam ratio, OM=Organic matter, CP=Crude protein, NDF=Neutral detergent fiber, ADF=Acid detergent fiber

The dry matter yield of *Brachiaria* cultivars was significantly different ( $P < 0.05$ ) as indicated in the above Table 2. The ILRI 12974 and *Brachiaria decumpens* had higher DM yield than the other cultivars. These results were in line with the (Helder *et al*, 2011) and (Mutimura and Everson, 2012). *Brachiaria decumpens* is adapted to infertile soil and withstands heavy grazing and trampling. The grass can tolerate shading and is suitable for soil erosion control (Kenaff, nd). In addition, concentration of CP varied significantly ( $p < 0.042$ ) among the treatments ranging from 6.2% for *Brachiaria* hybrid-ICARDA to 7.73% for ILRI Acc.687. However, the highest CP percentage was observed with ILRI 687 cultivar. The, organic matter (OM), Neutral detergent fiber (NDF) and acid detergent lignin (ADL) concentrations did not differ ( $p > 0.05$ ) among *brachiaria* cultivars. Ash content was significantly ( $p < 0.002$ ) higher for *Brachiaria* hybrid-ICARDA (15.45 %), while lower value was recorded for ILRI Acc. 11553 (10.95%).

### Conclusions and Recommendation

Based on the results, it can be concluded that, the forage agronomical parameters, yield and nutritive quality of *brachiaria* Cultivars showed significance differences. The *brachiaria decumpens* and ILRI 12974 have higher dry matter yield  $\text{tonha}^{-1}$  while 687 has lower yield. The leaf to stem ratio also showed the significance differences between the treatments of *brachiaria* cultivars. *Brachiaria decumpens* and ILRI 12974 showed the highest DM and other yields in the study area. Therefore, based on the dry matter and agronomical parameter, ILRI 12974 and *brachiaria decumpens* were recommended based on high biomass and DM yields. Thus, *brachiaria decumpens* and ILRI 12974 cultivars, that have the highest DM yield, should be used for further demonstration and scaling up among farmers. In addition, due to long drought season, perenniality and vegetativeness, using irrigation is very important for sustainable production. Determining cutting frequency, cutting interval for estimation the potentials of *brachiaria* cultivar in detail is needed as it can be harvested two times only with rain fall.

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# Evaluation of Desho Grass (*Pennisetum pedicellatum*) for Agronomic Performances, Biomass Yield and Nutritive Content at Bore and Adola districts, Guji Zone of Oromia

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## Abstract

The study was conducted at research sites of Bore and Adola districts representing highland and midland agro-ecologies respectively. The objective of the experiment was to identify adaptable and high biomass yield of Desho grass varieties. Accordingly, four Desho grass varieties; Kindo Kosha DZF No# 591, Areka DZF No# 590, Kulumsa DZF No # 592 and Kindo Kosha DZF No# 589 were evaluated in randomized complete block design (RCBD) with three replications. The result of highland site revealed that plant height was significantly ( $P < 0.05$ ) differ among the treatments. The highest value of plant height was measured from Kindo Kosha DZF No# 589 (102.6 cm) followed by Areka DZF No# 590, Kulumsa DZF No # 592 (97.7 cm) varieties, while the shortest plant height was recorded from Kindo Kosha DZF No# 591 (91.2 cm) variety. On the other hands, leaf to stem ratio, vigor, number of tiller per plant, number of leaf per plant, survive rate and dry matter yield (DM) were not shown significant ( $P > 0.05$ ) differences between treatments. Numerically the maximum DM yield (30.53t/ha) was obtained from Areka DZF No# 590 followed by Kindo Kosha DZF No# 589 (28.7t/ha). At Midland, number of tiller per plant, number of leaf per plant and plant height were significantly ( $P < 0.05$ ) differ among the treatments. The highest plant height was measured from Areka DZF No# 590 (108.6 cm) followed by Kindo Kosha DZF No# 589 (107 cm). On the other hands the result indicated that plot cover, vigor, survival rate, leaf to stem ratio and dry matter yield were not significant differ ( $P > 0.05$ ) among the treatments. Even if the dry matter yield was not statistically differ among the tested varieties, the maximum DM yield was measured from Areka DZF No# 590 (26.13t/ha) variety followed by Kindo Kosha DZF No# 591 (26.01 t/ha). Generally the result showed that Kindo Kosha DZF No# 589 and Areka DZF No# 590 were performed best for Bore (highland) site while Kindo Kosha DZF No# 591 variety was well performed at Adola (midland) site in agronomic, yield and quality parameters. Hence, these varieties should be promoted for the end users as an options of feed resources in the study areas and similar agro ecologies.

**Keywords;** Biomass yield, Desho grass, Forage varieties, *Pennisetum pedicellatum*,

## Introduction

Desho grass (*Pennisetum pedicellatum*) is native to tropical countries including Ethiopia (Ecocrop, 2010; Leta *et al.*, 2013; EPPO, 2014). In Ethiopia Desho grass is known as a perennial plant originated in Southern Nations (Welle *et al.*, 2006). Desho grass is a many-branched leafy grass growing up to 1 m high or more (FAO, 2010; Leta *et al.*, 2013). The culms are erect and branching, and the leaves are 15-25 cm long and 4-10 mm wide, flat and glabrous. The spikelets are 4 mm long, usually solitary (Ecocrop, 2010; FAO, 2010). Desho grass is mainly found on disturbed land, road edges and on recent fallow lands, where annual rainfall range between 600 mm and 1500 mm with a rainy season of 4-6 months and

average daily - temperatures of about 30- 35°C. Desho grass thrives on a wide range of soils (including degraded sandy or ferruginous soils) provided they are well drained. However, the grass is susceptible to water logging and frost but has some drought tolerance (Ecocrop, 2010; FAO, 2010). Currently it is utilized as a means of soil conservation practices and animal feed in the highlands of Ethiopia (Yakob *et al.*, 2015). The grass is popular, drought resistant plant, used as feed for ruminants (FAO, 2010; EPPO, 2014). It has the potential of meeting the challenges of feed scarcity since it provides more forage per unit area and ensures regular forage supply due to its multi-cut nature (Ecocrop, 2010). Desho grass is suitable for intensive management and performs well at an altitude ranging from 1500 to 2800 m.a.s.l (Leta *et al.*, 2013). Desho grass performs best at an altitude greater than 1700 m above sea level (Welle *et al.*, 2006).

Desho grass has a crude protein content of 9.6% on DM basis at early stage and 1.6% at straw stage, respectively. The digestibility and voluntary intake decrease with increase in stage of maturity which indicates that the grass should be fed at early stage of maturity. Mature Desho grass must be well supplemented with protein sources in order to sustain growth and/or milk production. Urea treatment may be a valuable option to improve its nutritive value (nitrogen content and digestibility) with the addition of adequate energy supplementation. From animal feed resource point of view, Desho grass is used in temporary pastures or in cut-and carry systems since it provides ample quantities of good quality green forage and stands several cuts a year. The grass is also useful for hay and silage preparation (Ecocrop, 2010). To improve availability of livestock feed in terms of quantity and quality, it is better to cultivate Desho grass forage that have better biomass yield and nutritional quality. Therefore, the objective of the study was to evaluate Desho grass varieties for its agronomic performances, biomass yield and nutritional content at the study area.

## **Materials and Methods**

### ***Description of the Study Area***

The experiment was carried out at Bore and Adola districts respectively representing highland and midland agro-ecologies of Guji Zone of Oromia. Bore district is located at 385 km to the south Oromia from Finfinne and 220 km from the Guji Zone capital city (Negele) with geographical location of 55°7'23" to 62°6'52" N latitudes and 38°25'51" to 38°56'21" E longitudes, South-eastern Oromia. The annual rain fall is about 1400-1800mm and the annual temperatures of the district ranged from 10.1 to 20°C. The major soil type of the site is black soil. Bore Agricultural Research station is located at 7 km from Bore town which is geographically located at 62°4'37" N latitude and 38°34'76" E longitudes. The research site was at an altitude of 2736 m.a.s.l. receiving high rainfall characterized by bimodal distribution. The first rainy season extends from April to October and the second season starts late November and ends at the beginning of March. Adola District is located at distance of 470 km from Addis Ababa and 120 km from the Zonal capital city, Negele Borena. It is an area where a mixed farming and semi-nomadic economic activity takes place, which is the major livelihood of the local people. The total area of District is 1254.56 km<sup>2</sup>. The district is situated at 5°44'10" - 6°12'38" N Latitudes and 38°45'10" - 39°12'37" E Longitudes. The District is characterized by three agro-climatic, namely highland (11%), midland (29%) and lowland (60%). The major soil types of the district are nitosols (red basaltic soils) and orthic acrosols (Yazachew E. and Kasahun D.2011).

### ***Experimental Treatments and Design***

The experiment was conducted using four varieties of Desho grasses; Kindo Kosha DZF No# 591, Areka DZF No# 590, Kulumsa DZF No # 592 and Kindo Kosha DZF No# 589. RCBD with three replications was applied. Splits of plant tiller were planting in rows spaced 50cm, 10cm between plants and 1m, 1.5m between plot and block respectively on plot size of 4m x 3m (12 m<sup>2</sup>). Other agronomic management practices were applied uniformly as per recommendation.

### ***Data Collection***

All relevant data like vigor, plot cover, number of tiller per plants, number of leaf per plants, number of survive rates, plant heights, leaf to stem ratio, biomass yield and nutritive value were recorded.

### ***Chemical Analysis***

Samples of feeds were dried in an oven for 72 hr at 65<sup>0</sup>C to determine the DM, Ash and N contents were determined according to the standard procedure of AOAC (1990). The ash content was determined by burning/igniting feed samples in a muffle furnace at 550<sup>0</sup>C and N content of feeds was determined according to Kejlhdhal procedure and the crude protein (CP) was calculated as N\*6.25. Neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) were analyzed according to the procedure of Van Soest and Robertson (1985).

### ***Statistical Analysis***

All collected data were analyzed using the general linear model procedure of SAS version 9.1. The statistical model for the analysis data was:

$$Y_{ijk} = \mu + A_j + B_i + e_{ijk}$$

Where;  $Y_{ijk}$ = response of variable under examination,  $\mu$  = overall mean,  $A_j$  =the  $j$ th factor effect of treatment/ cultivar,  $B_i$  =the  $i$ th factor effect of block/ replication,  $e_{ijk}$  = the random error.

## **Results and Discussion**

### ***Performances of Desho Grass Varieties at Bore (Highland) site***

Mean value of agronomic parameters and biomass yield of Desho grass are indicated in table 1. The analyzed result shows that there was a significantly ( $P < 0.05$ ) differences in plot cover and plant height among the tested treatments. The highest plot cover was produced from Kindo Kosha DZF No# 589(78.4%) while the lowest value (66.67%) was obtained from Kulumsa DZF No # 592 variety. This result is lower than result reported by Tekalegn *et al.* (2017). The highest plant height was measured from Kindo Kosha DZF No# 589 (102.6.6 cm) while the shortest plant height was recorded from Kindo Kosha DZF No# 591.

On the other hands, plant vigor, number of tiller per plants, number of leaf per plant, leaf to stem ratio and dry matter yield were shown non-significant ( $P > 0.05$ ) difference among the treatments. The highest

survive rate was measured from Areka DZF No# 590 (63.3 %) followed by Kindo Kosha DZF No# 589 (61.3 %) varieties. Numerically the highest plant vigor was recorded from Areka DZF No# 590 (78.4%). This finding lower than the value (96.3 %) reported by Tekalegn *et al.* (2017) for Areka-DZF # 590 Desho grass lines in Southern, Ethiopia. The result also indicated that numerically the highest value (30.5 t/ha) of dry matter yield were measured from Areka DZF No# 590 followed by Kindo Kosha DZF No# 589 (28.73 t/ha).

Table 1. Mean value of agronomic and yield component of Desho grass varieties at Bore site

Varieties	PC (%)	Vg (%)	SR (%)	NTPP	NLPP	PH (cm)	LSR	DMY (t/ha)
Kindo Kosha DZF No# 591	74 <sup>ab</sup>	74.8	58.6	137.9	11.14	91.2 <sup>b</sup>	0.76	24.8
Areka DZF No# 590	74 <sup>ab</sup>	78.4	63.3	145.7	13.77	97.7 <sup>ab</sup>	0.77	30.5
Kulumsa DZF No # 592	66.67 <sup>b</sup>	74.9	46.67	145.3	12.34	97.7 <sup>ab</sup>	0.66	27.2
Kindo Kosha DZF No# 589	78.4 <sup>a</sup>	77.7	61.33	135.3	14.17	102.6 <sup>a</sup>	0.78	28.7
<b>Mean</b>	<b>73.3</b>	<b>76.5</b>	<b>57.5</b>	<b>141.1</b>	<b>12.85</b>	<b>97.2</b>	<b>0.73</b>	<b>27.8</b>
<b>CV</b>	<b>6.2</b>	<b>10.3</b>	<b>25.7</b>	<b>5.6</b>	<b>14.4</b>	<b>4.4</b>	<b>17.5</b>	<b>11.4</b>
<b>Sig.level</b>	<b>*</b>	<b>ns</b>	<b>ns</b>	<b>ns</b>	<b>ns</b>	<b>*</b>	<b>ns</b>	<b>ns</b>

<sup>a,b,c</sup> Mean in a column within the same category having different superscripts differ significantly (p<0.05) PH =plant height PC=Plot cover, LSR=leaf to steam ratio, Vg=vigor, SR=survive rate, NTPP=number of tiller per plants, NLPP= number of leaf per plants, DMY =Dry matter yield, CV=Coefficient of variation, \*\*= highly significant, \*=significant and ns= None significant different.

The chemical compositions values of evaluated varieties are presented in Table 2. The laboratory result revealed that Kindo Kisha-DZF#589 variety had higher ash content (14.7%) than others. However, the Areka-DZF#590 variety had higher DM and CP content followed by Kindo Kosha DZF No# 591. The result obtained from this study on CP values for all tested varieties were higher than previously reported values by Denbela *et al.*, (2020) which ranges from 6.93-9.38% under different agro-ecologies. On the other hands, the value obtained from this study for NDF was lower than previously reported values which ranged 72.78-77.68% by Bimrew *et al.*, (2017).

Table 2: Mean chemical composition (DM %) of Desho grass varieties evaluated at Bore site

Varieties	DM	Ash	CP	NDF	ADF	ADL
Kindo Kosha DZF No# 591	94.2	12.5	17.07	72.6	42.2	12.03
Areka DZF No# 590	95.5	13.2	17.2	71.6	47.7	6.98
Kulumsa DZF No # 592	95.4	12.4	10.8	65.9	48.13	6.47
Kindo Kosha DZF No# 589	94.5	14.7	10.21	70.3	46.5	9.5

DM=dry matter, CP= crude protein, NDF=neutral detergent fiber, ADF=acid detergent fiber, ADL=acid detergent lignin

### **Performances of Desho Grass Varieties at Adola (Midland) Site**

Agronomic and biomass yield parameters of the tested Desho grass varieties evaluated at midland site are presented in table 3. Plant height, number of tiller per plant and number of leaf per plant were shown a significant (P<0.05) differences among the treatments. The highest mean value of plant height was measured from Areka DZF No# 590 (108.6 cm), followed by Kindo Kosha DZF No# 589 (107 cm) varieties while the shortest plant height (95.3 cm) was recorded from Kindo Kosha DZF No# 591. This



difference might be due to soil types and harvesting at different dates. On the other hands, the highest number of tiller per plant was recorded from Kulumsa DZF No # 592 (168.2) followed by Kindo Kosha DZF No# 589 (156.8) variety. The least value of number of tiller per plant was obtained from Areka DZF No# 590 (134.2). This result is higher than the finding reported by Bimrew *et al.* (2017) (39.4 cm) under irrigation at Northern Ethiopia. Numerically the highest (95.7%) mean value of survive rate was measured from Areka DZF No# 590 followed by Kindo Kosha DZF No# 591 (87.7%) varieties.

The analysis result of plot cover, vigor, leaf to stem ratio and dry matter yield indicated that there was non-significant differences ( $P>0.05$ ) observed among the treatments. This is might be due to shuttering of leaf at with fluctuation of temperature. The values of leaf to stem ratio recorded were ranged from 0.38-0.46. Numerically the he highest mean value of herbage dry matter yield (26.13 t/ha) was measured from Kindo Kosha DZF No# 589 followed Kindo Kosha DZF No# 591 (26.01t/ha). The lowest dry matter yield was measured from Areka DZF No# 590 and Kulumsa DZF No # 592 (24.57t/ha). This is higher than the result reported by Denbela *et al.*, (2020).

Table 3. Mean value of agronomic and yield component of Desho grass at Adola site

Varieties	PC (%)	Vg (%)	SR (%)	NTPP	NLPP	PH (cm)	LSR	DMY(t/ha)
Kindo Kosha DZF No# 591	72.2	81.4	87.7	151.5 <sup>ab</sup>	10.3 <sup>b</sup>	95.3 <sup>b</sup>	0.46	26.01
Areka DZF No# 590	71.0	79.2	95.7	134.2 <sup>b</sup>	11.4 <sup>b</sup>	108.6 <sup>a</sup>	0.38	24.57
Kulumsa DZF No # 592	63.6	77.4	76.6	168.2 <sup>a</sup>	10.5 <sup>b</sup>	101.6 <sup>ab</sup>	0.39	24.57
Kindo Kosha DZF No# 589	71.1	79.5	92.4	156.8 <sup>ab</sup>	12.6 <sup>a</sup>	107 <sup>a</sup>	0.45	26.13
<b>Mean</b>	<b>69.5</b>	<b>79.4</b>	<b>88.2</b>	<b>152.7</b>	<b>11.16</b>	<b>103.1</b>	<b>0.41</b>	<b>25.96</b>
<b>CV</b>	<b>20.4</b>	<b>4.8</b>	<b>15</b>	<b>7.4</b>	<b>6.8</b>	<b>4.3</b>	<b>18.1</b>	<b>7.7</b>
<b>Sig.level</b>	<b>ns</b>	<b>ns</b>	<b>ns</b>	<b>*</b>	<b>*</b>	<b>*</b>	<b>ns</b>	<b>ns</b>

<sup>a,b,c</sup> Mean in a column within the same category having different superscripts differ significantly ( $p<0.05$ )

PH =plant height PC=Plot cover, LSR=leaf to steam ratio, Vg=vigor, SR=survive rate, NTPP=number of tiller per plants, NLPP= number of leaf per plants, DMY =Dry matter yield, CV=Coefficient of variation, \*\*= highly significant, \*=significant and ns= None significant different.

The nutritional compositions of Desho grass varieties evaluated at Adola site are presented in Table 4. The result indicated that Areka DZF No# 590 contain the highest DM% (95.02%). Kindo Kisha-DZF#589 variety had higher ash content (19.7%) followed by Kindo Kosha DZF No# 591(19.5%). However, the higher (10.6%) CP content was obtained from Kulumsa DZF No # 592 followed by Kindo Kosha DZF No# 591 (9.8%). Kindo Kosha DZF No# 591 variety contains the highest (70.3%) NDF followed by Kindo Kosha DZF No# 589 (69.4%). On the other hands, the maximum value of ADF (51.3%) and ADL (26.7%) were recoded from Kulumsa DZF No # 592 Desho gass variety. The value obtained in this study for NDF was lower than previously reported values which ranged 72.78-77.68% by Bimrew *et al.* (2017).

Table 4: Mean chemical composition (DM %) of Desho grass varieties evaluated at Adola site

Varieties	DM	Ash	CP	NDF	ADF	ADL
Kindo Kosha DZF No# 591	94.7	19.5	9.8	70.3	46.8	11.4
Areka DZF No# 590	95.02	17.6	8.8	66	43.8	10.09
Kulumsa DZF No # 592	92.5	15.3	10.6	66.9	51.3	26.7
Kindo Kosha DZF No# 589	94.02	19.7	8.84	69.4	49.08	8.8

DM=dry matter, CP= crude protein, NDF=neutral detergent fiber, ADF=acid detergent fiber, ADL=acid detergent lignin

## Conclusion and Recommendations

Different varieties of Desho grass were evaluated for their agronomic parameters, biomass yield and nutritional composition. From the evaluated varieties, Kindo Kosha DZF No# 589 and Areka DZF No# 590 performed best at Bore site (highland) while Kindo Kosha DZF No# 591 variety have shown good potential at Adola (midland) site in agronomic, yield and quality parameters. Thus, it could be possible to conclude that these Desho grass varieties were performed well at the study site. Hence, these varieties should be promoted for the end users as an option of feed resources in the study areas and similar agro ecologies.

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# Adaptation Trial of Desho Grass (*Pennisetumpedicellatum*) at Midland and Highland Agro Ecology of East Hararghe Zone, Oromia, Ethiopia

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## Abstract

*The experiment was undertaken to identify the adaptable and high yielding Dasho grass line under two agro ecologies (midland and highland) of east Haraghe zone of Oromia region during 2018-2020 main cropping seasons. Four Dasho grass (*Pennisetum pedicellatum*) lines (Kindo kosha-DZF-591, Araka-DZF-590, Kulumsa, Kindo kosha-DZF#589) were evaluated in randomized complete block design with three replications. The general linear model procedures of SAS and least significance difference for data analysis and mean separation were employed respectively. The combined analysis revealed that Desho grass lines had significant effect on tiller number per plant ( $p < 0.05$ ) at both agro ecologies. The dry matter (DM), Ash, crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) contents were shown significant ( $p < 0.001$ ) difference among the Desho grass lines considered in the trial. The lines had no significant ( $p > 0.05$ ) effect on herbage dry matter, ground cover, plant height and leaf to stem ratio at both agro ecologies. Even though herbage dry matter yield was showed statistically non-significant results among the Desho grass lines, some lines were slightly dry matter yielder than other lines at their respective agro ecologies. Accordingly, Kindo kosha-DZF#589 (28.72 t/ha) and Kulumsa (28.51 t/ha) for midland and Kindo kosha-DZF-591 (25.06 t/ha) and Araka-DZF-590 (24.93 t/ha) for highland agro ecologies were recommended and further demonstration and scaling up/out should be implemented at the study area and similar agro-ecologies.*

**Key words:** Crude protein, Desho grass, Dry matter yield, Leaf to stem ratio, Plant height, Tiller number

## Introduction

Despite Ethiopia has large livestock population (CSA, 2016), the productivity of livestock is low. Feed shortage in terms of quantity and quality is the major constant for livestock production in Ethiopia (ILRI, 2009; Demeke *et al.*, 2017). The current report of CSA (2015) revealed that 56, 30 and 1.2% of the total livestock feed supply of the country is derived from grazing on natural pasture, crop residues and agro industrial byproducts, respectively. Since, natural pasture and crop residues are source of low quality feed it might be not met even the maintenance requirement for animal. To combat these nutritional constraints, use of improved forage species which are adaptable to wide agro-ecological conditions and grown with low inputs is important (Anele *et al.*, 2008). According to Lukuyu *et al.* (2011), to evaluate any feed and inclusion into livestock feeding program it is very important to have chemical composition and utilization information of locally available feed resources.

Among locally available potential feed resource in e Ethiopia, *Desho* grass (*Pennisetum pedicellatum*) is the most appropriate one (EPPO, 2014; Leta *et al.*, 2014). *Desho* grass is native to tropical countries including Ethiopia (Ecocrop, 2010; Leta *et al.*, 2013; EPPO, 2014). *Desho* grass is known as a perennial plant originated in Southern region of Ethiopia. It is palatable to cattle, sheep and other herbivores (FAO,

2010) and suitable for intensive management and performs well at an altitude ranging from 1500 to 2800 m.a.s.l. (Leta *et al.*, 2013). It has the potential of meeting the challenges of feed scarcity since it provides more forage per unit area and ensures regular forage supply due to its multi-cut nature suggests that it is a potential feed source in the dry season when feed availability in the tropics is critical (Cocrop, 2010). The grass is drought resistant plant, used as feed for ruminants (FAO, 2010; EPPO, 2014).

Desho grass serves as a business opportunity for farmers in Ethiopia (Shiferaw *et al.*, 2011; Tilahun *et al.*, 2017). Currently the grass is utilized as a means of soil conservation practices and animal feed in the highlands of Ethiopia (Welle *et al.*, 2006; Ecocrop, 2010). To obtain the highest yield of *Desho* grass should be cut within 4 months after sowing at 8 cm from ground level (Leta *et al.*, 2013). The yield and nutritional qualities of forage are influenced by numerous factors such as seasonal variations, stage of maturity, ecological conditions and management practices (Giovanni *et al.*, 2011). However, there is no adequate information on the agronomic characteristics, productivity and chemical composition of *Desho* grass in east Hararghe zone of Oromia, Ethiopia. Therefore, the current study was conducted with the objective to identify the adaptable and high yielding *Dasho* grass line under mid and high altitudes of East Hararghe. Zone of Oromia, Ethiopia.

## **Materials and Methods**

### ***Description of the study area***

The experiment was conducted under rain-fed conditions from 2018-2020 growing season in midland and highland of East Hararghe zone. Three districts namely Meta, Kersa and Kombolcha were selected for the study. The midland experimental study was conducted at the three districts, while the highland experimental study was conducted only at Meta district. For each agro ecology six locations were used.

Meta District is located at 445 km from the capital Addis Ababa and 80 km West of Harar town. It is located between 9°0'09" to 9°0'31" N latitude and 41°0'29" to 41°0'44" E longitude. Altitude of Meta District is 2830m a.s.l. The annual rainfall amount ranges from 600-900 mm and the temperature ranges between 15 °C-37 °C. Kersa district is bordering Haromaya district in the East, Kurfa Calle district in the South, Dire Dawa City administration in the North and Meta District in the West. The capital city of the district is located at 478km South of Addis Ababa and 42 km to the West of Harar town. The district contains 35 rural Peasant associations (PA) and the altitude ranges from 1,550 to 2,800 m a.s.l. Kombolcha district is one of the eighteen districts of East Hararghe Zone of Oromia Regional State. It is located at about 17 km north of Harar town and 542 km East of Addis Ababa, the nation's capital city. The altitude of the district ranges from 1200-2460 m a.s.l. Agro climatically, the district ranges from *Woina-dega* (*mid-altitude*) to Kola (low lands). The annual rainfall ranges from 600 mm to 900 mm with a bimodal and erratic pattern. The mean annual temperature of the area ranges between 16-25°C.

### ***Experimental treatments and design***

The experimental materials were obtained from Wando Genet Research Center. The experiment was conducted as a Randomized Complete Block Design (RCBD) with four replications. The experimental fields were ploughed and harrowed to a fine seedbed. Plot size was 4 x 3 m (12 m<sup>2</sup>). The root splits were planted with the intra and inter row spacing of 0.25 m and 0.5 m respectively. Land preparation, planting,

weeding and harvesting was made according to the recommendations (Leta *et al.*, 2013). NPS and urea were applied at the rate of 100 and 50 kg per ha.

### **Data collection**

Harvesting was done by hand using a sickle, leaving a stubble height of 8 cm above the ground (Leta *et al.*, 2013). The morphological parameters such as plant height were measured with measuring tape. The number of tillers was computed as mean of counts taken from ten plants that was randomly selected from the middle rows of each plot at 120 days after planting (Leta *et al.*, 2013). The leaf to stem ratio was determined by measuring 2 kg of fresh weight from the selected two middle rows (1 m<sup>2</sup>), separating in to leaves and stems, drying and weighing each component separately. Fresh herbage yield of the grass was measured immediately after each harvest and weighed on the field soon after mowing using a field balance. Sub-samples were taken from each plot at each site to determine dry matter yield. Finally the sub-samples were dried in oven dry at 65°C for 72 hours and stored in airtight bags to be used for chemical analysis.

### **Statistical analysis**

The values on agronomic parameters and dry matter yields were statistically evaluated by analysis of variance (ANOVA) using general linear model (GLM) procedure of Statistical Analysis Software to perform ANOVA (SAS 9.1). Mean were separated using least significant difference (LSD) at 5% significant level.

### **Results and Discussion**

#### ***Agronomic performances of Desho grass lines at midland agro ecology***

Combined analysis of variance for measured agronomic traits and herbage yields of Desho grass lines tested over environments is presented in Table 1. Since the interaction among the treatment and locations is non-significant, combined analysis was used. The analysis of variance shown that the lines had no significant ( $p>0.05$ ) effect on ground cover and plant height. However, numerically different mean value of ground cover and plant height were observed among the tested Desho grass lines. Thus, higher ground cover (97.75%) was obtained from Kindo kosha-DZF#589. The mean ground cover (96.35%) of the lines is within the range of mean ground cover reported (95.8-99.2%) by Tekalegn *et al.* (2017) for Dasho grass lines at Wondogenet Agricultural Research Center, Southern Ethiopia. The similarities of the findings were indicated that the ability of Desho grass adapted to different environments and soil types. This might be due to indigenous ecotype of Desho grass for Ethiopia (Smith, 2010). Similarly, Desho grass lines had no significant ( $p>0.05$ ) effect on plant height. The mean plant height was ranged from 125.6-151 cm. This result is in agreement with Shiferaw *et al.* (2011) who reported that Desho grass grows upright with the potential of reaching 120 cm based on soil fertility. However, the result of the current study is higher than the figure reported by Bimrew *et al.* (2017) (94 cm) for Desho grass at mid land altitude of Northern, Ethiopia.

The result of this study revealed that lines significantly ( $p < 0.001$ ) affect number of tillers per plant at midland agro ecology. The mean tiller per plant yield of the lines was ranged 115.9 -138.2. This result is higher than the report of Bimrew *et al.* (2016) (47.66) for Desho grass at mid land altitude of Northern, Ethiopia. This suggests a selection for increased tiller number per plant as an effective method for increasing biomass production and greater yields which is supported by Das *et al.* (2004), who found that tiller density was correlated with biomass production in grasses.

#### ***Dry matter yield and leaf to stem ratio***

The result of combined analysis showed that herbage dry matter yield did not significantly ( $p > 0.05$ ) affected by lines. Even though the dry matter yield in ton per hectare was not differ significantly ( $p > 0.05$ ) between Desho grass lines, optimum dry matter yield ranging from 24.67- 28.72 t/ha was produced. The mean herbage dry matter yield (24.67-28.72 t/ha) of the lines is within the range of mean herbage dry matter yield reported by Tekalegn *et al.* (2017) (28.43-30.9 t/ha) for similar Desho grass lines at Wondogenet Agricultural Research Center, Southern, Ethiopia.

The mean leaf to stem ratio of the Desho lines was 1.08, but no significant difference ( $p > 0.05$ ) was observed among the Desho lines in dry matter percentage. This is higher than the finding of Tekalegn *et al.* (2017) who reported that 0.72 for Desho grass lines, but it is lower than the finding of Bimrew (2016) who reported that leaf to stem ratio 1.18 harvested at 120 days at mid land altitude.

Table 1. The combined mean of agronomic parameters and biomass yield of Desho grass lines at midland agro ecology

<b>Treatments</b>	<b>GC%</b>	<b>PH(cm)</b>	<b>TN</b>	<b>DMY(t/ha)</b>	<b>LSR</b>
Kindo kosha-DZF-591	97.15	131	125.6 <sup>b</sup>	25.43	1.16
Araka-DZF-590	96.5	126.2	115.9 <sup>c</sup>	24.67	1.05
Kulumsa	94	133.2	131.8 <sup>ab</sup>	28.51	1.02
Kindo kosha-DZF#589	97.75	151	138.2 <sup>a</sup>	28.72	1.09
<b>CV (%)</b>	3.9	10.1	3.2	8.8	9.8
<b>LSD</b>	5.39	21.9	6.56	3.80	0.17
<b>p-value</b>	NS	NS	***	NS	NS

GC=Ground cover, PH=plant height, TN=tiller number, DMY=Total dry matter yield, LSR= Leaf to stem ratio, CV= coefficient of variation

#### ***Agronomic Performance of Desho Grass lines at Highland Agro ecology***

The combined analysis result for ground cover, plant height and tiller number of four Desho lines are indicated in table 2. Since the interaction among the treatment and location is non-significant combined analysis was used. The results showed that significant ( $p < 0.05$ ) effect was observed on ground cover and tiller number per plant among the Desho grass lines evaluated in the trial. The line Kindo kosha-DZF-591 was recorded the highest tiller number per plant (107) followed by Kindo kosha-DZF#589 (95.75) and Kulumsa-DZF-592 (92.5). This is higher than the value (49.47) obtained by Bimrew *et al.* (2017) for Desho grass at high land of Northern, Ethiopia. The least mean square was not indicated significant difference ( $p > 0.05$ ) on plant height among the tested Desho grass lines. However, numerically the

maximum plant height (166.65 cm) was recorded from Kindo kosha-DZF-591 followed by Kindo kosha-DZF#589 (107.75 cm). This result is in agreement with that of Shiferaw *et al.* (2011) who reported that Desho grass grows upright with the potential of reaching 90–120 cm based on soil fertility, but it is higher than the report of Bimrew *et al.* (2017) (87 cm) for Desho grass lines at high land areas of Northern, Ethiopia.

#### ***Dry matter yield and leaf to stem ratio***

Combined analysis result for herbage dry matter yields and leaf to stem ratio of Desho grass lines are presented in Table 2. Desho lines had no significant ( $p>0.05$ ) effect on both herbage dry matter yield and leaf to stem ration. The least square mean result on total dry matter yield and leaf to stem ratio were not revealed significant difference ( $p>0.05$ ) among Desho grass lines examined. Although the mean least square was not shown significant difference on dry matter yield, the treatment mean value is ranges between 24.51-25.06 t/ha. Numerically higher herbage dry matter yield (25.06/ha) was recorded from Kindo kosha-DZF-591 than the other tested lines. The results in the current study is greater than the reports of Bimrew *et al.* (2016) (19.9 t/ha) and Tilahun *et al.* (2017) (15.7 t/ha), respectively, for Desho grass lines at high land agro ecologies. However, it is lower than the result obtained (28.43-30.9 t/ha) by Tekalegn *et al.* (2017) at Wondogenet Agricultural Research Center, Southern, Ethiopia.

The mean leaf to stem ratio of the Desho lines was 1.11. This result is higher than the finding reported by Tekalegn *et al.* (2017) (0.44-0.55), but it is comparable with the finding of Bimrew (2016) who reported 1.15 that leaf to stem ratio for Desho grass lines at highland areas.

Table 2. The combined mean of agronomic parameters and biomass yield of Desho grass at highland agro ecology

<b>Lines</b>	<b>GC%</b>	<b>PH (cm)</b>	<b>TN</b>	<b>DMY (t/ha)</b>	<b>LSR</b>
<b>Kindo kosha-DZF-591</b>	90.50 <sup>a</sup>	116.25	107.00 <sup>a</sup>	25.06	1.11
<b>Araka-DZF-590</b>	87.50 <sup>ab</sup>	110.50	91.00 <sup>c</sup>	24.93	1.12
<b>Kulumsa</b>	85.50 <sup>b</sup>	103.25	92.50 <sup>bc</sup>	24.51	1.11
<b>Kindo kosha-DZF#589</b>	89.00 <sup>ab</sup>	107.75	95.75 <sup>b</sup>	24.62	1.10
<b>CV (%)</b>	2.1	6.5	2.1	1.5	3.2
<b>LSD (0.05)</b>	2.86	8.93	3.23	0.57	0.06
<b>P-value</b>	*	NS	**	NS	NS

GC=Ground cover, PH=plant height, TN=tiller number, DMY=Total dry matter yield, LSR= Leaf to stem ratio, CV= coefficient of variation

#### ***Nutritional quality of Desho grass lines***

The nutritional contents of the Desho grass lines are presented in Table 3. The result from dry matter yield (DM), Ash, crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) contents were shown significantly ( $p<0.001$ ) different among the Desho grass lines. The highest (91.75%) DM% concentration was obtained from Kindulosha-DZF-591 Desho grass lines followed by Araka-DZF-590(91.03%), while Kulumsa-DZF-592 was produced lower DM value (89.56%) as compared with other tested lines. The highest Ash content was produced from Kindo kosha-



DZF-591 (14.37%), while the lowest ash value (13.66%) was produced by Kindo kosha-DZF#589. The highest mean crude protein value was recorded from Kulumsa-DZF-592 (13.51), but Kindo kosha-DZF-591 line was produced the lowest (11.16%) CP value as compared to the other tested Desho grass lines. The highest NDF concentration was obtained from Kulumsa-DZF-592 line (72.42%), while the lowest value was recorded from Kindo kosha-DZF#589 (63.68%). The highest (34.33%) ADF content was recorded by Kindo kosha-DZF-591 whereas the lowest value (30.82%) was produced by Kindo kosha-DZF#589. Likewise, the highest (9.28%) ADL concentration was recorded by Kindo kosha-DZF-591 line, while the lowest value (4.77%) was noted by Kulumsa-DZF-592. The highest ADF concentration in the grasses shows less digestible than the other lines with lower ADF value. This is consistent with work of Albayrak *et al.* (2011) who reported that as the ADF increases the digestibility of the forage usually decrease causing consumption of the forage by animal to decrease.

Table 3. The combined mean value of chemical composition of the Desho grass lines at both agro ecology

Treatments	DM%	Ash%	CP%	NDF%	ADF%	ADL%
<b>Kindo kosha-DZF-591</b>	91.75 <sup>a</sup>	14.27 <sup>a</sup>	11.16 <sup>d</sup>	71.08 <sup>b</sup>	34.33 <sup>a</sup>	9.28 <sup>a</sup>
<b>Araka-DZF-590</b>	91.03 <sup>b</sup>	13.76 <sup>b</sup>	12.8 <sup>b</sup>	60.53 <sup>d</sup>	32.18 <sup>b</sup>	9.08 <sup>a</sup>
<b>Kulumsa-DZF-592</b>	89.56 <sup>d</sup>	14.37 <sup>a</sup>	13.51 <sup>a</sup>	72.42 <sup>a</sup>	31.08 <sup>c</sup>	4.77 <sup>c</sup>
<b>Kindo kosha-DZF#589</b>	90.67 <sup>c</sup>	13.66 <sup>b</sup>	11.96 <sup>c</sup>	63.68 <sup>c</sup>	30.82 <sup>c</sup>	6.49 <sup>b</sup>
<b>CV (%)</b>	1.1	1.5	1.7	1.9	1.5	7.8
<b>LSD</b>	0.07	0.08	0.26	0.76	0.58	0.70
<b>P-value</b>	**	**	**	**	**	**

DM: Dry matter, CP: Crude protein, NDF: Neutral detergent fiber, ADF: Acid detergent fiber, ADL: Acid detergent lignin, LSD: least significant difference, CV: coefficient of variation

### Conclusions and Recommendation

The results revealed non-significant differences in total dry matter yield ton per hectare and leaf to stem ratio among the four Desho grass lines. Although herbage dry matter yield was showed statistically non-significant results among the evaluated Desho grass lines, optimum herbage dry matter yield was produced at their respective agro ecologies. Accordingly, Kindo kosha-DZF#589 (28.72 t/ha) and Kulumsa (28.51 t/ha) were well performed at midland and Kindo kosha-DZF-591 (25.06 t/ha) and Araka-DZF-590 (24.93 t/ha) were well perfumed at highland agro ecologies. Additionally, the lines were produced reasonable CP concentration which ranges from 11.16-13.51% for Kindo kosha-DZF-591 to Kulumsa line, respectively. Generally, all Desho grass lines were produced optimum herbage dry matter yield and nutrient quality and should be recommended for promising forage biomass production at the study areas and other areas with similar agro ecologies. Therefore, Kindo kosha-DZF#589 and Kulumsa lines for midland and Kindo kosha-DZF-591 and Araka-DZF-590 for highland agro ecology were recommended and further demonstration and scaling-up/out should be implemented at the study area and similar agro-ecologies.

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# Adaptation trial of Brachiaria Grass Accessions for Herbage Yield and Nutritive Quality at Lowland and Midland Agro ecology of East Hararghe Zone, Oromia, Ethiopia

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## Abstract

*Livestock production in the Eastern Oromia of Ethiopia depends mainly on natural pastures and crop residues which are poor in quality and quantity particularly during dry season. Therefore, it need introduction of alternative improved forages of high quality which are adapted to the areas. This activity was conducted at four locations in which one site (Boko) was from lowland and three sites (Meta, Kersa and Kombolcha) were from midland of East Hararghe of Oromia, Ethiopia during 2018, 2019 and 2020 cropping season with the objective to identify and select the best Brachiaria grass accession/s for dry matter (DM) yield and nutritive quality in the study areas. Four brachiaria Urochloa decumbens accessions (ILRI-10871, ILRI-13205, ILRI-14721 and ILRI-14720) and five brachiaria Urochloa ruziziensis accessions (ILRI-13332, ILRI-14743, ILRI-10871, ILRI-14774, LRI-14813) and one local check were evaluated. The accessions were laid out in a randomized complete block design with three replications. Tiller numbers, plant height, forage dry matter yield and plot covers were recorded at their respective recommended stages. Accessions had significant ( $p < 0.05$ ) effect on plot cover, plant height, tiller number, dry matter yield and in nutritional contents. Based on the result, higher herbage DM yield (24.685 t/ha) was recorded from Brachiaria Urochloa ruziziensis ILRI-14813 followed by Urochloa decumbens ILRI-14721 (20.89 t/ha), while lower herbage dry matter yield (5.92 t/ha) was received from Urochloa decumbens ILRI-10871 at lowland. On the other hand, higher herbage dry matter yield (17.83 t/ha) was obtained from U. ruziziensis ILRI-14813, while lower DM yield (7.27 t/ha) was produced from Urochloa decumbens ILRI-10871 at mid land agro ecology. Concentration of CP varied significantly ( $p < 0.001$ ) among the accessions at midland ranging from 8.08%-10.87% and 10.49%-14.45% at lowland and midland, respectively. Thus, Brachiaria ruziziensis U. ruziziensis ILRI-14813 and Brachiaria decumbens Urochloa decumbens ILRI-14721 were produced optimum herbage yield and nutrient quality at lowland and midland areas and should be recommended for promising forage biomass production at the study areas and other areas with similar agro ecologies.*

**Keywords:** Dry matter yield, Plant height, Plot cover, Tiller numbers, Nutrition quality

## Introduction

Forages play an important role in agricultural economy of developing countries by providing the cheapest source of feed for the livestock. In lowlands of East Hararghe zone, one of the most important challenges to livestock production is scarcity of feeds during the dry season. Smallholder farmers face the challenge that grazing land is gradually becoming scarcer, and their current cattle productivity is too low for effective commercialization. Farmers depend on natural pasture and crop residue for livestock and more often give low priority to pasture establishment. These available feed resources in the smallholder mixed farms are inadequate in quantity and low in quality.

Past attempts to improve livestock production in this area focused mainly on crop residues and farmers usually harvest fodder from thinned crop plants, weeds, and defoliated leaves. Despite these efforts, cultivated forages account very low contribution mainly due to lack of suitable grasses adapted to environmental conditions of the area. In addition, land sub-division has also contributed to feed shortage through limited available land for pasture establishment. Planting nutritious forages on small parcels of land and cut-and-carry these to feed their penned cattle can considerably increase animal production and associated income here. Particularly as beef demand of the area is increasing in the country, this presents cattle-keeping smallholders in East Hararghe zone is with an opportunity to enhance their livelihoods. To address the challenges of feed shortage in the study area, there is need to select high quality forages that are adapted to East Hararghe zone.

The genus *Brachiaria* consists of about 100 species distributed across tropical and sub-tropical region (Renvoize *et al.*, 1996). *Brachiaria* have advantage over those in other genera including adaptation to drought and low fertility soils, sequesters carbon through its large roots system, enhance nitrogen use efficiency and subsequently minimize eutrophication and greenhouse gas emissions (Arango *et al.*, 2014; Moreta *et al.*, 2014; Rao *et al.*, 2014). *Brachiaria* plays important roles in soil erosion control and ecological restoration. *Brachiaria* species have been important component of sown pastures in humid low lands and savannas of tropical America with current estimated acreage of 99 million hectare in Brazil alone (Jank *et al.*, 2014). Africa is the center of origin of *Brachiaria* grasses. *Brachiaria* species are native to eastern and central Africa and are extensively grown as livestock forage in South America and East Asia (FAO, 2015). Important species include *B. ruziziensis*, *B. decumbens*, *B. brizantha* and *B. humidicola*.

Recently, the mounting demand for livestock products in Africa has renewed interest of farmers, researchers, and development and government agencies on forages particularly to climate resilient forages like *Brachiaria* grass. Therefore, there have been multiple repatriations of *Brachiaria* grass to Africa as hybrids and improved landraces (Maas *et al.*, 2015; Ghimire *et al.*, 2015). These materials have shown positive performance in terms of improved forage availability and livestock productivity. Despite the immense benefits demonstrated of these grasses in other countries, the potential of improved *Brachiaria* grass in East Oromia to address the challenge of livestock feed scarcity remain unexploited and there is no information on the production and uses of these grasses in the Eastern parts of the region. Therefore, there is a need for identifying *Brachiaria* grass accessions that are more productive and adaptable to the lowlands and midlands production systems of East Hararghe, Oromia, since accessions within a species differ in yield potential and quality of forage produced.

## **Materials and Methods**

### ***Description of the study area***

The study was conducted during 2018, 2019 and 2020 of main cropping season in lowland and midland of East Hararghe zone. Four districts were selected from lowland and midland. The lowland study was conducted at Fedis District. Fedis Agricultural Research Center, in Fedis District on Boko station, which is 550 km to the East of Addis Ababa and 24 km southeast of Harari city used as lowland site. The Fedis district is situated at an altitude of 1200 to 1600 m and 1500 m of Boko station above sea level, (Fuad *et al.*, 2018). The amount of rainfall are varies between 650 and 750 mm, while the average temperature of

the district ranges between 25 and 30 °C (Zenna, 2016). In the vicinity of the site; Vertisols and *Afilsols* soil type are common to the area (FARC, 2013).

The midland study was conducted in Meta, Kersa and Kombolcha districts of the Eastern Hararge Zone, Oromia Regional State. Meta District is located at 445 km from the capital Addis Ababa and 80 km west of Harar town. Meta District is located between 9°0'09" to 9°0'31" N latitude and 41°0'29" to 41°0'44" E longitude (Meta District Livestock Office, 2015). Altitude of Meta District is 2830 meters above sea level. The annual rainfall amount ranges from 600-900 mm and the temperature ranges between 15 °C-37 °C. Gara Muleta Mountain, one of the highest mountains in Oromia Regional State, is found in this District. Kersa district is bordering Haromaya district in the East, Kurfa Calle woreda in the south, Dire Dawa City administration in the north and Meta Woreda in the West. The capital city of the woreda is located at 478 km south of Addis Ababa and 42km to the West of Harar town which is the capital city of East Hararghe zone. The woreda contains 35 rural kebeles and the altitude ranges from 1,550 to 2,800 meters above sea level. Kombolcha district is one of the eighteen districts of East Hararghe Zone of Oromia Regional State. It is located at about 17 km north of Harar town and 542 km east of Addis Ababa, the nation's capital city. The altitude of the district ranges from 1200-2460 meters above sea level. Agro climatically, the district ranges from *Woina-dega (mid-altitude)* to Kola (low lands). The annual rainfall ranges from 600mm to 900mm with a bimodal and erratic pattern. The mean annual temperature of the area ranges between 16-25°C.

### ***Experimental design and treatments***

The experiment was conducted at two locations for three years during 2018 to 2020 cropping season in lowland and midland agro ecology. For each agro ecology six locations were used. The *Brachiaria* grass accessions used for the experiment were four *Brachiaria Urochloa decumbens* accessions (ILRI-10871, ILRI-13205, ILRI-14721 and ILRI-14720) and five *Brachiaria Urochloa ruziziensis* accessions (U. ruziziensis ILRI-13332, U. ruziziensis ILRI-14743, U. decumbens ILRI-10871, U. ruziziensis ILRI-14774, and ruziziensis ILRI-14813) and one local check as control. All the experimental materials were obtained from Holeta Agricultural Research Center except the local check. The accessions were arranged in a randomized complete block design with three replications. The plot sizes were 4 m x 3 m with a 1 m path between plots and 1.5 m between blocks. The grass roots were planted at about 0.5 m and 0.25 m between rows and plants, respectively on a well prepared seed bed.

### ***Data collection***

Plot cover, plant height and tiller number were recorded at 16 weeks after planting in both agro-ecologies. At 16 weeks after planting, the plants were harvested for dry matter (DM) yield determination. Plant height was determined by measuring the primary shoots from the base of the plant to the topmost flag leaf of four tagged plants as described by Rayburn and Lozier (2007). The percentage of plot cover was determined from a 1 m x 1 m quadrat sub-divided into 25 squares as described by Njarui and Wandera (2004). Tillers were counted for tagged plants. During the DM yield determination, the plants were cut to a stubble height of 5 cm in an area of 1 m<sup>2</sup>. Fresh herbage was harvested, weighed and a sub-sample taken, oven dried at of 65 °C for 72 hours to a constant weight.

### ***Chemical Composition***

Partially dried feed samples were ground to pass through a 1 mm sieve screens using Wiley mill and stored in airtight plastic bags for chemical analysis. The DM content was determined by oven drying at 105 °C for 24 hours. The ash component was determined by igniting the dried sample in a muffle furnace at 550 to 600 °C for 4 hours (Sluiter *et al.*, 2008). The nitrogen was determined using the micro-Kjeldahl technique. The CP was calculated as 6.25 x Nitrogen. The method of Van Soest and Robertson (1985) was used to determine neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL). Hemicelluloses were calculated by subtracting the ADF from the NDF content while cellulose was determined by subtracting the ADL from the ADF content. Chemical composition was analyzed at Haramaya University animal nutrition laboratory.

### ***Data Analysis***

The values on agronomic parameters and dry matter yields were statistically evaluated by analysis of variance (ANOVA) using general linear model (GLM) procedure of Statistical Analysis Software to perform ANOVA (SAS 9.1). Means were separated using Tukey's test at  $p < 0.05$  (Gomez and Gomez, 1984).

## **Results and Discussion**

### ***Morphological and yield data of Brachiaria grass accessions at lowland***

Combined analysis of the results for growth parameters (plant height, plot cover, tiller number) and dry matter yield during the experimental period (2018-2020) are presented in Table 1. Since the interaction among the treatment and location is non-significant combined analysis was used. There were significant differences ( $p < 0.01$ ) on groundcover, plant height and dry matter yield and tiller number ( $p < 0.05$ ) among the Brachiaria grass accessions considered in the trail. The plot cover and plant height of the tested Brachiaria accessions ranges from 20% to 100% and 66.05 cm to 155 cm, respectively. Generally, the entire Brachiaria accessions were established successfully and attained optimum ground cover within 16 weeks after establishment (Table 1). The Brachiaria accessions *U. ruziziensis* ILRI-14813 (100%) and *U. decumbens* ILRI-14721 (100%) were consistently recorded higher plot cover than the rest accessions at all study sites. (Table 1). This shows that the soil and agro ecology were favorable for the indicated Brachiaria accessions. In contrast *U. decumbens* ILRI-10871 revealed significantly the lowest plot cover (20%) and maintained the lowest plant height (66.05 cm). The difference in plot cover among the grass accessions might be attributed to differences in regeneration ability and growth rate among the grasses accessions genetically. Plant height was shown significant differences ( $p < 0.05$ ) at 16 weeks after establishment among the Brachiaria accessions (Table 1). The difference in plant height might be related to their different in growth habit.

The brachiaria accessions tiller number ranges from 36.05 to 90.13. The lowest tiller number was recorded by *U. decumbens* ILRI-10871, while the rest had almost similar tiller number (Table 1). The highest dry matter yield was recorded from accessions *U. ruziziensis* ILRI-14813 (24.685 t/ha) followed by *U. decumbens* ILRI-14721 (20.89t/ha); whereas the lowest dry matter yield was recorded from *U. decumbens* ILRI-10871 (5.92 t/ha) (Table 1). The higher dry matter yield of these accessions might be

related to their high plant height. The results agreed with the findings of Tessema *et al.* (2003) who reported that increasing foliage height had a direct relationship with increased foliage herbage yield. The higher dry matter yield might be also due to their high tiller number, plot cover and plant height. Because the tiller numbers increased the chances of survival for most grasses and that large number of tillers produced allowed grasses to attain relatively high dry matter. The results are in agreement with Mganga (2009) who reported that tillering ability increases dry matter yield. The DM yield obtained in the current study was higher than the findings of Hare *et al.* (2009) who reported 16.3 t/ha DM yield. However, the result is almost similar to those of Hare *et al.* (2013a) and FAO (2015) who reported that DM yield of *Brachiaria* grass up to 20 t/ha. The accessions *U. ruziziensis* ILRI-14813 and *U. decumbens* ILRI-14721 had 75.36 % and 48.37% forage dry matter yield advantage over the local check (14.08 t/ha), respectively.

Table 1. Composite mean of agronomic parameters and herbage dry matter yields of *Brachiaria* accessions at lowland agro ecology.

Treatments	GC %	PH (cm)	TN	DMY(t/ha)
<i>U. decumbens</i> ILRI-10871	20.00 <sup>d</sup>	66.05 <sup>c</sup>	36.05 <sup>c</sup>	5.92 <sup>d</sup>
<i>U. decumbens</i> ILRI-13205	58.33 <sup>c</sup>	98.67 <sup>bc</sup>	90.13 <sup>a</sup>	9.31 <sup>cd</sup>
<i>U. ruziziensis</i> ILRI-13332	69.17 <sup>bc</sup>	135.28 <sup>ab</sup>	57.67 <sup>bc</sup>	12.29 <sup>bcd</sup>
<i>U. decumbens</i> ILRI-14720	98.33 <sup>a</sup>	131.67 <sup>ab</sup>	73.94 <sup>ab</sup>	11.13 <sup>cd</sup>
<i>U. decumbens</i> ILRI-14721	100.00 <sup>a</sup>	155.00 <sup>a</sup>	81.10 <sup>ab</sup>	20.89 <sup>ab</sup>
<i>U. ruziziensis</i> ILRI-14743	81.67 <sup>abc</sup>	127.17 <sup>ab</sup>	78.48 <sup>ab</sup>	12.66 <sup>bcd</sup>
<i>U. decumbens</i> ILRI-10871	90.83 <sup>ab</sup>	148.50 <sup>a</sup>	79.06 <sup>ab</sup>	15.30 <sup>bc</sup>
<i>U. ruziziensis</i> ILRI-14774	69.17 <sup>bc</sup>	119.17 <sup>ab</sup>	71.55 <sup>ab</sup>	8.30 <sup>cd</sup>
<i>U. ruziziensis</i> ILRI-14813	100.00 <sup>a</sup>	143.00 <sup>ab</sup>	82.19 <sup>ab</sup>	24.69 <sup>a</sup>
Local check	98.33 <sup>a</sup>	131.83 <sup>ab</sup>	79.28 <sup>ab</sup>	14.08 <sup>bc</sup>
<b>CV (%)</b>	30.62	33.24	36.7	36.42
<b>LSD (0.05)</b>	13.89	24.11	15.457	4.5007
<b>p-value</b>	**	**	*	**

LSD: least significant difference, GC: ground cover, DMY: dry matter yield, PH: plant height, TN: tiller number, CV: Coefficient of variation, SL: significance level

### ***Nutritional quality of Brachiaria grass accessions at Lowland agro ecology***

The composite mean of proximate composition (OM%, Ash%, CP%, NDF%, ADF% and ADL %) of *Brachiaria* accessions during the study time are presented in Table 2. The result of analysis of variance on OM%, Ash%, CP%, NDF%, ADF% and ADL% were indicated significant ( $p < 0.05$ ) among the tested accessions in this trial (Table 2). The highest CP% was recorded from *U. decumbens* ILRI-13205 (10.87%) accession, in contrast the lowest was recorded from accessions *U. ruziziensis* ILRI-13332 (8.08%). The results were comparable to values obtained by Villela *et al.* (2008), who found that the *brachiaria decumbens* had CP content of 9.43% in autumn and Hare *et al.* (2009), who recorded average CP ranging from 9.8 to 11.8% for *brachiaria* grass. However, the results were lower than the values of CP 13-16% recorded by Vendramini *et al.* (2014) for *brachiaria* grass. Generally, the CP content in all the *Brachiaria* grasses accessions ranges 8.08-10.87% (Table 2). This result was almost similar to 7-10% reported by Nguku *et al.* (2015).



The neutral detergent fiber (NDF %) content of the grasses was significantly ( $p < 0.05$ ) affected among the tested accessions and ranges from 57.94-64.65. This result was lower than result obtained by Pariz *et al.* (2011) who reported 66.4%-74.3% for brachiaria grasses. The highest NDF% concentration was obtained from accessions U. decumbens ILRI-10871 (64.65%) followed by U. ruziziensis ILRI-13332 (64.09%). U. ruziziensis ILRI-14743 (63.93%), U. ruziziensis ILRI-14774 (64.10%) and local check (63.78%) but statistically at par, while lower NDF% value was recorded from accessions U. decumbens ILRI-14720 (57.94%) and U. decumbens ILRI-13205 (58.12%) (Table 2). The higher NDF% value obtained from the current study was comparable with that of Pariz *et al.* (2011), who reported higher NDF% ( $> 60\%$ ) for brachiaria ruziziensis. The ADL% content of the Brachiaria grass were statistically similar among the tested accessions, except accessions U. decumbens ILRI-10871 (5.39%) and U. ruziziensis ILRI-14743 (5.29%) which had significantly ( $p < 0.05$ ) higher than the local check (4.56%) (Table 2).

Table 2. Composite mean value of chemical composition of the Brachiaria accessions at lowland agro ecology

Treatments	DM	Ash	CP	NDF	ADF	ADL
U. decumbens ILRI-10871	92.127 <sup>a</sup>	9.63 <sup>a</sup>	9.17 <sup>b</sup> <sup>c</sup>	64.65 <sup>a</sup>	45.38 <sup>a</sup>	5.39 <sup>a</sup>
U. decumbens ILRI-13205	90.84 <sup>abc</sup>	9.09 <sup>abc</sup>	10.87 <sup>a</sup>	58.12 <sup>c</sup>	42.76 <sup>bc</sup>	4.83 <sup>ab</sup>
U. ruziziensis ILRI-13332	91.38 <sup>ab</sup>	8.37 <sup>c</sup>	8.08 <sup>c</sup>	64.09 <sup>a</sup>	41.53 <sup>cd</sup>	4.91 <sup>ab</sup>
Local check	88.92 <sup>c</sup>	8.86 <sup>abc</sup>	9.06 <sup>bc</sup>	63.78 <sup>a</sup>	42.03 <sup>cd</sup>	4.56 <sup>b</sup>
U. decumbens ILRI-14720	89.81 <sup>bc</sup>	9.42 <sup>ab</sup>	10.34 <sup>ab</sup>	57.94 <sup>c</sup>	41.79 <sup>cd</sup>	4.09 <sup>ab</sup>
U. decumbens ILRI-14721	90.68 <sup>abc</sup>	9.24 <sup>ab</sup>	9.13 <sup>b</sup> <sup>c</sup>	61.94 <sup>b</sup>	41.12 <sup>cd</sup>	4.83 <sup>ab</sup>
U. ruziziensis ILRI-14743	91.76 <sup>ab</sup>	8.86 <sup>abc</sup>	8.36 <sup>c</sup>	63.93 <sup>a</sup>	44.93 <sup>ab</sup>	5.29 <sup>a</sup>
U. decumbens ILRI-10871	90.42 <sup>abc</sup>	9.35 <sup>ab</sup>	9.19 <sup>bc</sup>	61.02 <sup>b</sup>	43.12 <sup>abc</sup>	4.99 <sup>ab</sup>
U. ruziziensis ILRI-14774	89.30 <sup>c</sup>	9.23 <sup>ab</sup>	8.09 <sup>c</sup>	64.10 <sup>a</sup>	41.19 <sup>cd</sup>	4.83 <sup>ab</sup>
U. ruziziensis ILRI-14813	90.06 <sup>abc</sup>	8.58 <sup>bc</sup>	10.03 <sup>ab</sup>	61.29 <sup>b</sup>	40.23 <sup>d</sup>	5.03 <sup>ab</sup>
CV (%)	1.31	6.1	10.05	1.53	3.47	12.59
LSD	0.97	0.45	0.76	0.78	1.2	0.31
p-value	**	**	**	***	**	*

DM: Dry matter, CP: Crude protein, NDF: Neutral Detergent Fiber, ADF: Acid Detergent Fiber, ADL: Acid Detergent Lignin, LSD: least significant difference, CV: coefficient of variation

### ***Agronomic and yield performances of Brachiaria grass accessions at midland agro ecology***

The result of combined analysis of variance showed that ground cover, plant height, tiller number and dry matter yield at harvesting was significantly difference ( $p < 0.05$ ) among the tested Brachiaria accessions (Table 4). Since the interaction among the treatment and location is non-significant combined analysis was used. The ground cover and plant height of the grass ranges from 20% to 100% and 58.67 cm to 101.7 cm, respectively. The highest (87.23%) mean ground cover was recorded for Brachiaria grass accessions U. ruziziensis ILRI-14813, while the lowest (62.00%) mean ground cover was obtained from U. ruziziensis ILRI-14743 The Brachiaria grass U. decumbens ILRI-14721 and U. ruziziensis ILRI-14813 accessions were significantly ( $p < 0.001$ ) taller than U. decumbens ILRI-10871 and U. ruziziensis ILRI-14774 accessions. However, no significant differences observed in plant height between U. decumbens ILRI-14721 and U. ruziziensis ILRI-14813 accessions. There was statistically significant variation ( $p < 0.004$ ) was observed in numbers of tillers among the brachiaria grass accessions evaluated. The higher

(69.95%) tiller number was recorded from *Brachiaria* grass accession U. ruziziensis ILRI-14813, while U. decumbens ILRI-10871 had the lower tiller number.

There were also significant ( $p < 0.001$ ) differences among the *Brachiaria* accessions on DM yields in t/ha. The mean value of herbage DM yield was ranged between 7.27 – 17.83 t/ha. The highest mean value was obtained from U. ruziziensis ILRI-14813 (17.83 t/ha) followed by U. decumbens ILRI-14721 (16.57 ton/ha) accessions. The higher dry matter production of these two accessions might be related to their higher plant height and ground cover. This result was supported by Mganga (2009) and Nguku *et al.*, (2016), who observed that pasture species which grow fast and tall are more efficient, competitive and higher biomass production. The higher mean value of DM yields in the current study was comparable with that of Hare *et al.*, (2009), who reported 16.3 t/ha annual DM yield for *brachiaria* grass in Thailand.

Table 3. Composite mean Agronomic and yield of *Brachiaria* grass accessions at midland agro ecology

Treatments	GC (%)	PH (cm)	TN	DMY (t/ha)
U. decumbens ILRI-10871	65.31 <sup>b</sup>	58.67 <sup>bc</sup>	37.60 <sup>c</sup>	7.27 <sup>d</sup>
U. decumbens ILRI-13205	67.0 <sup>b</sup>	74.33 <sup>abc</sup>	46.34 <sup>abc</sup>	9.98 <sup>cd</sup>
U. ruziziensis ILRI-13332	70.34 <sup>ab</sup>	76.33 <sup>abc</sup>	53.35 <sup>abc</sup>	11.37 <sup>cd</sup>
U. decumbens ILRI-14720	80.34 <sup>ab</sup>	80.0 <sup>abc</sup>	54.60 <sup>abc</sup>	10.97 <sup>cd</sup>
U. decumbens ILRI-14721	78.68 <sup>ab</sup>	101.67 <sup>a</sup>	68.32 <sup>ab</sup>	16.57 <sup>ab</sup>
U. ruziziensis ILRI-14743	62.00 <sup>b</sup>	82.33 <sup>abc</sup>	53.40 <sup>abc</sup>	8.77 <sup>cd</sup>
U. decumbens ILRI-10871	66.99 <sup>b</sup>	86.67 <sup>abc</sup>	44.64 <sup>abc</sup>	10.20 <sup>cd</sup>
U. ruziziensis ILRI-14774	62.01 <sup>b</sup>	56.33 <sup>c</sup>	44.00 <sup>abc</sup>	12.18 <sup>bc</sup>
Local check	76.67 <sup>ab</sup>	76.00 <sup>abc</sup>	42.48 <sup>bc</sup>	7.43 <sup>cd</sup>
U. ruziziensis ILRI-14813	87.23 <sup>a</sup>	91.67 <sup>a</sup>	69.95 <sup>a</sup>	17.83 <sup>a</sup>
CV (%)	9.6	21.6	17.2	14.6
LSD (5%)	11.78	14.94	15.09	2.83
P-value	**	**	**	**

GC=Ground cover, PH=Plant height, TN=tiller number; DMY=Dry matter yield; CV= Coefficient of variation

#### ***Nutritional quality of Brachiaria grass accessions at Midland agro ecology***

Nutritional qualities of the tested *Brachiaria* grass accessions are presented in Table 4. The result of analysis of variance for DM, Ash, CP, NDF, ADF and ADL content were indicated significantly different ( $P < 0.001$ ) among the *Brachiaria* accessions. Local check had higher NDF content (72.73%), while U. ruziziensis ILRI-14813 was produced lower NDF (60.34%). The overall mean value of ADF obtained from *Brachiaria* grass accessions were ranged from 28.68-35.21%. Accession U. ruziziensis ILRI-14774 and U. ruziziensis ILRI-14743 were recorded the highest (34.83 and 35.21) % ADF values, respectively among the tested grass accessions. This is specified that these accessions considerably enhanced less digestible than the other *Brachiaria* accessions. This result was comparable with the work of Albayrak *et*

al. (2011), who reported that as the content of ADF increased the digestibility of the forage usually decreased. The least square mean value of all the Brachiaria accessions excluding *U. decumbens* ILRI-13205, *U. decumbens* ILRI-10871 and *U. decumbens* ILRI-14720 accessions had recorded higher CP content (13.29-14.45%) than the local check (11.76%). Brachiaria grass accession *U. decumbens* ILRI-14721 accumulated the higher CP content than the other evaluated brachiaria grass accessions. All the Brachiaria accessions considered in the trial were recorded lower NDF than local check and consequently they were more digestible than the local check. The CP content of the whole accessions except *U. decumbens* ILRI-13205, *U. decumbens* ILRI-10871, *U. decumbens* ILRI-14720 and local check were higher when compared with the findings of Nguku (2015), who reported CP content of 7-12.8% in a semi-arid region of Kenya.

Table 4. Chemical composition of Brachiaria grass accessions at midland agro ecology

Treatment	DM%	Ash%	CP%	NDF%	ADF%	ADL%
<i>U. decumbens</i> ILRI-10871	91.39 <sup>b</sup>	11.59 <sup>b</sup>	12.49 <sup>ef</sup>	68.07 <sup>c</sup>	32.21 <sup>b</sup>	8.24 <sup>ab</sup>
<i>U. decumbens</i> ILRI-13205	91.13 <sup>bc</sup>	11.7 <sup>b</sup>	10.49 <sup>g</sup>	62.45 <sup>ef</sup>	32.2 <sup>b</sup>	7.72 <sup>ab</sup>
<i>U. ruziziensis</i> ILRI-13332	90.8 <sup>de</sup>	12.36 <sup>a</sup>	13.42 <sup>bcd</sup>	64.93 <sup>d</sup>	30.52 <sup>c</sup>	8.24 <sup>ab</sup>
<i>U. decumbens</i> ILRI-14720	90.45 <sup>f</sup>	11.16 <sup>c</sup>	12.6 <sup>def</sup>	61.78 <sup>fg</sup>	28.53 <sup>d</sup>	7.32 <sup>bc</sup>
<i>U. decumbens</i> ILRI-14721	90.73 <sup>def</sup>	11.73 <sup>b</sup>	14.45 <sup>a</sup>	64.01 <sup>de</sup>	32.38 <sup>b</sup>	5.54 <sup>d</sup>
<i>U. ruziziensis</i> ILRI-14743	90.92 <sup>cd</sup>	12.42 <sup>a</sup>	13.96 <sup>abc</sup>	70.64 <sup>b</sup>	35.21 <sup>a</sup>	8.71 <sup>a</sup>
<i>U. decumbens</i> ILRI-10871	91.12 <sup>bc</sup>	11.48 <sup>b</sup>	14.18 <sup>ab</sup>	69.25 <sup>bc</sup>	28.56 <sup>d</sup>	6.04 <sup>d</sup>
<i>U. ruziziensis</i> ILRI-14774	90.55 <sup>ef</sup>	10.6 <sup>d</sup>	13.29 <sup>cde</sup>	63.39 <sup>def</sup>	34.83 <sup>a</sup>	6.24 <sup>d</sup>
<i>U. ruziziensis</i> ILRI-14813	92.09 <sup>a</sup>	11.46 <sup>b</sup>	13.71 <sup>abc</sup>	60.34 <sup>g</sup>	28.68 <sup>d</sup>	6.51 <sup>cd</sup>
Local check	90.0 <sup>g</sup>	11.6 <sup>b</sup>	11.76 <sup>f</sup>	72.73 <sup>a</sup>	29.68 <sup>cd</sup>	7.41 <sup>bc</sup>
CV	1.4	1.8	2.7	1.6	1.3	4.9
LSD	0.17	0.16	0.52	1.22	0.72	0.60
p-value	***	***	***	***	***	***

DM: Dry matter, CP: Crude protein, NDF: Neutral Detergent Fiber, ADF: Acid Detergent Fiber, ADL: Acid Detergent Lignin, LSD: least significant difference, CV: coefficient of variation

## Conclusions and Recommendations

The current study was revealed that the brachiaria accessions had more tillers, ground cover, plant height and dry matter yield during the production seasons in lowland than midland. Except *U. decumbens* ILRI-10871 DM yield for all the Brachiaria accessions was higher at lowland than at midland agro-ecology. Brachiaria grass accession of *U. ruziziensis* ILRI-14813 was recorded higher herbage dry matter yield followed by *U. decumbens* ILRI-14721. In both agro-ecologies, they were the most productive grasses and yielded 24.69 and 20.89 t/ha and 17.83 and 16.57 t/ha in lowland and midland, respectively. At lowland agro-ecology most Brachiaria accessions showed upright growth characteristics, whereas in midland the same accessions showed a spreading growth habit which possibly had adverse effect on herbage DM production. Additionally, Brachiaria grass accessions *U. ruziziensis* ILRI-14813 and *U. decumbens* ILRI-14721 had produced better nutritive value among the other accessions were tested, In conclusion, the two Brachiaria accessions were produced optimum herbage yield and nutrient quality at lowland and midland areas and should be recommended for promising forage biomass production at the study areas and other areas with similar agro ecologies.

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# **Apiculture Research Results**

# Establishing honey bee floral calendar in West Arsi and East Shewa zones of Oromia, Ethiopia

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## Abstract

Adequate knowledge of the honey bee flora is important for beekeeping production and the study was under taken to identify and document honeybee plants in West Arsi and East shoa Zones using melissopalynological analysis of honey samples, pollen load collection , plant inventory, structured questionnaires and field observation. Eighteen honey samples (18) were collected from different parts of the zones for honey pollen analysis. Accordingly a total of 83 plant species belonging to 46 families in West Arsi zone and 69 plant species belonging to 36 families in East shoa zone were identified as honeybee source plants. Fifty eight (58) plant species were identified as honey source plants based on melissopalynological analysis of honey. Among the identified plant species, *Guizotia scarba* (89.7%), *Eucalyptus globulus* (69.2%), *Eucalyptus camadulensis* (66.2%), *Acacia tortolis* (60.9%), *Schefflera abyssinica*(70.2%) and *Croton macronstachys*(57.2%) are the most dominant honey source plants in honey .Out of the analyzed honey samples, for botanical compostion, 14 were identified as monofloral honeys and 4 as multifloral honeys .On the otherhand 14 honey samples were dominated by few major bee plants due to their abundance in addition to their quality for honey production.The flowering calendar of plants in the area indicated that two major honey flow periods starting from April to June and October to November were identified. The scarcity of honeybee forages were observed in July to August and January to February and upto March. In most districts of the West Arsi and east shoa zones, herbaceous honeybee forages were the dominant honey source plants during September to November flowering season. While, during March to May trees and shrubs were also abundant due to variation in phenological patterns of plants. Inorder to apply seasonal honeybee colony management practice , beekeepers should manage honeybee colonies following floral calendar of honeybee plants of the area to increase the honey production of the area.

**Keywords:** Bee plants, pollen, monofloral honey, phenology, plant diversity

## Introduction

Ethiopia is endowed with natural and cultivated flora and diverse agro-ecological and climatic condition that are well-suited for beekeeping (Fichtl & Admassu, 1994; Admassu *et al.*, 2014). Oromia region is one of the regional states in Federal Republic of Ethiopia rich in natural resources and favorable climatic condition for beekeeping development. One of the natural resources suitable for beekeeping is the forest resources which include Harena, Yayu, Dindin, Anfarara, Munessa, Jibat, Chilimo and Menagesha-Suba forests which are highly potential for beekeeping. Moreover the area is also known for the growth of different field cultaived crops such as oil crops, pulses, horticultural crops and cereals. In order to boost the honey production from these floral resources , identification and documentation of economic bee forages and establishing their flowering calendar is critical for the sub-sector development.

Despite the richness of the bee flora, suitable climatic condition and its accessibility to market,the knowledge of the bee flora in the region is incomplete. Therefore identification of bee plants and

establishment of the floral calendar of important bee plant species of the area has paramount importance for practical beekeeping such as adding super, reducing super, honey flow period of the area and to predict the frequency of honey harvest (Mardan, 1984, Nuru & Admassu, 2001, Desalegne, 2005).

Therefore preparation of flowering calendar in the mid rift valley region of east shoa and west Arsi zones would be important for better honey production and to improve managements of honeybee colonies based on different seasonal calendar bee forages of the area. However bee forages of the zones are not adequately identified and their relationships with seasonal colony management plan is not established to the required level to increase the honey production. Moreover areas with unique production opportunities are not identified in both zones with aim of contributing to wards to the economy of beekeepers, in the area. Thus study was designed to characterize major bee forages, prepare flowering calendar and to assess the mono floral honey types in the area.

## **Materials and Methods**

### ***Description of the study area***

The study was conducted in West Arsi and East Shewa zones of Oromia. From each zones three districts were selected based on potentiality of area for beekeeping and agro-ecological condition of the area.

### ***Survey of the bee forages***

For this survey, well-structured questionnaires or checklist was developed. Both primary and secondary data were collected from the respective districts of each zone. Moreover Participatory Rural Appraisal (PRA) techniques through focused group discussion was employed. The group discussion with experts, community groups, development agents and farmer beekeepers were carried out to generate relevant information about the bee forages and floral calendar of the area.

### ***Sampling methods for collection of plant specimen***

For plant sample collection, each study zone was classified into three agro-ecologies (Highland, mid land and low lands) and from each agro-ecology one district was selected. Then the district was categorized into three PAs based on vegetation density, vegetation type and agro-ecology. Then samples of bee forage was collected from the selected PAs during the main flowering seasons using the standard Herbarium techniques and identified at National Herbarium of Addis Ababa University using the published flora books and Herbarium specimen. Moreover field observation was made to determine whether the plant is visited by honeybees, flowering period and food sources.

### ***Preparation of reference materials***

In order to prepare reference pollen slide, fully matured but un-opened flower buds were collected. Collection of un-open buds is to allow anthesis to take place in a closed environment so that other pollens present in the air, especially those from anemophilous plants, do not be contaminate the pollen samples. Then reference slide was prepared following method prescribed by the International Commission for bee botany (Louveaux et al., 1978). For this, either the ripe anthers, or the entire flower was picked and washed with ether; after the remaining ether has evaporated, then the pollen grains were taken with a needle and a small drop of glycerinated jelly was added and placed on a watch glass to melt at 40 °C;



the pollen slide is then covered with a coverslip and observed under the light microscope. The prepared reference slide was used as a pollen data base for palynologic analysis or for determination of botanical origin of honey. In addition to the collection of the different kinds of pollen samples of the flower a photographic archive for each pollen type was taken to understand the pollen exina sculpture, the polar and the equatorial views of the pollen grains.

### ***Honey sample collection and Laboratory analysis***

Fresh honey samples from different agro-ecologies of the study area at different seasons were collected for laboratory analysis. The honey pollen analysis was made following the methods adopted by Louvuex *et al.* (1978) for determination of botanical origin and frequency of pollen grains in the honey. Ten grams of honey was dissolved in 20 ml of warm water (40°C). The solution was centrifuged for 10 min at 2500 r/ min, the supernatant solution was decanted and the sediments were collected (Erdtman, 1960). The sediments were rinsed with distilled water to enhance further extraction of pollen from honey, centrifuged for 5 min at 2500 r/min, and preserved. Then after, two slides were prepared from each honey sample and identified under a light microscope. Pollen types were identified by comparison with reference slides of pollen collected directly from the plants in the study area. For quantification of the pollen types, at least 500 pollen grains were counted from each samples. The percentage frequency of the pollen taxa in all the samples was calculated. The percentage of pollen count were allocated to one of four frequency classes for nectar source plants: predominant pollen types (>45% of the total pollen grains counted); secondary pollen types (16%-45%); important minor pollen types (3%-15%); and minor pollen types (<3%) (Louveaux, Maurizio, & Vorwohl, 1978). Honeys with predominant pollen types were considered as monofloral honey.

### ***Colony Establishment for pollen collection***

A total of 24 honeybee colonies were established in 18 sites of the 6 districts of the study zones. For each site 4 honeybee colonies were established (two for pollen trapping and two for honey harvesting). Pollen traps having 16% pollen trapping efficiency were fitted at the entrance of beehives and pollen loads were collected for 12 months. The collected fresh pollen samples were placed in the clean paper bags and left for 24hr to dry at room temperature. The dried pollen was sorted into colors and identified to genus or species level using the pollen atlas of Ethiopia (Nuru, 2002).

## **Results and Discussions**

### ***Seasonal availability of honeybee plants***

On the basis of the response of beekeepers, secondary data collection and focus group discussion a total of 83, 69 plant species belonging to 46, 36 plant families were identified as honeybee source plants in West Arsi and East shoa zones respectively. In East shoa zone 38.75% of the honeybee forages were flowered from September to October while 19.6% flowered March to May and 41.6% from June to August. Similarly in West Arsi zone 35.5% of the honey bee forage are flowered from September to November, 33.3% flowered March to May and 31.2% of the plant species flowered during dearth period (Dec-Feb and Jun-Aug). In terms of the growth habit of the plant in the zones, in west Arsi zone out of 83 species, 39.65% are trees, 33.72% are shrubs and 26.6% herbs including cultivated crops. Similarly in east shoa zone out of 69 species 44.8% are trees, 37.06% are shrubs and 18.1% are herbs (Appendix 1-

2). In east shewa and west Arsi zones experienced beekeepers also familiar with honeybee plants that provide good honey, when they bloom and for how long they remain in blooming. Furthermore, some beekeepers are always paying attention to monitor the herbaceous plants, shrubs and trees that are especially important for honeybees (Niguse & Haftom, 2015).

### *Melissopalynological Analysis*

On the basis of the honey pollen analysis a total 58 bee plant species were identified representing 29 families in the study districts of East Shewa and West Arsi zones Appendix 3. The families with the highest number of species composition were Fabaceae 9 (15.5%); Astraceae 5 (8.6%); and Brassicaceae, & Rosaceae each of them with 3 (5.1%) species; Anacardiaceae and Cactaceae each of them with two (3.4%) species and the others with one species.

The most predominant pollen types (> 45%) were recorded for *Eucalyptus camaldulensis*, *Acacia tortolis*, *Guizotia scabra*, *Eucalyptus globulus*, *Hypoestes forskalii* due to their massive flowering in the area (Table 1). The secondary pollen source plants (16-45%) recorded were *Vernonia spp*, *Bidens spp*, *plantago lanceolata*, *Justicia scmhperrina*, *Trifolium spp*, *Mangifera indica*, *Guizotia abyssinica*, and *Dovyalis abyssinica*. The minor important pollen source plant species (3-15%) were *Trifolium quartinianum*, *Zea mays*, *Olea europia* and *crisium schimperii* and the rest of the species were rare (<3%). The diversity of secondary and important minor honey source plant species were higher than predominant species.

The honey pollen analysis also showed that a number of anemophilous (wind pollinated) pollen grains from *Plantago lanceolatum*, *Andropogon abyssinica*, *Zea mays*, *Cyperus species* and *Sporobolus consimilis*. Some of the plant species were found more frequent in sample plots *Achyranthes aspera*, *Tagetes minuta*, *Balanite aegyptica*, *Leucas abyssinica* but did not appear in honey samples. This might be due to less potential for nectar and pollen and the species density of the bee forage per plot might be very low to attract honeybees.

In many districts of the zones, herbaceous honeybee flora were the dominant honey source plants during September to November due to disturbances and expansion of agricultural crops. Some of these herbaceous honeybee forage plants include *Guizotia spp*, *Bidens spp*, *Trifolium spp* and *Hypoestes spp* which grow in farm land and at the edge of the forest. However, in March to May majority of honey source plants were trees and shrubs in comparison to herbaceous. The tree species such as *Schefflera abyssinica*, *Acacia tortolis*, *Croton macrostachyus* and *Eucalyptus spp* are flowered in this season. Admasu *et al.* (2014) also stated that *Schefflera abyssinica*, *Acacia spp* and *Croton macrostachyus* are the most important honey producing trees and flowered from April to May. The high scarcity of honeybee forage was observed in July to August and January to February and upto March. When honeybee plant identification sorted as major and minor honeybee plants during the pollen analysis of the honey, the main problem was that a given honeybee plant is major in one district and minor in the other districts. This is due to variation in the abundance of the species and anthropogenic influences on plants. For example in Kofale *Guizotia scabra* was not widely distributed and considered as the minor honeybee plants (1.5% in honey pollen analysis) and 87.6% in Gimbichu District which is major honeybee plants. This indicated that the abundance of a given honeybee plant species has great impact on honey production potential and mislead to consider the plant as best honeybee forage plant. Thus adjusting a number of honeybee colonies with available resource is important to increase the productivities of honeybee

colonies by overcoming the problem of colony overstocking (Nuru *et al.*, 2017). Nigusse & Haftom (2015) also reported that a good beekeeping area is in which honeybee plants grow abundantly and with a relatively long flowering duration of bee forages. Hence, beekeepers should select appropriate site that comprises enough supply of nectar and pollen source plants within the flight range of honeybees for honey production (Crane, 1990).

Table 1: Predominant, Secondary, important minor and minor honey source plants in districts of east shoa and west Arsi Zone

District	Predominant pollen source (>45%)	Secondary pollen source (16-45)	Important minor pollen source (3-15)	Minor pollen source(< 3%)
Dugda	<i>Guizotia scabra</i> <i>Hypoestes forskoolii</i>		<i>Eucalyptus camaldulensis</i> <i>Trifolium quartinianum</i> <i>Zea mays</i> <i>Acacia tortolis</i>	<i>Plantago lanceolata</i> <i>Lepidium sativium</i> <i>Phyllanthus</i> <i>Olea europea</i> <i>Vernonia amygdalina</i> <i>Crisium schimperia</i> <i>Acacia saligna</i> <i>Acacia tortilis</i>
Gimbichu	<i>Guizotia scabra</i>	<i>Justicia scmhpserina</i>	<i>Eucalyptus camaldulensis</i> <i>Vernonia amygdalina</i> <i>Crisium schimperia</i> <i>Eucalyptus globulus</i>	<i>Plantago lanceolata</i> <i>Lepidium</i> <i>Olea europea</i> <i>Maesa lanceolata</i> <i>Hypoestes forskoolii</i> <i>Datura arborea</i>
Ada`a	<i>Guizotia scarba</i>		<i>Eucalyptus camaldulensis</i> <i>Phyllanthus spp</i> <i>Olea europa</i> <i>Lepidum sativium</i> <i>Plantago lanceolata</i>	<i>Sesamum indicum</i> <i>Rubus steudreri</i> <i>Vernonia amygdalina</i> <i>Trifolium spp</i> <i>Acacia spp</i> <i>Olea europea</i> <i>Plantago lanceolata</i> <i>Vernonia amygdalina</i> <i>Acacia abyssinica</i> <i>Acacia seyal</i>
Wando	<i>Eucalyptus camaldulensis</i>	<i>Guizotia spp</i> <i>Plantago lanceolata</i> <i>Mangifera indica L</i>	<i>Plantago lanceolata</i> <i>Lepidium sativium</i>	<i>Crambe hispanica</i> <i>Olea europa</i> <i>Bidens spp</i> <i>Ocimum basilicum</i> <i>Trifolium spp</i> <i>Phoenix abyssinica</i>
Kofale	<i>Guizotia scabra</i> <i>Acacia tortolis</i>	<i>Vernonia amygdalina</i> <i>Eucalyptus Camaldulensis</i> <i>Eucalyptus globolus</i>	<i>Schefflera abyssinica</i> <i>Guizotia abyssinica</i> <i>Vernonia auriculifera</i> <i>Rumex nervosus</i> <i>Trifolium quartinianum</i> <i>Ricinus communis</i> <i>Erythrina abyssinica</i> <i>Acacia abyssinica</i>	<i>Apodytes dimidiata</i> <i>Pterolobium stellatum</i> <i>Carissa spinarum L</i> <i>Croton macrostachyus</i> <i>Brassica carinata</i> <i>Medicago polymorpha L.</i> <i>Euphorbia nubica</i> <i>Dichrostachys cinera</i> <i>Acacia saligna</i> <i>Dovyalis abyssinica</i>

District	Predominant pollen source (>45%)	Secondary pollen source (16-45)	Important minor pollen source (3-15)	Minor pollen source(< 3%)
Arsi Nagele	<i>Croton macrostachyus</i> <i>Guizotia scabra</i> <i>Eucalyptus globulus</i>	<i>Vernonia auriculifera</i> <i>Hypoestes forskalii</i> <i>Eucalyptus Camaldulensis</i> <i>Guizotia abyssinica</i> <i>Dovyalis abyssinica</i> <i>Schefflera abyssinica</i>	<i>Vernonia amygdalina</i> <i>Eucalyptus globulus</i> <i>Cordia africana</i> <i>Buddleja polystachya</i> <i>Trifolium quartinianum</i> <i>Dombeya torrida</i> <i>Olea europea</i> <i>Acacia tortilis</i>	<i>Vicia faba</i> <i>Mangifera indica.</i> <i>Euphorbia abyssinica</i> <i>Prunus africana</i> <i>Ficus vasta</i> <i>Solanum incanum.</i> <i>Calpurnia aurea</i> <i>Milletia ferruginea</i> <i>Acacia seyal</i> <i>Persea americana</i>

### ***Monofloral honey types***

Based on the number of a total pollen count, out of 18 analyzed honey samples 14 were identified as mono floral honeys and 4 samples were identified as multifloral honeys. Based on this identification five dominant plant species contributing for monofloral honey production were identified. These are *Guizotia scabra*, *Eucalyptus camadulensis*, *Acacia tortolis*, *Schefflera abyssinica* and *Croton macronstachys*. The dominance of these plant species in honey samples is due to their abundance, nectar and pollen potentiality. *Eucalyptus camaldulensis* is major honey source plants that provided monofloral honey in wando district of west Arsi Zone. Gemechis (2013) also reported that *Eucalyptus* mono-honey comes mainly from *E.globulus* though there are other important species like *E. camaldulensis* and *E. citrodora* that serve as sources of honey. *Guizotia scabra* is major honey source plants in Gimbichu Dugda, Wando and Ada'a Districts of East Shewa and West Arsi Zones that provide mono floral honey (Table 2). Gemechis, 2013 also reported that *Guizotia mono* honey mostly comes from *G.scabra* and along with this plant *G. abyssinica* and other weeds which flower in the same season and partly contribute to this honey (Admassu *et al.*, 2014). Due to their wide distribution, *Guizotia scabra* and *Acacia tortolis* are predominant plant species in mid and lowlands of the both zones. Monofloral honeys of *Schefflera abyssinica* was also produced in kofale district of Wes Arsi (Table 2).

Microscopic analysis revealed that plant species variability is greatest in the minor pollen group (less than 3%), followed by the important minor pollen, secondary, and dominant groups. Sabo *et al.* (2011) also reported that variability is always small among pollen species in the dominant groups, while greater among minor pollen (less than 3%), important minor pollen and secondary pollen groups. Analysis of the pollen content of honey is used to investigate the provenance and provide a quantitative measure of floral origin for use in market (Louveaux *et al.*, 1978). It is very effective to determine and control the geographical origin of honeys and it also provides information about other important quality aspects (Werner *et al.*, 2004).

Table 2: Monofloral Honey collected from districts of east shoa and west Arsi with its botanical origin and honey harvesting Season

No. of sample	District	Plant species	Pollen frequency in (%)	Harvesting season
1	Dugda	<i>Guzizotia scarba</i>	89.7	September-october
2	Wando	<i>Eucalyptus</i>	69.2	December-january
3	Gimbichu	<i>Guzizotia scarba</i>	81.4	September-november
4	Ada'a	<i>Guzizotia scarba</i>	76.9	September-october
5	Dugda	<i>Guzizotia scarba</i>	89.3	September-october
6	Nagele Arsi	<i>Guzizotia scarba</i>	88.9	September-november
7	Ada'a	<i>Guzizotia scarba</i>	81.95	September-october
8	Gimbichu	<i>Guzizotia scarba</i>	87.6	September-november
9	Ada'a	<i>Guzizotia scarba</i>	82	September-october
10	Wando	<i>Eucalyptus camadulensis</i>	66.2	May-june
11	Wando	<i>Eucalyptus camadulensis</i>	54.9	May-june
12	Dugda	<i>Acacia tortolis</i>	60.9	March –may
13	Kofale	<i>Schefflera abyssinica</i>	70.2	April-june
14	Nagele Arsi	<i>Croton macronstachys</i>	57.2	May-june

### **Flowering period of major bee forages**

Majority (50.6%) of honey source plants identified from honey pollen analysis are flowered from September to November followed by March to May (29.6%) while 19.8% flowered June to August (Table 3-4). As a result Sept- Nov and March to May are considered as major honey harvesting season in in East shewa and West Arsi Zones. The high scarcity of honeybee forage was observed in July to August and January to February up to mid march in both zones. The flowering period of the plants is very short at lower altitudes of the rift valley due to high temperature, insufficient moisture and erratic rainfall while the flowering periods of the plants in the higher altitudes extends up to end of November due to availability of high rainfall which lasts until end of October. In lower altitudes of the rift the major honey flow seasons is during early October as compared to higher altitudes, which is during November, and minor flow during May-June due to availability of field crops and protected forests.

Table 3. Flowering seasons of major honeybee plants in East Shewa Zone

Botanical name	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
<i>Acacia abyssinica</i>				■	■							
<i>Acacia albida</i>									■	■	■	
<i>Acacia bussei</i>				■	■							
<i>Acacia etabaica</i>		■	■	■								
<i>Acacia lahai</i>												
<i>Acacia oerfata</i>			■	■	■							
<i>Acacia pilispina</i>									■	■	■	
<i>Acacia Senegal</i>									■	■	■	
<i>Acacia seyal</i>									■	■	■	■
<i>Acacia tortolis</i>			■	■	■							

Botanical name	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
<i>Balanites aegyptiaca</i>									██████████	██████████	██████████	██████████
<i>Bidens macroptera</i>									██████████	██████████	██████████	██████████
<i>Brassica carinata</i>									██████████	██████████		
<i>Carthamus tinctorius</i>									██████████	██████████	██████████	██████████
<i>Cordia africana</i>								██████████	██████████	██████████		
<i>Ocimum sanctum</i>									██████████	██████████		
<i>Croton macrostachyus</i>					██████████	██████████	██████████	██████████				
<i>Dovyalis abyssinica</i>		██████████	██████████	██████████								
<i>Ekebergia capensis</i>	██████████	██████████	██████████									
<i>E. Camaldulensis</i>			██████████	██████████	██████████							
<i>Eucalyptus globulus</i>			██████████	██████████	██████████							
<i>Euphorbia abyssinica</i>									██████████	██████████	██████████	
<i>Olea europea</i>					██████████	██████████	██████████					
<i>Papaya carica</i>									██████████	██████████	██████████	
<i>Guizotia scabra</i>									██████████	██████████	██████████	
<i>Rhus natalensis</i>									██████████	██████████	██████████	
<i>Vernonia amygdalina</i>	██████████	██████████	██████████									██████████
<i>Zea mays</i>												██████████

Table 4. Flowering seasons of major honeybee plants in West Arsi Zone

Botanical name	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
<i>Acacia abyssinica</i>									██████████	██████████	██████████	██████████
<i>Acacia oerfata</i>				██████████	██████████	██████████						
<i>Acacia seyal</i>				██████████	██████████	██████████			██████████	██████████	██████████	
<i>Acacia tortolis</i>				██████████	██████████	██████████						
<i>Balanites aegyptiaca</i>									██████████	██████████	██████████	
<i>Brassica carinata</i>									██████████	██████████		
<i>Callistemon citrinus</i>	██████████	██████████	██████████	██████████	██████████	██████████	██████████	██████████	██████████	██████████	██████████	██████████
<i>Carica papaya</i>										██████████	██████████	██████████
<i>Carissa spinarum</i>										██████████	██████████	
<i>Coffea arabica</i>	██████████	██████████	██████████	██████████								
<i>Cordia africana</i>									██████████	██████████	██████████	██████████
<i>Coriandrum sativum</i>									██████████	██████████	██████████	██████████
<i>Croton macrostachyus</i>					██████████	██████████	██████████	██████████				
<i>Datura stramonium</i>												
<i>Dovyalis abyssinica</i>		██████████	██████████	██████████	██████████							
<i>Ekebergia capensis</i>	██████████	██████████	██████████	██████████								
<i>Erythrina abyssinica</i>									██████████	██████████	██████████	██████████
<i>Eucalyptus Camaldulensis</i>			██████████	██████████	██████████	██████████						
<i>Eucalyptus globulus</i>			██████████	██████████	██████████	██████████						

Botanical name	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
<i>Euphorbia abyssinica</i>										██████████	██████████	██████████
<i>Euphorbia nubica</i>	██████████	██████████								██████████	██████████	
<i>Guizotia abyssinica</i>										██████████	██████████	
<i>Guizotia scabra</i>										██████████	██████████	
<i>Mangifera indica</i>										██████████	██████████	
<i>Maytenus obscura</i>	██████████	██████████										
<i>Nigella sativa</i>										██████████	██████████	
<i>Olea europaea</i>					██████████	██████████				██████████	██████████	
<i>Persea americana</i>										██████████	██████████	
<i>Prunus Africana</i>										██████████	██████████	
<i>Psidium guajava</i>										██████████	██████████	
<i>Schefflera abyssinica</i>			██████████	██████████								
<i>Schinus molle</i>										██████████	██████████	
<i>Vernonia amygdalina</i>	██████████	██████████										
<i>Vernonia auriculifera</i>	██████████	██████████										
<i>Zea mays</i>											██████████	██████████

### ***Plant inventory of bee forage***

As the result of the inventory 35 honeybee plant species were identified belonging to 26 families. The families with the highest number of species (from highest to lowest) were *Astraceae* with 8(22.8%) species; *Fabaceae* 3(8.5%); and *Myrtaceae*, *celastraceae* & *Brassicaceae* each of them with two (5.7%) species and the others with one species. From these 35 species 60.9%, 12.1% and 29.2% were Herbs, Trees and Shrubs respectively (Table 5). This result indicated that herbaceous plant species including cultivated crops have also great contribution for honey production.

Table: 5. List of honey bee plants identified from pollen Analysis in East shoa zone of oromia

<i>Plant spp</i>	Family	Local name	Herb
<i>Acacia spp</i>	Fabaceae	Xedecha	Tree
<i>Andropogon abyssinica</i>	Poaceae	Balami	Herb
<i>Aspilia africana</i>	Asteraceae		Herb
<i>Bersama abyssinica</i>	Meliantaceae	Lolchiisa	Tree
<i>Bidens pilosa</i>	Asteraceae	Samie/abare	Herb
<i>Bidens spp</i>	Asteraceae	Kelo	Herb
<i>Brassica carinata</i>	Brassicaceae	Gomana	Herb
<i>Carduus nyassanus</i>	Asteraceae	Koshoshila	Herb
<i>Caylusea abyssinica</i>	Resedaceae	Rendi/arranchi	Herb
<i>Cirsium schimperi</i>	Asteraceae	Kosheshila	Herb
<i>Datura arborea</i>	Solanaceae	Tiruba	Shrub
<i>Dracaena schizantha</i>	Agavaceae	Lankiso	Herb
<i>Echinops macrochaetus</i>	Asteraceae	Koshoshila	Herb
<i>Eucalyptus camadulensis</i>	Myrtaceae	Berzafi dima	Tree
<i>Eucalyptus globulus</i>	Myrtaceae	Berzafi adi	Tree
<i>Glycine max</i>	Fabaceae	Atara	Herb
<i>Grevillea robusta</i>	Proteaceae	Giravilia	Tree
<i>Guizotia scarab</i>	Asteraceae	Adaa	Herb
<i>Helminthotheca echioides</i>	Asteraceae		Herb
<i>Hypoestes forskoolii</i>	Acanthaceae	Dergu	Herb
<i>Lepidium sativum</i>	Brassicaceae	Feto	Herb

<i>Plant spp</i>	Family	Local name	Herb
<i>Maesa lanceolata</i>	Myrsinaceae	Abayi	Shrub
<i>Maytenus obscura</i>	Celastraceae	Kombolcha	Tree
<i>Musa X paradisiaca L.</i>	Musaceae	Muzi	Herb
<i>Ocimum basilicum</i>	Lamiaceae	Qinidaba	Herb
<i>Olea europea</i>	Oleaceae	Ejersa	Tree
<i>Phoenix reclinata</i>	Arecaceae	Meti	Tree
<i>Plantago lanceolata</i>	Plantaginaceae	Qorxobi	Herb
<i>Ranunculus multifidus</i>	Ranunculaceae	Buter cup	Herb
<i>Rosa abyssinica</i>	Rosaceae	Qaqawe	Shrub
<i>Rumex spp</i>	Polygonaceae	Mucha arba	Herb
<i>Rumex nepalensis</i>	Polygonaceae	Mucha arba	Herb
<i>Schinus molle</i>	Anacardiaceae	Qundu berbere	Tree
<i>Silybum marianum</i>	Asteraceae	Holy thistle	Herb
<i>Syzygiumguineense</i>	Myrtaceae	Bedesa	Tree
<i>Terminalia spp</i>	Combretaceae		Tree
<i>Terminalia brownie</i>	Rubiaceae	Galo	Shrub
<i>Trifolium spp</i>	Papilionoideae	Sidisa	Herb
<i>Vernonia adoensis</i>	Asteraceae	Sanqii farad	Shrub
<i>Vernonia leopoldi</i>	Asteraceae		Herb
<i>Vicia faba</i>	Fabaceae	Baqela	Herb

### Species abundance

The most abundant bee forage species from field inventory were *Hypoestes forskali*, *Guizotia scarba*, *Eucalyptus Camaldulensis*, *Eucalyptus globulus*, *Bidens spp*, *Andropogon abyssinica*, *Plantago lanceolata* and *Balanites aegyptiaca*, (Figure 1). The higher abundance of bee forages in sample plots is due to an adaptation of the plant species to local climate and soil condition suggesting that higher frequency occurrence of the plants, is an indicator of adaptation to the area. For instance *Hypoestes forskali* is the most frequent species in sample quadrat due to its growing habit under shade of different Acacia trees.

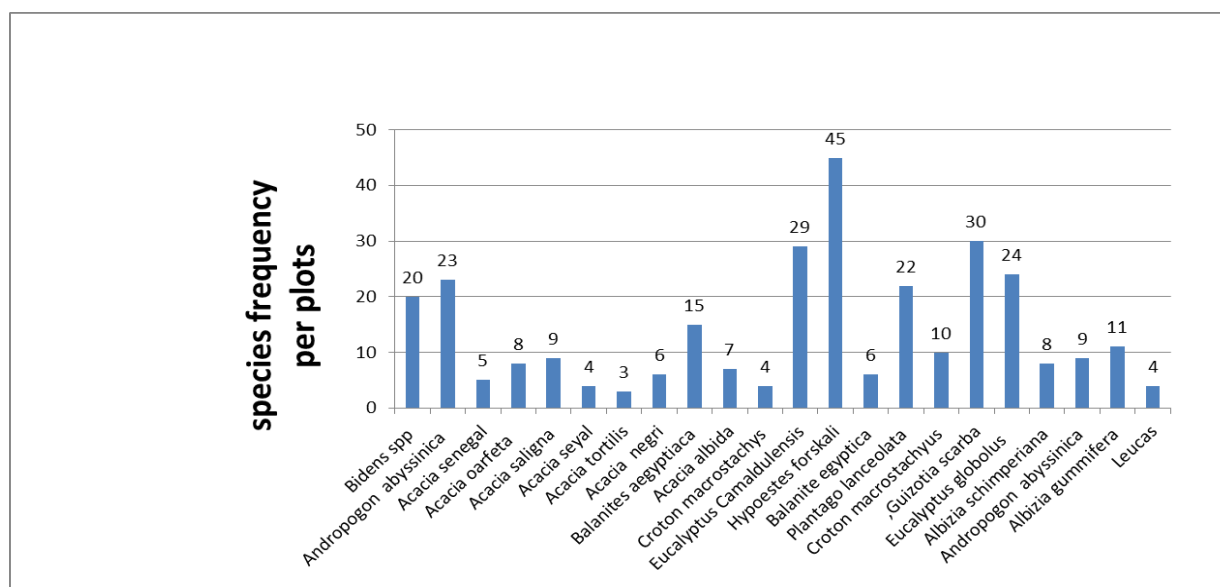


Figure 1. The frequency distribution of honey bee plant species in sample plots



**Plant density per plot**

The density and cover value of the plant species per plots were higher for herbaceous plants species (*Hypoestes forskaloi*, *Achyranthes aspera*, *Andropogon abyssinica*, *Tagetes minuta*, *Leucas spp* & *Solanum spp*) (Figure 2). This is due to smaller seed size, which occupies large areas of the plots. The trees and shrubs density was lower due to deforestation. However the cover values of trees were higher due to large canopy, which is advantageous for more flower production to be foraged by honeybees.

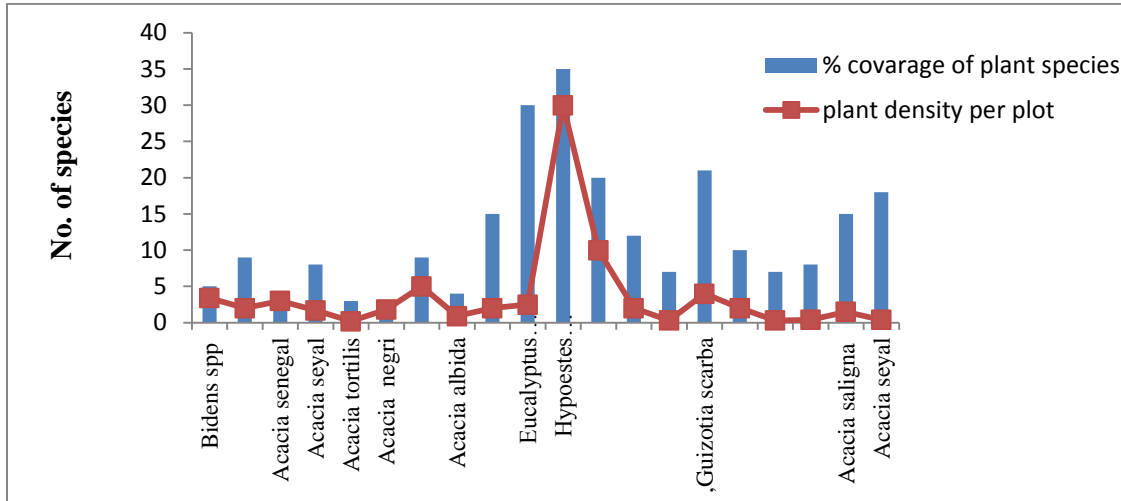


Figure 2. The relationship between plant density and cover abundance of plant species in sample plots

**Plant diversity**

The species diversity in sample plots were generally higher at the lower altitudes of the rift valley (Dugda, Ada'a & wando ) and decreasing to wards higher altitudes (Negale-Arsi Gumbichu & Kofale) (Figure 3). The higher diversity of bee forages at lower altitude of the mid riftvalley is due to the location of the study site in one of the Biodiversity Hotspot areas which is known for higher plant diversity. These agree with Friis et al. (2010), Tura et al (2019) who stated that the central riftvalley and low lands of Borena zone are known for higher plant diversity and encompassing endemic flora.

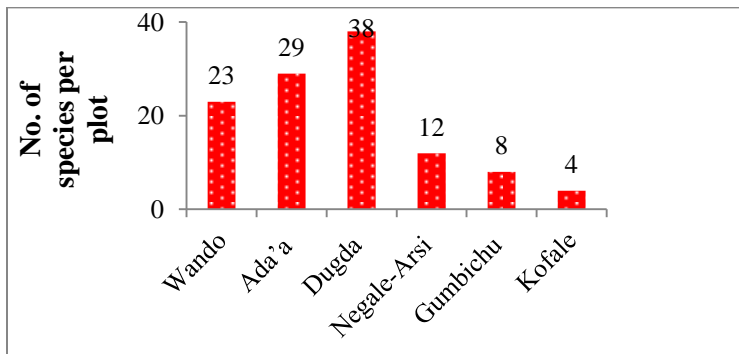


Figure 3. Plant diversity from different districts of the study area

## Conclusion and Recommendations

Based on field observation, field interview and honey pollen analysis 83 bee forage species were identified as honey source plants in the area. About 50% of the plants identified by the beekeepers were appeared in honey sample analysis indicating beekeepers knows bee forage plants and honeybees are efficiently utilizing local floral resource for production of honey. Five types of mono-floral honeys were identified which include : *Guzizotia scabra*, *Eucalyptus camadulensis*, *Acacia tortolis*, *Schefflera abyssinica* and *Croton macronstachyus* in East and West Arsi zones.

In many districts of the study area, herbaceous honeybee flora species are the dominant honey source plants from September to November. However, in March to May majority of honey source plants are trees and shrubs. The high scarcity of honeybee forage was observed in July to August and Dec- Jan to mid of March. Therefore, beekeepers should feed their honeybee colonies following floral calendar of honeybee plants.

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Appendix 1. Important honeybee plant species, habits and flowering periods based on survey and field observation in East shoa zone

<b>Botanical name</b>	<b>Family</b>	<b>Local name</b>	<b>Habit</b>	<b>Flowering period</b>
<i>Acacia abyssinica</i>	Fabaceae	Lafto	Tree	April-May
<i>Acacia albida</i>	Fabaceae	Garbi	Tree	March-April
<i>Acacia bussei</i>	Fabaceae	Ajoo	Tree	Jan-Feb
<i>Acacia etabaica</i>	Fabaceae	Hamaressa	Tree	Sep-Oct
<i>Acacia lahai</i>	Fabaceae	Xadacha	Tree	March- April
<i>Acacia oerfota</i>	Fabaceae	Ajjo	Herb	Sep-Oct
<i>Acacia pilispina</i>	Fabaceae	Girari	Tree	Oct-Nov
<i>Acacia saligna</i>	Fabaceae	-	Herb	Sep-Oct
<i>Acacia Senegal</i>	Fabaceae	Saphansa	Herb	Sept-Oct
<i>Acacia seyal</i>	Fabaceae	Waccu	Herb	Sept-Oct
<i>Acacia spp</i>	Fabaceae	-	Herb	Oct-Nov
<i>Acacia tortolis</i>	Fabaceae	Xadecha	Shrub	April- May
<i>Acanthus pubescens</i>	Acanthaceae	-	Tree	Sep-Oct
<i>Acokanthera schimperi</i>	Acanthaceae	-	Tree	April –June
<i>Agave Americana</i>	Agavaceae	Kancha	Herb	Sep-Oct
<i>Allium cepa</i>	Alliaceae	Qulubi dimma	Shrub	Oct-Nov
<i>Aloe macrocarpa</i>	Agavaceae	Arigessa	Shrub	April-May
<i>Balanites aegyptiaca</i>	Balanitaceae	Badeno	Tree	Sep-Oct
<i>Bidens macroptera</i>	Asteraceae	Kello	Shrub	Sep-Oct
<i>Brassica carinata</i>	Brassicaceae	Gommana	Herb	Oct-Nov
<i>Brassica napus</i>	Brassicaceae	Rafuu	Shrub	Sep-Oct
<i>Calpurnia aurea</i>	Fabaceae	Ceeke	Tree	Sept-Oct
<i>Carissa spinarum</i>	Apocynaceae	Agamasaa	Herb	Jul-Aug
<i>Carthamus tinctorius</i>	Asteraceae	Sufi	Shrub	Sep-Oct
<i>Celtis Africana</i>	Ulmaceae	-	Tree	Sept-Oct
<i>Cicer arietinum</i>	Fabaceae	Missira	Herb	Sept-Oct
<i>Citrus aurantifolia</i>	Rutaceae	Butikana	Tree	Dec-Jan
<i>Clematis simensis</i>	Ranunculaceae	Fiti	Shrub	May-June
<i>Cordia Africana</i>	Boraginaceae	wadessa	Shrub	Sep-Oct
<i>Croton macrostachyus</i>	Euphorbiaceae	Bakanisa	Tree	March-April
<i>Dichrostachys cinerea</i>	Fabaceae	Jerime	Herb	Sep-Oct
<i>Dodonaea anugustifolia</i>	Sapindaceae	Itaacha	Herb	Sep-Oct
<i>Dovyalis abyssinica</i>	Flacourtiaceae	Koshomi	Shrub	Feb-Mar
<i>Ehretia cymosa</i>	Boraginaceae	Ulagaa	Tree	Sept-Nov
<i>Ekebergia capensis.</i>	Meliaceae	Sombo	Tree	Jan- Feb
<i>Erythrina abyssinica</i>	Fabaceae	Sept-Oct	Shrub	Mar-Apr
<i>Eucalyptus camalduensis</i>	Myrtaceae	Barzafi dima	Tree	Mar-Apr
<i>Eucalyptus globules</i>	Myrtaceae	Adami Barzafi adi	Shrub	May-June
<i>Euphorbia abyssinica</i>	Euphorbiaceae	Adami	Tree	Oct-Nov
<i>Ficus sycomorus</i>	Moraceae	Oda	Herb	Sep-Oct

<i>Ficus sur</i> Forssk.	Moraceae	shola	Herb/crop	Sep-Oct
<i>Grevillea robusta</i>	Proteaceae	Gravillea	Shrub	Oct-Nov
<i>Grewia bicolor</i>	Tiliaceae	Haroressa	Shrub	Dec-Jan
<i>Grewia ferruginea</i>	Tiliaceae	Haroressa	Herb/crop	Sep-Oct
<i>Guizotia abyssinica</i>	Asteraceae	Nugi	Shrub	Oct-Nov
<i>Guizotia scabra</i>	Asteraceae	Tufo	Herb	Oct-Nov
<i>Helianthus annuus</i>	Asteraceae	Sufi	Herb/crop	Oct-Nov
<i>Hypoestes forskoolii</i>	Acanthaceae	Dergu	Shrub	Dec- Jan
<i>Justicia schimperiana</i>	Acanthaceae	Dhumuga	Tree	Oct-Nov
<i>Kalanchoe petitiiana</i>	Crassulaceae	Dodoti	Tree	Sept-Nov
<i>Lablab purpureus</i>	Fabaceae	Lablab	Tree	Sept-Nov
<i>Lantana camara</i>	Verbenaceae	Yewof kollo	Tree	Sept-Nov
<i>Lathyrus sativus</i>	Fabaceae	Gayyo	Tree	Sept-Nov
<i>Lens culinaris</i>	Fabaceae	Misira	Tree	Sept-Nov
<i>Leucaena leucocephala</i>	Fabaceae	Lukenia	Shrub	Sep-Oct
<i>Lippia adoensis</i>	Verbenaceae	Kussaye	Shrub	Sept-Nov
<i>Lonchocarpus laxiflora</i>	Fabaceae	-	Herb	Sep-Oct
<i>Mangifera indica</i>	Anacardiaceae	Mango	Tree	May-June
<i>Maytenus gracilipes</i>	Celastraceae	Kombolcha	Tree	Sep-Oct
<i>Medicago polymorpha</i>	Fabaceae	-	Tree	Sep-Oct
<i>Ocimum sanctum</i>	Lamiaceae	Bosobilla	Shrub	Sep-Oct
<i>Olea europea</i>	Oleaceae	Ejerssa	Shrub	Mar-April
<i>Otestegia minusli</i>	Lamiaceae	Tungiti	Shrub	Sep-Oct
<i>Papaya carica</i>	Caricaceae	Papya	Tree	Sep-Oct,
<i>Persea Americana</i>	Lauraceae	Avocado	Tree	Sep-Nov
<i>Pisum sativum</i>	Fabaceae	Atara	Tree	Sep-Nov
<i>Prunus Africana</i>	Roseaceae	Homi	Tree	Jan-Feb
<i>Pterolobium stellatum</i>	Fabaceae	Hargama	Tree	Sep-Oct
<i>Rhus natalensis</i>	Anacardiaceae	Xaxessa	Herb	Sep-Oct
<i>Sesbania sesban</i>	Fabaceae	sesbania	Herb	Sep-Oct
<i>Satureja paradoxa</i>	Lamiaceae	-	shrub	Dec- Jan
<i>Schinus molle</i>	Anacardiaceae	Qundoberbere	Herb	Sept-Oct
<i>Snowdenia polystachya</i>	Poaceae	-	herb	Nov-Dec
<i>Solanum incanum</i>	Solanaceae	Hidi	Tree	Sept-Oct
<i>Thymus schimperii</i>	Lamiaceae	Tosgnii	Herb	Sep-Oct
<i>Trichilia emetic</i>	Meliaceae	-	Tree	May-June
<i>Trigonella foenumgraecum</i>	Legumlnosae	Abeshii	Shrub	Feb-Mar
<i>Vernonia amygdalina</i>	Asteraceae	Ebicha	shrub	July-Aug
<i>Vicia faba</i>	Fabaceae	Bakela	Herb	Aug-Sep
<i>Zea mays</i>	Poaceae	Bokollo	Herb	Mar-April
<i>Ziziphus mucronata</i>	Rhmnaceae	Qurqura	Tree	Sept-Nov

Appendix 2. Important honeybee plant species, habits and flowering periods based on survey and field observation in Arsi Negelle district of West Arsi Zone

Botanical name	Family	Local Name	Habit	Flowering period
<i>Acacia abyssinica</i>	Fabaceae	Lafto	Tree	April-May
<i>Acacia oarjeta</i>	Fabaceae	Ajjo	Tree	April-May
<i>Acacia seyal</i>	Fabaceae	Waccu	Tree	March-April
<i>Acacia tortilis</i>	Fabaceae	Xadecha	Tree	April-May
<i>Acacia saligna</i>	Aloaceae	-	Tree	Sep-Oct
<i>Agava Americana</i>	Agavaceae	Qaca	Herb	Oct-Nov
<i>Albizia schimperiana</i>	Fabaceae	Sessa	Tree	Jan-Feb
<i>Allophylus abyssinicus</i>	Spindaceae	-	Tree	Aug-Sep
<i>Aloe macrocarpa</i>	Aloaceae	Argissa	shrub	Mar-April
<i>Andropolon abyssinicus</i>	Poaceae	Balami	Herb	Sept-Oct
<i>Apodytes dimidiata</i>	Icacinaceae	Calalaqa	Tree	Sep-Oct
<i>Balanites aegyptiaca</i>	Balanitaceae	Bedeno	Tree	Sep-Oct
<i>Brassica carinata</i>	Brassicaceae	Gomana	Herb	Sept- Nov
<i>Buddleia polystachya</i>	Loganiaceae	Adadao	Tree	Mar-Apr
<i>Callistemon citrinus</i>	Myrtaceae	Bottle brush	shrub	Sept - Jun
<i>Calpurnia aurea</i>	Fabaceae	Ceeke	Shrub	Oct-Nov
<i>Capparis tomentosa</i>	Capparidaceae	Gora	climber	May-June
<i>Carica papaya</i>	Caricaceae	Papaya	Tree/shrub	Aug-Sep
<i>Carissa spinarum</i>	Apocynaceae	Agamssa	shrub	April-May
<i>Cicer arietinum</i>	Fabaceae	Misera	Herb	Nov- Dec
<i>Citrus sinensis</i>	Rutaceae	Lemon	Shrub	Sept- Nov
<i>Clematis simensis</i>	Ranunculaceae	Fiti	climber	Dec-Jan
<i>Coffea Arabica</i>	Rubiaceae	Buna	Shrub	Jan-Feb
<i>Cordia africana</i>	Boraginaceae	Oda	Tree	July- Sept
<i>Coriandrum satium</i>	Ranunculaceae	Abadabo	Herb	Sep-Oct
<i>Croton macrostachys</i>	Euphorbiaceae	Dobbii	Tree	May
<i>Dombeyatorrida</i>	Sterculiaceae	Gravilia	Tree	Sep-Oct
<i>Dovyalis abyssinica</i>	Flacourtiaceae	Nugi	Shrub	Feb- Mar
<i>Ehretia cymosa</i>	Boraginaceae	Ada	shrub	Sep-Oct
<i>Ekebergia capensis</i>	Meliaceae	Sombo	Tree	Jan- Feb
<i>Ensete ventricosum</i>	Musaceae	Ensete	Herb	Oct-Nov
<i>Erythrina abyssinica</i>	Fabaceae	Walensu	Tree	Sep-Oct
<i>Eucalyptus Camaldulensis</i>	Myrtaceae	Bargamo dimma	Tree	March- May
<i>Eucalyptus globulus</i>	Myrtaceae	Bargamo adi	Tree	March- May
<i>Euphorbia abyssinica</i>	Euphorbiaceae	Adami	Tree	Sept- Nov
<i>Euphorbia nubica</i>	Euphorbiaceae	-	Shrub	Oct-Nov
<i>Ficus sycomorus</i>	Moraceae	Oda	Tree	Nov-Dec
<i>Ficus vasta</i>	Moraceae	Qilitu	Tree	Oct-Nov
<i>Ficussur Forssk.</i>	Moraceae	Harbu	Tree	Rainy season
<i>Galinsogaparviflora</i>	Asteraceae	Ejersa	Herb	Sept- Nov
<i>Grevillea robusta</i>	Proteaceae	Gravilla	Tree	Sept-Nov
<i>Guizotia abyssinica</i>	Asteraceae	Nugi	Herb	Sept-Nov
<i>Guizotia scabra</i>	Asteraceae	Mech	Herb	Sept-Nov

<i>Hagenia abyssinica</i>	Rosaceae	Hexo	Tree	Sep-Oct
<i>Hypericum quartinianum</i>	Hypericaceae	Ambelta	Shrub	Sep-Oct
<i>Hypoestes forskalii</i>	Acanthaceae	Dergu)	Dergu	Sept- Nov
<i>Justicia Schimeriana</i>	Acanthaceae	Dhmuga	Shrub	Sep-Oct
<i>Lantana camera</i>	Verbenaceae	-	shrub	Sept-Nov
<i>Lens culinaris</i>	Fabaceae	Misira	herb	Dec- Jan
<i>Leucaena leucocephala</i>	Fabaceae	Nyata hori	shrub	Sept-Nov
<i>Linum usitatissimum</i>	Linaceae	Talbaa	herb	Sept-Nov
<i>Lycopersicon esculentum</i>	Solanaceae	Timatimi	Herb	Nov-Dec
<i>Maessa lanceolata</i>	Myricineae	Abayii	Shrub	Aug- sept
<i>Mangifera indica</i>	Anacardiaceae	Mango	Tree	Jan-Feb
<i>Maytenus gracilipes</i>	Celastraceae	Komblocha	shrub	Oct-Nov
<i>Maytenus obscura</i>	Celastraceae	Komblocha	Shrub	Feb-March
<i>Medicago polymorpha</i>	Fabaceae	-	Herb	Sep-Oct
<i>Medicago sativa</i>	Fabaceae	alfalfa	Herb	Oct-Nov
<i>Nicotiana rustica</i>	Solanaceae	Tembo	Herb	Dec-Jan
<i>Nigella sativa</i>	Ranunculaceae	Abusuda gurcha	Herb	Sep-Oct
<i>Olea europea</i>	Oleaceae	Ejerssa	Tree	May
<i>Opuntia ficus-indica</i>	Cactaceae	Beles	Shrub	Dec-Jan
<i>Phytolacca dodecandra</i>	Phytolacceaeae	Andode	Shrub	Sep-Oct
<i>Pterolobium stellatum</i>	Fabaceae	Hargama	shrub	Sept- Dec
<i>Rhamnus prinoides</i>	Rhamnaceae	Gesho	shrub	Sept- Dec
<i>Ricinus communis</i>	Euphorbiaceae	qoboo	Shrub	Sep-Oct

Appendix 3. List of honeybee plants identified from Pollen analysis in west Arsi and East shewa Zones of Oromia

District	Botanical Name	Family	Local name	Habit
Wando	<i>Eucalyptus camadulensis</i>	Myrtaceae	Barzafi dima	Tree
	<i>Datura stramonium</i>	Solanaceae	Manji	Herb
	<i>Hibiscus rosensis</i>	Malvaceae		Herb
	<i>Justicia spp</i>	Acanthaceae	Dhumuga	Shrub
	<i>Plantago lanceolata</i>	Plantaginaceae	Korisa	Herb
	<i>Terminalia spp</i>	Combretaceae		Tree
	<i>Guizotia scabra</i>	Asteraceae	Ada	Herb
	<i>Datura arborea</i>	Solanaceae		Shrub
	<i>Croton macrostachyus</i>	Euphorblaceae	Bakanisa	Tree
	<i>Guizotia abyssinica</i>	Asteraceae	Nugi	Herb
	<i>Zea mays</i>	Poaceae	Boqolo	Herb
	<i>Ocimum bassilicum</i>	Lamiaceae	Basobila	Herb
	<i>Trifolium spp</i>	Leguminosae	Sidisa	Herb
	<i>Schinus molle</i>	Anacardiaceae	Barbare guracha	Herb
	<i>Vernonia amygdalina</i>	Asteraceae	Ebicha	Shrub
	<i>Hypoestes forskalii</i>	Acanthaceae	Dargu	Herb
	<i>Rosa abyssinica</i>	Rosaceae	Gora	Shrub
<i>Polyscias tulva</i>	Araliaceae	Ankisa	Tree	

	<i>Hagenia abyssinica</i>	Rosaceae	Kosoo	Tree
	<i>Clausena anisata</i>	Rutaceae	Walaya /adesa	Shrub
Kofale	<i>Eucalyptus globulus</i>	Myrtaceae	Berzafi adi	Tree
	<i>Vernonia spp</i>	Asteraceae	Reji/ebicha	Shrub
	<i>Brassica carinata</i>	Brassicaceae	Gomana	Herb
	<i>Justitia schimperana</i>	Acanthaceae	Dhumuga	Shrub
	<i>Rubus steudneri</i>	Rosaceae	Gumure/haltufa	Shrub
	<i>Cirsium schimperi</i>	Asteraceae	Kosheshila	Herb
	<i>Zea mays</i>	Poaceae	Boqolo	Herb
	<i>Rumex nervosus</i>	Polygonaceae	Dangego	Shrub
	<i>Trifolium spp</i>	Leguminosae	Siddisa	Herb
Gimbichu	<i>Plantago lanceolata</i>	Plantaginaceae	Korrisa	Herb
	<i>Guizotia spp</i>	Asteraceae	Adaa	Herb
	<i>Olea europea</i>	Oleaceae	Ejersa	Tree
	<i>Cirsium schimperi</i>	Asteraceae	Kosheshila	Herb
	<i>Dracaena schizantha</i>	Agavaceae	Lankiso	Herb
	<i>Maytenus obscura</i>	Celastraceae	Kombolcha	Tree
	<i>Bidens spp</i>	Asteraceae	Kelo	Herb
	<i>Vernonia leopoldi</i>	Asteraceae		Herb
	<i>Caylusea abyssinica</i>	Resedaceae	Rendi/arranchi	Herb
	<i>Helminthotheca echioides</i>	Asteraceae		Herb
	<i>Bersama abyssinica</i>	Melanthaceae	Lolchisa	Tree
	<i>Vernonia adoensis</i>	Asteraceae	Sanqi farda	Shrub
	<i>Carduus nyassanus</i>	Asteraceae	Koshoshila	Herb
	<i>Eucalyptus globulus</i>	Myrtaceae	Berzafi adi	Tree
	<i>Bidens pilosa</i>	Asteraceae	Samie/abare	Herb
	<i>Schinus molle</i>	Anacardiaceae	Barbare gurracha	Herb
	<i>Silybum marianum</i>	Asteraceae	Holytistle	Herb
	<i>Echinops macrochaetus</i>	Asteraceae	Koshoshila	Herb
	<i>Zea mays</i>	Poaceae	Boqolo	Herb
	<i>Glycine max</i>	Fabaceae	Atara	Herb
	<i>Maytenus spp</i>	Celastraceae	Kombolcha	Tree
	<i>Andropogon abyssinica</i>	Poaceae	Balami	Herb
	<i>Ocimum basilicum</i>	Lamiaceae	Urgoftu	Herb
	<i>Aspilia africana</i>	Asteraceae		Herb
	<i>Terminalia spp</i>	Combretaceae		Tree
	<i>Brassica carinata</i>	Brassicaceae	Gomana	Herb
Ada'a	<i>Zea mays</i>	Poaceae	Boqolo	Herb
	<i>Guizotia scabra</i>	Asteraceae	Ada	Herb
	<i>Eucalyptus camadulensis</i>	Myrtaceae	Barzafi dima	Herb
	<i>Vicia faba</i>	Fabaceae	Baqela	Herb
	<i>Acacia spp</i>	Fabaceae	Dedecha	Tree
	<i>MusaXparadisiacaL.</i>	Musaceae	Muzi	Herb
	<i>Brassica carinata</i>	Brassicaceae	Gomana	Herb
	<i>Ocimum basilicumL.</i>	Lamiaceae	Qinidaba	Herb
	<i>Rumex spp</i>	Polygonaceae	Mucha arba	Herb
<i>Trifolium spp</i>	Papilionoideae	Siddisa	Herb	



	<i>Bidens spp</i>	Asteraceae	Kelo	Herb
	<i>Cirsium schimperi</i>	Asteraceae	Koshoshila	Herb
	<i>Plantago lanceolata</i>	Plantaginaceae	Qorxobi	Herb
	<i>Echinops macrochaetus</i>	Asteraceae	Koshoshila	Herb
	<i>Datura arborea</i>	Solanaceae		Shrub
Dugda	<i>Guizotia scarab</i>	Asteraceae	Ada	Herb
	<i>Acacia spp</i>	Fabaceae	Dedecha	Tree
	<i>MusaXparadisiacaL.</i>	Musaceae	Muzi	Herb
	<i>Ocimum basilicum</i>	Lamiaceae	Qinidaba	Herb
	<i>Terminalia brownie</i>	Rubiaceae	Galo	Shrub
	<i>Zea mays</i>	Poaceae	Boqolo	Herb
	<i>Maesa lanceolata</i>	Myrsinaceae	Abayi	Shrub
	<i>Lepidium sativum</i>	Brassicaceae	Feto	Herb
	<i>Rumex nepalensis</i>	Polygonaceae	Mucha arba	Herb
	<i>Silybum marianum</i>	Asteraceae	Holy thistle	Herb
	<i>Rosa abyssinica</i>	Rosaceae	Qaqawe	Shrub
	<i>Glycine max</i>	Leguminosae	Soybean	Herb
	<i>Grevillea robusta</i>	Proteaceae	Giravilia	Tree
	<i>Phoenix reclinata</i>	Arecaceae	Meti	Tree
	<i>Eucalyptus camadulensis</i>	Myrtaceae	Berzafi dima	Tree
	<i>Ranunculus multifidius</i>	Ranunculacea	Buter cup	Herb
	<i>Syzygium guineense</i>	Myrtaceae	Bedesa	Tree
	<i>Schinus molle</i>	Anacardiaceae	Qunduberbere	Tree
	<i>Vicia faba</i>	Leguminosae	Bakela	Herb
	<i>Hypoestesforsaolii</i>	Acanthaceae	Dergu	Herb
Nagalle Arsi	<i>Guizotia spp</i>	Astrceae	Ada	Herb
	<i>Plantago lanceolata L.</i>	Plantaginaceae	Qorxobi	Herb
	<i>Dombeya aethiopica</i>	Sterculiaceae		Tree
	<i>Brassica carinata</i>	Brassicaceae	Gomana	Herb
	<i>Olea europea</i>	Oleaceae	Ejersa	Tree
	<i>Eucalyptus camaldulensis</i>	Myrtaceae	Berza dima	Tree
	<i>Zea mays L.</i>	Poaceae	Bokolo	Herb
	<i>Olea capensis</i>	Oleaceae	Agarguri	Tree
	<i>Vernonia auriculifera</i>	Asteraceae	Reji	Shrub
	<i>Dombeya torida</i>	Sterculiaceae	Danisa	Tree
	<i>Musa X paradisiaca L.</i>	Musaceae	Muzi	Herb
	<i>Rosa abyssinicaa</i>	Rosaceae	Qeqewi	Shrub
	<i>Echinopus macrochoetus</i>	Asteraceae	Koshoshila	Herb
	<i>Justicia schimperiana</i>	Acanthaceae	Dhumuga	Shrub
	<i>Croton macrostachyus Hochst.</i>	Euphorblaceae	Bekenis	Tree
	<i>Ocimum basilicum L.</i>	Lamiaceae	Damakase	Shrub
	<i>Acacia abyssinica</i>	Fabaceae	Xadacha	Tree
	<i>Guizotia abyssinica</i>	Astrceae	Nuugii	Herb
	<i>Maesa lanceolata</i>	Myrsinaceae	Galaba/abaye	Shrub
	<i>Rumex nepalensls</i>	Polygonaceae	Mucha arba	shrub
	<i>Justitiaschimperana</i>	Acanthaceae	Dumuga	Shrub
	<i>Plantago lanceolata</i>	Plantaginaceae	Korisa	Herb
	<i>Terminalia brownie</i>	Combretaceae	Bridhessa	Tree

# Establishment of floral calendar in Guji Zone of Southern Oromia Region, Ethiopia

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## Abstract

*The success of beekeeping primarily depends on the availability of bee flora based on its abundance, nectar and pollen yield and prolonged flowering periods. Identification and establishment of flowering calendar are important for appropriate site selection and for determination of honey flow period of the area. The study was carried out in east Guji zone of southern Oromia with the objective of characterizing and, documenting the major bee forages and establishing floral calendar for effective management of honeybee colonies in different agro-ecological condition of the area. For this study six districts were selected purposively and from each districts three PAs were selected. Honeybee colonies were established to collect pollen by fitting pollen traps having 16% pollen trapping efficiency at the entrance of honeybee colonies and pollen pellets were sorted by color and analyzed for bee forage identification. Honey samples were collected from established colonies and the honey pollen analysis was done for determination of botanical origin and frequency of pollen grains. On top of this field observation, inventory of bee forages and field interview were done to capture bee forage types and flowering calendar of the area. The result showed that more than 80 honeybee plant species were identified and three honey harvesting seasons including two major and one minor honey flow seasons were known in highland and midland agro-ecologies of the area. In addition seven types of monofloral honey types were identified namely Terminalia brownii, Guizotia scabra, Croton macrostachyus, Coffea arabica, Schefflera abyssinica, Eucalyptus globulus and Vernonia spp. However, from the information obtained from field interview and focus group discussion indicated that the diversity of bee flora and honey production potential of the area is declining at rapid rate due to expansion of farmland, unwise application of pesticide and herbicide. Thus conservation of major bee flora and effective management of the honeybee colonies following the floral calendar of bee flora in different agro-ecological condition of the area is recommended to increase harvesting frequency of honey.*

**Keyword:** Bee flora, floral calendar, Agro-ecology, Beekeeping, Plant inventory

## Introduction

Ethiopia is endowed with natural and cultivated flora and diverse agro-ecological and climatic condition that are well-suited for beekeeping. Oromia is one of the regional states in Federal Republic of Ethiopia rich in natural resources and favorable climatic condition for beekeeping development. Oromia region is characterized by plateau and very limited low land areas. Due to variation in geomorphology of the region and other environmental factors resulted for the presence of different natural vegetation suitable for beekeeping. Out of the 58 National Forest Priority Areas of the Country, 49 are found in Oromia. The well-known forest resource of Oromia rich in biodiversity include Hareenna, Yayu, Dindin, Anfarara, Munessa, Jibat, Chilimo and Managesha-Suba which are highly potential for honey production. The region also comprises cultivated crops such as oil crops, horticultural, spices and pulses, which can enhance beekeeping development further. The availability of conducive environment for beekeeping, the

region is leading in honey production accounting for 42.7% of the apiculture resources of the country with annual honey production volume of 24.8 thousand tones out of the total 54 thousand tones (CSA, 2017).

Guji zone is one of the 21 zones of Oromia regional state and is located at the southern part of the region. It is characterized by encompassing all agro-ecologies (highland, midland and lowland). As the result of different agro-ecologies which enables the zone to have rich diversity of bee flora resources highly suitable for honey production. There are more than five (5) different colored honeys in the zone indicating the zone is rich in diversity of bee flora.

In order to boost the honey production from this huge resource of the Zone, identification and documentation of economic bee forages and establishing their flowering calendar is critical for the sub-sector development. Establishing floral calendar is a critical tool for planning various beekeeping management operations such as supering bee hives, predicting the frequency and honey flow period of the area. Therefore identification and documentation of bee plants is important to characterize major bee forage plants and also to establish the flowering calendar for planning various beekeeping management operations. There are strong associations between the seasonal cycles of honeybee colonies and calendar of bee plants which can be applied in practical management of honeybee colonies. Timing of management colonies corresponding to phenological pattern of bee plants is critical in building up colony populations before the main nectar flow.

There is lack of information about bee forages and floral calendar of the Guji zone in relation to seasonal colony management which is entirely depend on the agro-ecological condition of the area. Moreover areas with unique production opportunities and honey production potential are not identified since beekeeping has immense contribution to the economy of local communities in the area. Thus assessing the availability of bee forage and establishing flowering calendar of honey plants in different agro-ecologies of the zone for effective seasonal colony management is paramount important for increased honey production. Therefore the study was aimed to characterize and document major bee forages contributing for honey production and to establish appropriate floral calendar for effective bee management of honeybee colonies in Guji zone.

## **Materials and Methods**

### ***Description of the study area***

The activity was carried out in six potential districts (Bore, A/Sorra, Adola, Shakiso, Gorodola and Liban) of East Guji zone of southern Oromia region from September 2019 to August 2021. Geographically, the study area is located at 5°20'03.830"– 6°21'40.709"N latitudes and 39°40'52.051"-38°36'56.051"E longitudes. For this study six districts and from each district three PAs were selected purposively based on agro-ecology.

### ***Colony establishment and pollen load collection***

Eighteen (18) honeybees' colonies were established at 12 sites of the six districts of Guji zone of Oromia. Pollen trap was fitted at the entrance of bee hives having the pollen trapping efficiency of 16% and pollen loads was collected four times per week. The pollen loads were sorted into colors and identified to the genus species level using the pollen atlas of Ethiopia (Nuru,2002).

### ***Honey pollen analysis***

The pollen analysis was done by following the methods adopted by Louvuexet *al.* (1978) for determination of botanical composition and frequency of pollen grains in the honey. Accordingly ten grams of honey sample was weighed and placed in centrifuge tube and added with 20 ml of distilled water to dissolve the honey. Then after the solution was centrifuged for 10 minutes and the supernatant liquid was decanted. Another distilled water of 20 ml was added to completely dissolve the remaining sugar crystals and centrifuged again for 5 minutes and supernatant was removed completely. The residue was spread evenly with a micro spatula on a microscope slide and allowed to dry. One drop of glycerin jelly was applied to the cover slip and the sample was examined through the microscope and the picture of the pollen was taken by the camera connected to the microscope (Carl ZEISS microscope Germany). The frequency occurrences of pollen were determined by counting 500 pollens from a single slide. The pollen count was converted into percent to calculate the predominant, secondary, tertiary and rare enrichment of honey plant pollen in honey samples.

### ***Plant inventory***

To assess the bee forages abundance and diversity, four transect lines was laid out from apiary sites to North, South, West and East within 2 km radius following GPS. Plant species was sampled systematically after 2 km distance from one to the other in order to avoid redundancy. Along these transects plots of 20x20 m was laid out within 400 m interval between the sample plots. In order to retain accuracy, five (5) small plots measuring 2x2m was laid out within the larger plot to capture herbaceous and grasses. All the plant species encountered in each sample plots was recorded and percentage cover of each species was estimated visually to determine frequency, density and abundance of bee forages. For those plant species which could not identified in the field, sample of bee plant specimens were collected, pressed and dried and identified at National Herbarium of Addis Ababa University. The Shannon-Wiener Index was calculated using the following formula.

$$\text{Shannon Index } (H') = \sum_{i=1}^s P_i \ln P_i$$

### ***Survey of bee forages***

For this study well-structured questionnaires was developed and both primary and secondary data were collected. The participatory Rural Appraisal (PRA) techniques were applied through focused group discussions. The group discussions with experts, development agents, farmer beekeepers, were done to generate relevant information about major honey source plants, floral calendar, major honey types, and honey harvesting period. Moreover, field observation was made on bee forage availability and abundance in the area.

## **Results and Discussions**

### ***Plant Inventory of bee Forages***

According to plant inventory of the bee forages in different all agro-ecologies of east Guji zone showed that 80 plant species belonging to 36 families were identified Appendix 1. Among the identified plant

families Asteraceae, Fabaceae, Lamiaceae, Acanthaceae and Euphorbiaceae comprising high species composition accounting for 20.25%, 17.72%, 7.5%, 7.79%, 3.79% respectively and the rest of the families are represented by one species. The growth forms of bee forage utilized by honeybees comprises Climber (4%), Herbs (27%), shrub (33%), and trees (36%) Figure1. The trees and shrubs are high in the area due to low level of disturbance and most the study areas are covered by natural forest including Anfara forest. The species diversity including Shannon wiener index and equitability and evenness of the area is 2.85 and 0.67 respectively which is higher bee forage diversity as compare to other known forests in southwest Ethiopia.

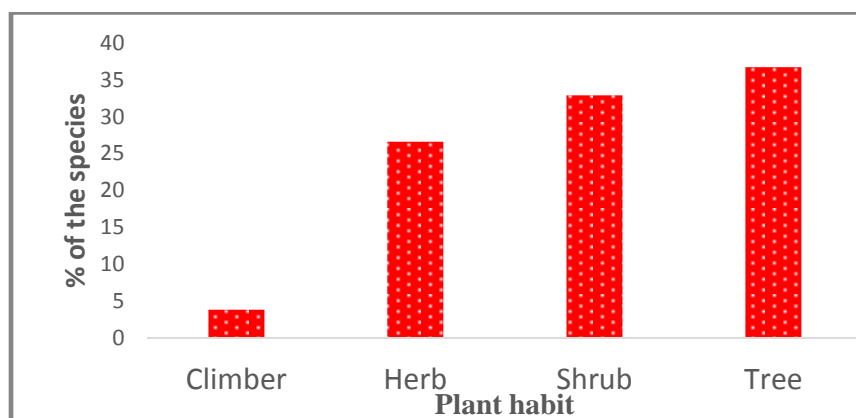


Figure 1. The growth habit of the bee forage in the area

### ***Flowering period and honey flow season of East Guji Zone***

According to analysis of the flowering period of bee forages 37% of the species flowered from Sept-Nov, 43% flowered Dec-Feb, 23% flowered from March–April and 13% from June-August. The presence of higher percentage of flowering species during Sept–Nov and Dec-Feb due to summer rain which starts in the middle of June and extends up to December Figure 2. Following the flowering period of bee forages three honey flow period was identified. From these three honey flow periods two are the major ones and one is the minor honey harvesting seasons. The two majors honey flow periods in the area are February to April up to the beginning of May and the minor honey flow period is June. During the first honey flow period, *Vernonia amygdalina*, *Vernonia auriculifera* and *Schefflera abyssinica* from which honeybees produce white mono floral honey. The second honey flow period was at the end of flowering of *Eucalyptus* spp and *Croton macrostachyus*. Honey flow seasons are varies based on the agro-ecologies, weather conditions and diversity of bee flora. The honey flow seasons of the area at different agro-ecologies were indicated in the (Table1).

Table 1:- Honey harvesting seasons in East Guji zone

Agro-ecology	Frequency	Major honey flow	Minor honey
Highland	3	February, April-May	June
Midland	3	February-March, June-July	September
Lowland	2	July-September, December	

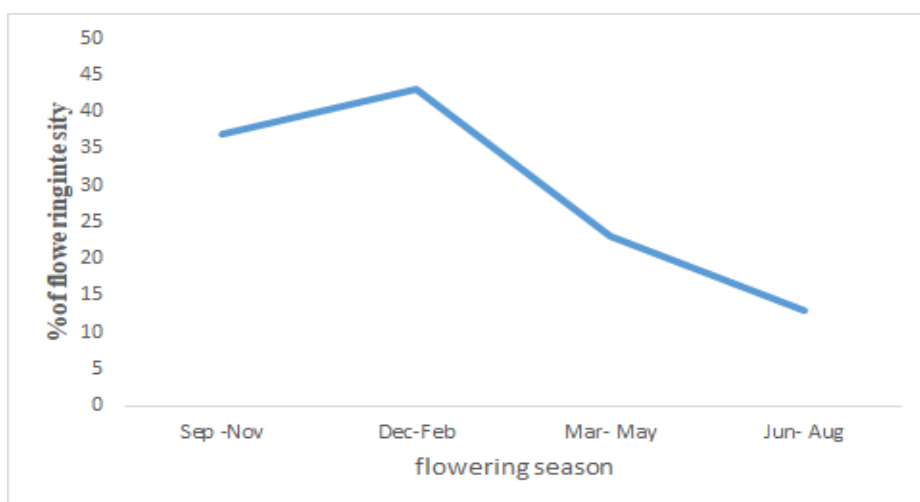


Figure 2. The percentage of flowering period of bee forages in the area

### ***Pollen load analysis***

From the analysis of pollen load collection, 34 plant species were identified ( Table2) and the most frequently visited by honeybees for pollen were *Guizotia scabra*, *Hagenia abyssinica*, *Rumex nepalensis*, *Aspilia africana*, *Vernonia amygdalina*, and *Bidens pilosa* 18.52%, 5.56%, 4.32%, 6.17%, and 11.73% respectively. The rest of the species are rare and indirectly contributing for brood rearing. The contribution of each species as major source of pollen is due to abundance and pollen yielding potential of the plants. This result is also supported with findings of Admassu and Tura 2021 Abebe *et al* 2013 and Admassu 2003, who reported that honeybees forage on more productive and profitable plants that provide necessary nutrition nearby their hives.

Table 2- Plant species identified from pollen load analysis

<b><i>Botanical name</i></b>	<b>Pollen weight (gm)</b>	<b>Foraging length</b>	<b>Agro-ecology</b>
<i>Guizotia scabra</i>	18.52	Nov.-Feb.	All agro-ecology
<i>Hagenia abyssinica</i>	5.56	Oct.-Jan.	Highland
<i>Kniphofia pilosa</i>	1.85	Nov.-Dec.	Highland
<i>Rumex nepalensis</i>	4.32	Nov.-January	Highland
<i>Hypoestes Spp</i>	0.62	January	Highland
<i>Eucalyptus Camaldulensis</i>	1.85	Nov.-January	Highland
<i>Aspilia Africana</i>	6.17	Oct. –Feb.	All Agro-ecology
<i>Apodytes dimidata</i>	0.62	January	Highland
<i>Crassiocephalum Vitellinum</i>	0.62	Dec. –Jan.	Highland
<i>Discopodium penninervium</i>	0.62	Jan. –Feb.	Highland
<i>Vernonia amygdalina</i>	5.56	Dec.-Feb.	All agro-ecology
<i>Brassica carinata</i>	0.62	November	Highland
<i>Trifolium spp.</i>	1.23	November	Highland
<i>Eucalyptus globulus</i>	1.23	Nov.-Dec	Highland
<i>Plantago Lanceolata</i>	2.47	Nov.-Dec	Highland
<i>Cyperus longus</i>	0.62	December	Highland
<i>Terminalia Spp.</i>	0.62	December	Midland
<i>Vernonia adoensis</i>	1.85	Dec. –Feb	Midland

<b>Botanical name</b>	<b>Pollen weight (gm)</b>	<b>Foraging length</b>	<b>Agro-ecology</b>
<i>Coffea Arabica</i>	1.23	Jan.-Feb.	Midland
<i>Bidens pilosa</i>	6.79	Sept.-January	Midland
<i>Maesa lanceolata</i>	2.47	Dec.-Jan	Midland
<i>Ocimum basilium</i>	1.23	January	Midland
<i>Phyllanthus reticulatus</i>	0.62	December	Lowland
<i>Melilotus alba</i>	0.62	January	Lowland
<i>Justicia spp</i>	0.62	December	Lowland
<i>Crambe hispanica</i>	3.70	Nov.-Dec.	Highland
<i>Carduus nyssanus</i>	0.62	Nov.-Jan.	Highland
<i>Calpurnia subdecandra</i>	0.62	December	Lowland
<i>Gallineria saxifraga</i>	0.62	December	Lowland
<i>Vernonia leopoldii</i>	3.09	Nov.-Dec.	Midland
<i>Rumex spp</i>	3.09	Nov.-Jan.	Highland
<i>Guizotia abyssinica</i>	0.62	November	Highland

### **Honey pollen analysis**

The honey pollen analysis indicated that 28 plant species were identified from the study districts Table 3. The predominant honey source plants representing > 45% pollen count were recorded for *Terminalia brownii*, *Guizotia scabra*, and *Croton macrostachyus* for Liban district. On other hand *Eucalyptus spp* , *Croton macrostachyus* and *Coffea arabica* are predominant honey source plants from Adola and Bore districts while *Schefflera abyssinica* and *Vernonia spp* are predominant honey source plants in Ana sora district Table 3. The secondary pollen source plants representing (16-45%) were *Achyranthes aspera*, *Vernonia spp* and *Terminalia laxiflora*. The minor important pollen source plants ((3-15%) were *Bidens pilosa*, *Crambe hispanica*, *Hypoestes spp* and, *Ocimum basilicum*, *Achyranthes aspera*. The plant species representing below 3% include *Cyperus longus*, *Maesa lanceolata*, *Justicia spp*, *Hypoestes forkaoli* and *Acacia Senegal*. The contribution of the plant species as source monofloral honey is due to their abundance, nectar yielding potential of the plants and duration of flowering. In this study seven types of monofloral honey were identified which include *Coffea* honey, *Guizotia* honey, *Vernonia spp*, honey, *Croton* honey, *Eucalyptus* honey, *Schefflera* honey and *Terminalia* honey Table 3. The presence of the above monofloral honey types have been reported in different parts of the country as indicated by Admassu and Tura ( 2021), Abera *et al* (2015), Nuru *et al* ( 2017), and Mesert Gemed (2018). Among the identified monofloral honey types *Guizotia scabra* covers large portion of cultivated areas of Liban district. This is agreeing with Tura and Admassu, 2019 who reported that *Guizotia scabra* was a predominant monofloral honey source plant in east and west shoa and Borena zones of Oromia. The Eucalyptus honey is also found in Adola, Bore and Gorodola districts due to the wider adaption of Eucalyptus tree to wide range of altitude. The availability of Eucalyptus Monofloral honey has been reported by Abera 2015 in Addis Ababa.

Table 3 Predominate and secondary pollen source plants in different districts of Guji zone of Oromia

District	Predominant pollen source (>45%)	Secondary pollen source (16-45%)	Important minor pollen source (3-15%)	Minor pollen source (< 3%)
Liban	<i>Terminalia brownii</i>		<i>Bidens pilosa</i>	<i>Justicia spp</i> <i>Hypoestes forkaoli</i> <i>Acacia Senegal</i> <i>Guizotia scabra</i> <i>Salvia merjamie</i>
	<i>Guizotia scabra</i> <i>Croton macrostachyus</i>		<i>Bidens pilosa</i> <i>Vernonia spp</i> <i>Crambe hispanica</i> <i>Chirra</i> <i>Hypoestes spp</i> <i>Ocimum basilicum</i> <i>Euphorbia hypericifolia</i> <i>Achyranthes aspera</i>	<i>Plectranthus schmperi</i> <i>Justicia spp</i> <i>Vicia faba</i> <i>Phaseolus vulgaris</i>
Gorodola	<i>Eucalyptus globulus</i>			<i>Achyranthes aspera</i> <i>Cyperus longus</i> <i>Maesa lanceolata</i>
Adola	<i>Eucalyptus globulus</i> <i>Coffea arabica</i> <i>Croton macrostachyus</i>	<i>Achyranthes aspera</i> <i>Vernonia spp</i>	<i>Guizotia scabra</i> <i>Hypoestes forskaoli</i> <i>Achyranthes aspera</i>	<i>Syzygium guineense</i>
Adola	<i>Eucalyptus globulus</i> <i>Coffea arabica</i>	<i>Achyranthes aspera</i> <i>Vernonia spp</i>	<i>Guizotia scabra</i> <i>Hypoestes forskaoli</i> <i>Achyranthes aspera</i>	<i>Syzygium guineense</i>
A/Sorra	<i>Vernonia spp</i> <i>Schefflera abyssinica</i>	<i>Terminalia laxiflora</i>	<i>Pterolobium stellatum</i>	

### Conclusion and Recommendation

The result shown that the presence of more than 80 bee forage species and three honey flow period, of which two major honey flow season was identified in lowlands and one minor honey flow season was identified in highland and mid lands agro-ecologies of Guji zone. As reported from beekeepers interview and field observation, the bee forage diversity and honey production potential of the zone is declining due to expansion of farmland and use of pesticide and herbicide. Therefore, in order to increase honey production and maintain the colony population in the area, honeybee forage should be conserved through better management of forest and also avoiding unwise application of pesticide and herbicides in the area.

### Acknowledgement

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Appendix 1:- Checklist of bee forages obtained from field inventory in the study area

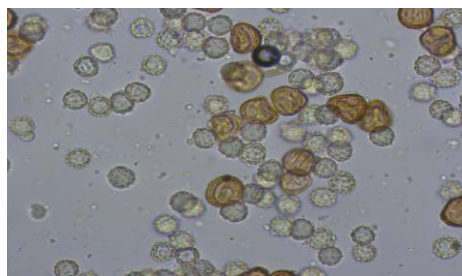
Botanical name	Family	Local Name	Habit	Flowering period	No of Individuals	Diversity indices		
						ni	Ln ni	(ni/lnni)
<i>Schefflera abyssinica</i>	<i>Araliaceae</i>	Gatame	Tree	Feb.-Apr	25	0.008	-7.13	0.0057
<i>Vernonia amygdalina</i>	<i>Asteraceae</i>	Ebicha	Shrub	Dec.-Jan.	184	0.006	-5.08	0.032
<i>Syzygium guineense</i>	<i>Myrtaceae</i>	Badessa	Tree	Mar-Apr.	19	0.002	-7.35	0.0047
<i>Eucalyptus camaldulensis</i>	<i>Myrtaceae</i>	Bargamo Dima	Tree	Mar-May	1788	0.006	-2.81	0.17
<i>Vernonia auriculifera</i>	<i>Asteraceae</i>	Reji	Shrub	Dec.-Feb.	1931	0.007	-2.73	0.18
<i>Cordia Africana</i>	<i>Boraginaceae</i>	Wadessa	Tree	March	22	0.001	-7.21	0.005
<i>Acacia brevispica</i>	<i>Fabaceae</i>	Hamarresa	Shrub	Dec.-Feb.	6	0.001	-8.72	0.002
<i>Acacia nilotica</i>	<i>Fabaceae</i>	Burra	Tree	Aprl-Mar	16	0.05	-7.52	0.0041
<i>Croton macrostachyus</i>	<i>Euphorbiaceae</i>	Mokonnisa	Tree	April-June	218	0.007	-4.92	0.036
<i>Ilex mitis</i>	<i>Aquifoliaceae</i>	Hangadhi	Tree	Dec.	4	0.001	-9.21	0.0009
<i>Dombeya torrida</i>	<i>Sterculiaceae</i>	Danisa	Tree	Dec-Feb	16	0.05	-7.52	0.0041
<i>Euphorbia abyssinica</i>	<i>Euphorbiaceae</i>	Adami	Tree	Oct.-May	839	0.02	3.56	0.10
<i>Hypoestes forskoolii</i>	<i>Acanthaceae</i>	Qaxine	Herb	Jun	2183	0.07	-2.62	0.19
<i>Senecio mriocephalum</i>	<i>Asteraceae</i>	Gimbodha	Shrub	Oct.-Jan.	37	0.01	-6.73	0.008
<i>Allophyllus abyssinicus</i>	<i>Sapindaceae</i>	Saraji	Tree	Aug-sept.	6	0.002	-8.72	0.002
<i>Ekebergia capensis</i>	<i>Meliaceae</i>	Anonu	Tree	Jan.-Mar	18	0.007	-7.42	0.0045
<i>Phytolacca dodecandra</i>	<i>Phytolaccaceae</i>	Andode	Climber	Nov.-Mar	5	0.0002	8.52	0.0017
<i>Maytenus obscura</i>	<i>Celastraceae</i>	Kom'olcha	Tree	Dec.	65	0.0022	-6.12	0.0135
<i>Brucea antidysenterica</i>	<i>Simaroubaceae</i>	komonyo	Shrub	Nov.-Dec.	44	0.0015	-6.50	0.0096
<i>Pterolobium stellatum</i>	<i>Fabaceae</i>	Gora	Shrub	Dec	472	0.0158	-4.15	0.066
<i>Coffea arabica</i>	<i>Rubiaceae</i>	Buna	Shrub	Mar	505	0.017	-4.08	0.069
<i>Acacia abyssinica</i>	<i>Fabaceae</i>	Lafto/Ambo	Tree	Mar	13	0.00044	-7.73	0.0034
<i>Hagenia abyssinica</i>	<i>Rosaceae</i>	Heto	Tree	Oct-Jan	29	0.001	-6.91	0.01
<i>Acanthus eminens</i>	<i>Acanthaceae</i>	Gogodhu	Shrub	Dec.-Apr	222	0.0074	-4.91	0.036
<i>Trifolium spp</i>	<i>Fabaceae</i>	Sidisa/Mirtu	Herb	Sep	517	0.17	-1.77	0.3012
<i>Satureja paradoxa</i>	<i>Lamiaceae</i>	Aredo	Herb	July	804	0.027	-3.61	0.098
<i>Carduus spp</i>	<i>Asteraceae</i>	Qore Hare	Herb	Sept	15	0.001	-6.91	0.007
<i>Senna septemtrionalis</i>	<i>Fabaceae</i>	Sisiko	Shrub	Sept.-Oct	37	0.0012	-6.73	0.01
<i>Pavonia urens</i>	<i>Malvaceae</i>	Hinchini	Shrub	De.-feb.	107	0.0361	-3.32	0.12
<i>Bothriocline Schimperii</i>	<i>Asteraceae</i>	Soyoma	Herb	Oct.-Apr	150	0.01	-4.61	0.05
<i>Urera hypselodendron</i>	<i>Urticaceae</i>	Hajija	Climber	Oct.-Dec.	11	0.0004	-7.82	0.0031
<i>Clematis hirsute</i>	<i>Ranunculaceae</i>	Fiti	Climber	Sep.-Jan.	0	0	0	0
<i>Ziziphus mauritiana</i>	<i>Rhamnaceae</i>	Hagala	Tree	Jan.-June	25	0.001	-6.91	0.01
<i>Terminalia laxiflora</i>	<i>Combretaceae</i>	Rukessa	Tree	Dec	3	0.0001	-9.21	0.001
<i>Erythrina brucei</i>	<i>Fabaceae</i>	Walena	Tree	Nov.-Jan	108	0.004	-5.52	0.0221
<i>Ocimum sauve</i>	<i>Lamiaceae</i>	Hanchabi	Shrub	Oct-Jan	103	0.035	-3.35	0.12
<i>Datura stramonium L.</i>	<i>Solanaceae</i>	Atafaris/Bubu	Herb	July-Aug	54	0.002	-6.21	0.012
<i>Grewia ferruginea</i>	<i>Tiliaceae</i>	Dokonu	Shrub	Oct.-Dec.	0	0	0	0
<i>Carica papaya L.</i>	<i>Caricaceae</i>	Papaya	Tree	Year round	1	0.00003	-10.41	0.0003
<i>Justicia schimperiana</i>	<i>Acanthaceae</i>	dumuga	Shrub	Nov	490	0.016	-4.31	0.066
<i>Salvia tiliifolia</i>	<i>Lamiaceae</i>	Basha	Herb	July	1471	0.05	-3.00	0.15

<i>Maesa lanceolata</i>	Myrsinaceae	Abayi	Shrub	Nov-Dec	28	0.00094	-6.97	0.007
<i>Milletia ferruginea</i>	Fabaceae	Sariti	Shrub	Dec.	131	0.0044	-5.43	0.024
<i>Combretum molle</i>	Combretaceae	Bika	Tree	March	9	0.0003	8.11	0.0024
<i>Acacia Senegal</i>	Fabaceae	Saphansa	Shrub	Mar-Apr	12	0.0004	-7.82	0.0031
<i>Grewia bicolor</i>	Tiliaceae	Haroressa	Shrub	Dec.	8	0.0003	-8.11	0.0024
<i>Acacia tortolisi</i>	Fabaceae	Dhadacha	Tree	March	0	0	0	0
<i>Acacia lahai</i>	Fabaceae	Garbi	Tree	April	5	0.0002	-8.52	0.0017
<i>Prunus Africana</i>	Rosaceae	Sukke	Tree	Nov-Feb	1	0.00003	-10.4	0.0003
<i>Eucalyptus globulus</i>	Myrtaceae	Bargamo Adi	Tree	Mar-May	69	0.0023	-6.07	0.014
<i>PPhoenix reclinata</i>	Arecaceae	Mexi	Tree	Sept	4	0.0001	-9.21	0.0009
<i>Plectranthus garckeianus</i>	Lamiaceae	Ajo'a	Herb	Nov-Dec	28	0.00094	-6.97	0.007
<i>Rhamnus prinoides</i>	Rhamnaceae	Gesho	Shrub	Nov.-Jan	4	0.0001	-9.21	0.0009
<i>Agave sisalana</i>	Agavaceae	Alge	Herb	Dec	24	0.0008	-7.13	0.0057
<i>Bidens pilosa</i>	Asteraceae	Bila/Matane	Herb	Sept.-Nov	750	0.025	-3.6	0.092
<i>Carduus Schimperii</i>	Asteraceae	Qore Hare	Herb	Nov.-Jan	98	0.0033	-5.7	0.019
<i>Crassocephalu Macropappum</i>	Asteraceae	Tambo bera	Herb	Jun	156	0.005	-5.3	0.026
<i>Caesalpinia decapetala</i>	Fabaceae	Harangama	Shrub	Oct.-Feb.	52	0.0017	-6.3	0.011
<i>Clausena anisata</i>	Rutaceae	Uluma	Shrub	Oct.-Apr	0	0	0	0
<i>Galineria saxifrage</i>	Fabaceae	Kudhumi	Tree	Nov.-Jan.	7	0.0002	-8.5	0.0017
<i>Vernonia Spp</i>	Asteraceae	Mararo	Shrubs	Dec.-Feb.	219	0.0073	-4.9	0.036
<i>Aspilia Africana</i>	Asteraceae	Hadaa	Shrubs	Oct.-Dec.	355	0.012	-4.4	0.053
					29.8			2.85

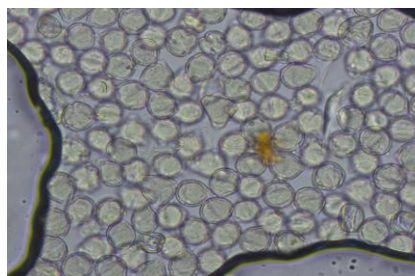
Appendix 2 List of plant species identified from honey pollen analysis in different districts of the zone

No	Identified Plants	Frequency	Percentage	Season (month)	Districts	Kebeles
1	<i>Justicia spp</i>	10	0.70	August	Liban	Miessa
2	<i>Hypoestes forkaoli</i>	30	2.30			
3	<i>Acacia Senegal</i>	20	1.50			
4	<i>Guizotia scabra</i>	5	0.40			
5	<i>Bidens pilosa</i>	50	3.80			
6	<i>Terminalia browni</i>	1200	90.90			
7	<i>Salvia merjamie</i>	5	0.40			
1	<i>Crambe hispanica</i>	50	3.64	December	Liban	Miessa
2	<i>plectranthus schmperi</i>	15	1.10			
3	<i>Guizotia scabra</i>	1000	72.73			
4	<i>Bidens pilosa</i>	100	7.27			
5	<i>Vernonia spp</i>	60	4.36			
6	<i>Justicia spp</i>	40	2.90			
7	<i>Chirra</i>	100	7.27			
8	<i>Vicia faba</i>	10	0.73			
1	<i>Bidens pachyloma</i>	20	1.30	August	Liban	Guradhera
2	<i>Hypoestes spp</i>	50	3.20			
3	<i>Terminalia Browni</i>	1500	94.90			
4	<i>Phaseolus vulgaris</i>	10	0.60			
1	<i>Ociumum basilicum</i>	60	4.60	July 20 - August	Liban	Q/Mallee
2	<i>Hypoestes spp</i>	40	3.10			
3	<i>Euphorbia hypericifolia</i>	50	3.80			
4	<i>Croton macrostachyus</i>	1000	77.00			

5	<i>Achyranthes aspera</i>	50	3.80			
6	<i>Bidens pilosa</i>	100	7.70			
1	<i>Eucalyptus globulus</i>	1400	97.20	September	Gorodola	Harqalo
2	<i>Achyranthes aspera</i>	10	0.70			
3	<i>Cyperus longus</i>	10	0.70			
4	<i>Maesa lanceolata</i>	20	1.40			
1	<i>Eucalyptus globulus</i>	1000	72.70	March	Adola	Q/sorsa
2	<i>vernonia spp</i>	200	14.50			
3	<i>Guizotia scabra</i>	100	7.30			
4	<i>Hypoestes forkaoli</i>	75	5.50			
1	<i>Achyranthes aspera</i>	1000	33.3	March	Adola	04(Sub-site)
2	<i>Eucalyptus globulus</i>	2000	66.70			
1	<i>Eucalyptus globulus</i>	500	32.30	February	Adola	Dole/Anfara
2	<i>coffea arabica</i>	1000	64.50			
3	<i>Achyranthes aspera</i>	50	3.20			
1	<i>Croton macrostachyus</i>	250	70.42	July	Adola	04/Sub-site
2	<i>Syzygium guineense</i>	5	1.96			
3	<i>Euculyptus globulus</i>	100	28.17			
1	<i>Eucalyptus globulus</i>	100	99.00	June	Bore	A/Quxure
2	<i>Terminalia Spp</i>	1	1.00			
1	<i>Vernonia spp</i>	1000	47.62	April	A/Sorra	B/Qorsa
2	<i>Schefflera abyssinica</i>	1000	47.62			
3	<i>Pterolobium stellatum</i>	100	4.76			
1	<i>Vernonia spp</i>	2000	80	February	A/Sorraa	B/Qorsa
2	<i>Terminalia laxiflora</i>	500	20			



*Pollen morphollgy of Bidens pilosa*



*Pollen grain morphology of Coffea arabica monofloral honey*

## Herbaceous bee forage establishment in low lands of Borana Zone Southern Oromia

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### Abstract

*For alleviating the shortage of bee forage and increasing honey production, the use of improved perennial and annual nectar and pollen sources plants are essential for beekeeping development. The study was conducted with the objective of evaluating and selecting adaptable honey bee forage species for sustaining honeybee colonies during the dearth period in mid land and lowland agro-ecologies of the Borana zone. The planting materials were *Aeschynomene uniflorum*, *Melilotus alba*, *Fagopyrum esculentum*, *Sinaps alba* and *Vicia sativa*. The planting materials were planted in a plot size of 2mx2m arranged in randomized block design with three replications. **The plant species were evaluated based on germination date, time to set flower, days to 50% flowering, number of flower heads per plants, flowering length, Days to maturity and Plant height.** Among five evaluated herbaceous bee forage species three of them: *Aeschynomene uniflorum*, *Fagopyrum esculentum* and *Sinaps alba* were well adapted and performed for the most important agronomic parameters. Therefore, the selected bee forage species can be demonstrated under beekeepers apiary in study the areas as well as other areas with similar agro-ecology to be utilized for beekeeping development. Moreover, further evaluation on other agronomic parameters such as determining seeding rate, fertilizer rate, nectar/pollen yield, foraging intensity, and carrying capacity at different location of Borana zone is highly recommended.*

**Keywords:** *Bee Forage, Herbaceous, Flowering Period, Borana zone*

### Introduction

Ethiopia is endowed with wide agro- climatic condition and huge water resources and other favorable ecological factors that have enabled the country to sustain large number of honeybee colonies and also the country is known for long established practices of beekeeping (Fichtl and Admassu, 1994).

Honeybee colony performance as well as production of honey, wax and other hive products are depends on availability of bee forage plants which can provide nectar and pollen for honeybees. These food sources provide the nutritional requirements of the bee colonies: nectar as sources of honey which provides heat and energy for honey bees and pollen provides protein, vitamins, fatty substance, and other nutrients (Mulualem *et al.* 2021). From 250,000 species of flowering plants, 4000 plants species are documented to be important for honey bees as source of nectar and pollen and also reported to be the source of world honey (Crane, 1990).

The diversified agro-climatic conditions of the Ethiopia create environmental conditions conducive for the growth of 6000 to 7000 flowering plant species (Fichtl and Admassu, 1994) and most of them are bee plants. However, currently shortage of bee forage is reported as serious constraint in different parts of the country including Borana zone (Tamirat T. *et al.*, 2013). This is due to rapid human population growth, expansion of agriculture due to forest encroachment, agro-chemicals and drought. Generally, the effort of conserving and managing bee floral resources for maintaining honeybee colonies and the sustaining honey production is poor in the country including the zone.

In order to survive, become productive, honeybee colonies must have a continuous supply of nectar and pollen sources from perennial and annual bee forages in adequate quantities around the apiary. Moreover, planting of improved bee forage flowers will ensure the availability of bee forage and also solve the problem of the bridge of floral gaps and minimize the absconding of honeybee colonies during the dearth period (Malede *et al.*, 2015). Thus introduction of improved bee forages is crucial for beekeeping development in the Borana zone since there is high demand for improved honey bee forage from different stake holders like government, non-government organization, and beekeeper farmers. Therefore, in this study attempts were made to screen and select the best performing herbaceous bee forage species for colony maintenance and honey production in Borana zone of Oromia.

## **Materials and Methods**

### ***Description of the study area***

The study was conducted in Borana zone in southern Oromia. Borana zone is characterized by its arid to semi-arid climates with an annual mean annual temperature ranging from 19°C to 24°C (Coppock, 1994; Kamara *et al.*, 2005). The rainfall pattern is bimodal, with 60% occurring from March to May (long rainy season) and locally named as “*Ganna*” for the area. The short rainy season (*Hagaya*) which extends from September to November (Coppock, 1994). Generally, the average annual rainfall of the area ranges between 350 and 900 mm. On the other hand, the long dry season (*Bona*) occurs from December to February, and the short dry season (*Adolessa*) occurred from June to August. The predominant soil types of the area include red soil, black soil, white or gray soil and sandy soil.

### ***Seed collection***

Seeds of honeybee forage species including *Fagopyrum esculentum*, *Melilotus alba*, *Sinaps alba*, *Vicia sativa* and *Aeschynomene uniflorum* were collected from Adami Tullu and Bako Agricultural Research Centers of Oromia Agricultural Research Institute. The plant species were selected on the basis of wide range of agro-ecological adaptation, the degree of importance as bee forages, similarity in growth habit and ease of propagation from seeds.

### ***Experimental design and management***

To evaluate the performance of the selected plant materials, seeds were planted in rows on prepared seed bed by covering with a thin layer of the soil in plot size of 2m x 2m and arranged in Randomized complete block design with three replications. During planting 20cm between rows and 30cm between plants were used and sowing was carried out during the main rainy season.

The agronomic practice common to the forage plants such as weeding and protection from the pest were applied. The data on days to germinate, density of plant per plot, time to set flower and days to 50% flowering. At 50% flowering, number of flower heads was counted for each species by taking 1m<sup>2</sup> plot area as well as foraging intensity of honeybees on flowers was counted starting from 6: 00 a.m. to 6: 00 p.m. for ten minutes at every 2-hour interval. Five plants from middle of two rows per plots were randomly taken at 50 % flowering and height of plant was measured and average height of plant was recorded for each species. Finally seed yield and drought tolerance ability of the plant were also recorded.

## **Data analysis**

The collected data was analyzed using SAS software version 9.0 and descriptive statistics, One Way Anova. The mean separation was analyzed using LSD test.

## **Results and Discussions**

In this study the mean value of investigated agronomic parameters for *Aeschynomene uniflorum*, *Fagopyrum esculentum* and *Sinaps alba* were indicated in Table 1 and 2. While the other two forages did not reach to germination stage in both years of study periods.

### **Date to emergency (DTE)**

The result shows that the mean emergency date of selected forages were 5.5, 5.6 and 5.2 days for *Aeschynomene uniflorum*, *Fagopyrum esculentum* and *Sinaps alba* respectively. The result of *F. esculentum* is similar with the finding of Tura Bareke *et al* (2014), Tusa Gemechu and Amaslu Arega., and Ofijan Tesfaye, 2017 but higher than Habte Arega *et al.* (2019), *Sinaps alba* has taken 5.5 days which also within the ranges of previous findings Ofijan Tesfaye, 2017, Habte Arega *et al.* (2019). The result shows only *Fagopyrum esculentum* has significance difference ( $P<0.05$ ) in days to germination across planting years (Table 2).

### **Days to flowering (DTF)**

The current finding in days to flowering of selected forages was 66 days, for *Aeschynomene uniflorum*, 27days for *Fagopyrum esculentum* and 45 for *sinaps alba*. The result is within the range of previous reports of different authors. Accordingly, *F. esculentum* has taken 40.3 days to flowering which similar with finding of Tura Bareke *et al.*, 2014, Admasu Adi *et al.*, 2017; Habte Arega *et al.*, 2019; Tusa Gemechu and Amsalu Arega, 2017 and Ofijan Tesfaye 2015 days to flowering of 38 ,25.94 , 26.95 and 29.50days respectively. *Sinaps alba* also has taken 44.7, 35.83, and 58.25 days according to Tura Bareke *et al.*, 2014; Ofijan Tesfaye, 2015; Habte Arega *et al.*, 2019 respectively. Both *Aeschynomene uniflorum* and *Fagopyrum esculentum* showed significance difference ( $P<0.05$ ) in number of days to flower across planting years.

### **Duration to flowering (DF)**

The result shows that the mean duration of flowering for the selected forages were 29, 27 and 27 days for *Aschynomene uniflorum*, *Fagopyrum esculentum* and *Sinaps alba* respectively. This finding agrees with Habte Arega *et al.*, 2019, Tusa Gemechu and Amsalu Arega, 2017; 33.16 and 31 days respectively. Girma Chalchisa *et al.*, 2014 reported that *Aeschynomene uniflorum* has long blooming duration, of 43 days. Bee forage plants which have long flowering period from blooming to shedding are very important for honey production.

### **Days to maturity (DM)**

The current finding on days to maturity of selected forages was 103.6, 66 and 80 days for *A. uniflorum*, *F. esculentum* and *S. alba* respectively. The result indicated that each forages *has significance difference* ( $P<0.05$ ) in days to maturity across planting years (Table 2).

### Plant height (PH)

Plant height is a good indicator of growth rate and adaptation of forages to the environment. The mean performance of plant height of tested bee forage species were 79.75cm, 59cm and 58.3cm for *Aeschynomene uniflorum*, *Fagopyrum esculentum* and *Sinaps alba* respectively. The result shows only *sinaps alba* has significant difference ( $P < 0.05$ ) in plant height across planting years.

Table 1. The mean Agronomic Parameters of bee forage species.

Forage species	Parameters	Year1( 2019/20)			Year 2 (2020/21)			Pooled
		Mean $\pm$ SD	Min	Max	Mean $\pm$ SD	Min	Max	
<i>Aeschynomene uniflorum</i>	DTE	5.00 $\pm$ 0	5	5	6.00 $\pm$ 0	6	6	5.5
	DTF	66.00 $\pm$ 0	66	66	64.00 $\pm$ 0	64	64	65
	NFHP	9.53 $\pm$ 1.5	8	11	15.00 $\pm$ 7	10	23	12.3
	DF	29.67 $\pm$ 1.15	29	31	29.00 $\pm$ 0	29	29	29
	DM	103.67 $\pm$ 0.58	103	104	98.00 $\pm$ 0	98	98	100.8
	PH	81.63 $\pm$ 12.22	68.3	92.3	77.88 $\pm$ 6.57	70.33	82.3	79.75
<i>(Fagopyrum esculentum)</i>	DTE	5.33 $\pm$ 0.58	5	6	6.00 $\pm$ 0	6	6	5.6
	DTF	26.00 $\pm$ 0	26	26	29.00 $\pm$ 0	29	29	27
	NFHP	16.20 $\pm$ 3.12	12.6	18.2	12.57 $\pm$ 3.01	9.2	15	14.4
	DF	30.00 $\pm$ 0	30	30	24.00 $\pm$ 0	24	24	27
	DM	66.00 $\pm$ 0	66	66	64.00 $\pm$ 0	64	64	65
	PH	62.53 $\pm$ 8.44	56.6	72.2	55.53 $\pm$ 2.84	53	58.6	59
<i>Sinaps alba</i>	DTE	5.33 $\pm$ 0.58	5	6	5.00 $\pm$ 0	5	5	5.2
	DTF	44.33 $\pm$ 3.06	41	47	46.00 $\pm$ 0	46	46	45
	NFHP	6.1 $\pm$ 2.85	3.3	9	3.83 $\pm$ 0.29	3.5	4	4.7
	DF	33.67 $\pm$ 4.73	30	39	21.00 $\pm$ 0	21	21	27
	DM	80.00 $\pm$ 0	80	80	75.00 $\pm$ 0	75	75	77.5
	PH	75.53 $\pm$ 5.61	72	82	41.00 $\pm$ 1	40	42	58.3

DTE= Date to emergency DTF= Days to flowering NFHP= Number of flower head per plant DF= Duration of flowering PH= plant height at flowering stage DM= Maturity date

Table 2. Performance of three different bee forage species across planting years

Planting year	Parameters					
	DTE	DTF	NFHP	DF	DM	PH (cm)
	<i>Aeschynomene uniflorum</i>					
2019/20	5.00b	66.00a	9.53a	29.67a	103.67a	81.63a
2020/21	6.00a	64.00b	15.00a	29.00a	98.00b	77.88a
	<i>Fagopyrum esculentum</i>					
2019/20	5.33a	26.00b	16.20a	30.00a	66.00a	62.53a
2020/21	6.00a	29.00a	12.57a	24.00b	64.00b	55.53a
	<i>Sinaps alba</i>					
2019/20	5.33a	44.33a	6.10a	33.67a	80.00a	75.53a
2020/21	5.00a	46.00a	3.83a	21.00b	75.00b	41.00b

\*Means under similar Forage Species and columns with the same superscript letter are not significantly different  $p > 0.05$ , DTE= Date to emergency DTF= Days to flowering NFHP= Number of flower head per plant DF= Duration of flowering PH= plant height at flowering stage DM= Maturity date



## Conclusions and Recommendations

In conclusion the study reveals that among five evaluated herbaceous bee forages species three of them: *Fagopyrum esculentum*, *Aeschynomene uniflorum* and *Sinaps alba* were adapted and well performed based on important agronomic parameters. The performances of each forages species were different across planting years. Further evaluation on other agronomic parameters such as determining seeding rate, fertilizer rate, nectar/pollen yield, foraging intensity honeybees at different agro pastoral areas of the Borana zone is highly recommended.

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# Establishing Floral calendar in Southwest shoa, West shoa and Jimma zones of Oromia Ethiopia

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## Abstract

*Establishment of flowering calendar of honey plants is critical stages in improving honey yields of the area. The study was conducted to identify the major bee forages and preparing floral calendars in west shoa, south west and Jimma zones of Oromia. Bee forage inventory was made using transect methods in a plot size of 20mx20m, for woody plants and 2mx2m for herbs. Pollen traps having 16% pollen trapping efficiency was fitted at the entrance of beehives for pollen load collection. Honey pollen analysis procedure was followed to determine the botanical origin of honey. The study revealed that 101 bee forages were identified of which Asteraceae, Fabaceae, Acanthaceae, Lamiaceae and Myrtaceae are the major plant families comprising 30.8%, 23.1%, 11.5%, 9.6% and 7.7%, species composition and the rest of the families are represented below one percent. From pollen load collection, 90 plant species were identified of which *Bidens prestinaria*; *Eucalyptus camaludensis*, *Eucalyptus globulus*, *Guizotia scabra*, *Plantago lanceolata*, *Maesa lanceolata*, *Trifolium spp*, and *Vernonia spp* are dominant pollen source plants and foraged by honey bees for more than 50 days. The fresh weight of pollen collected at different months of the year indicated that significant amount of pollen was collected during September, October to November but decline of the fresh weigh of t pollen was recorded during, February and March in most parts of the study area. From analysis of fresh weight of pollen on the basis of seasonal variation indicated that two major honey flow period were identified. The pollen analysis of honey samples also revealed that five monofloral honey types were identified which include *Guizotia spp*, *Erica arborea*, *Eucalyptus spp*, *Coffea arabica* and *Croton macrostachyus* comprising the pollen frequency ranging from 45% to 100%. The study concluded that floral calendar of bee forages strictly dependent on the seasonal availability of pollen and beekeepers should get awareness on management of honeybee colonies with the flowering calendar in their respective district to increase frequency of honey production.*

**Key words:** Flowering calendar, pollen analysis, pollen weight, plant diversity, honey flow period

## Introduction

Ethiopia is endowed with natural and cultivated flora and diverse agro-ecological and climatic condition that are well-suited for beekeeping. Oromia is one of the regional states in Federal Republic of Ethiopia rich in natural resources and favorable climatic condition for beekeeping development. The vegetation covers of the Oromia accounts for about 69% of the total area of the Region. Out of the 58 National Forest found in Ethiopia, about 49 are found in Oromia region (Hailu, 2020). The region is also known for growth of diverse cultivated crops such as oil crops, pulses, cereals and horticultural crops. This makes the region is one of the major honey producing area with production volume of 15,492 tons( 41%) of the country's honey production (Tura and Admassu 2019). For proper utilization of the immense beekeeping potential of the region, the identification, documentation of economic bee forages and preparing their flowering calendar are important to increase honey production that contributes to wards boosting the income of the household. The floral calendar of bee plant is a timetable that indicates the

date and duration of flowering of important bee plants. It is an important tool that informs the availability of certain bee forage for particular area, to predict time of honey flow period and their values to bees (Haftom, 2016).

Central and western Oromia including southwest, West shewa and Jimma zones are covered with forestlands including Chilimo, Gedo and Jibat forests, which are potential for beekeeping (Kassa et al. 2009, Teshome and Ensermu 2013). In addition, most areas of west shoa and southwest shoa zones are covered with vertisols which is suitable for growth of pulses and oil crops providing pollen and nectar for honey bees (Nuru and Admassu, 2001). On other hand Tolera and Dejene, 2014 reported that western Oromia including Jimma Zone has considerable beekeeping potential with rich flora and high honeybee population.

Even though apiculture resource is immense in zones, due to lack of information on flowering calendar corresponding to seasonal colony management practices, beekeepers are losing significant amount of honey. Therefore, identification of major bee forages and preparing floral calendars which can correspond with seasonal colony management practice is important for determining appropriate honey flow period for multiple honey production. Thus study was designed to identify major bee forages and prepare flowering calendar and to assess the mono floral honey types in the area.

## **Materials and Methods**

### ***Description of the study area***

The study was conducted in central highlands and western Oromia of Ethiopia with particular zones of Southwest Shewa, West Shewa, and Jimma zones. The study districts covered were Woliso, Amaya, Wonchi, Ejere, Gedo, Bako, Tolay, Omonada and Gomma. These districts have different agro-ecologies ranging from highlands, midland and low lands and covered with different natural vegetation and variety of cultivated crops including oil crops, cereals, pulses and horticultural crops suitable for beekeeping.

### ***Inventory of bee forages***

To assess the bee forages abundance and diversity, four transect lines were laid out from apiary sites to North, South, West and East within 2 Km radius following GPS. Apiary sites were selected systematically within 2 km distance from one to the other in order to avoid redundancy. Along these transects, a plot of 20mx20 m were laid out within 400 m interval between the sample plots. In order to retain accuracy, five (5) sub plots measuring 2mx2 m (4m<sup>2</sup>) were laid out within the larger plot to capture herbaceous and grasses. All the plant species encountered in each sample plots were recorded and percentage cover of each species was estimated visually. For those plant species which could not identified in the field, sample of the specimens were collected using the standard Herbarium techniques and identified at Holeta Bee Research center and National Herbarium of Addis Ababa University.

### ***Colony establishment and pollen load collection***

Twenty-seven (27) honeybees' colonies were established at 12 sites of the nine districts of southwest, west shoa and Jimma zone of Oromia. Pollen trap was fitted at the entrance of bee hives having the pollen trapping efficiency of 16% and pollen loads was collected four times per week. The pollen samples were placed in the clean paper bags and left for 24hr to dry at room temperature. The pollen grains were

collected and sorted into colors and identified to the genus species level using the pollen atlas of Ethiopia (Nuru, 2002).

### *Pollen analysis of honey*

For pollen analysis of honey, a total of 27 honey samples were collected from farm gates of the beekeepers during the honey harvesting seasons. The samples of honey about 500 gm. were collected from each district for analysis. All samples were kept in sealed glass jars and frozen at -20°C until analysis. The pollen analysis was made following the methods adopted by Louvuex *et al.* (1978) for determination of botanical composition and frequency of pollen grains in the honey.

### *Data analysis*

Descriptive statistics was used to present abundance and frequency of the species in different districts of the study area and variation of pollen collection among the study districts. The abundance of bee flora was calculated as the number of individuals plant in sample plots divided by total number of the species.

$$Abundance = \frac{\text{number of individuals}}{\text{Total number of species}}$$

### **Results**

From inventory of bee forage, pollen load collection and pollen analysis of honey, 101 plant species were identified belonging to 32 families Appendix I. Among the plant families Asteraceae, Fabaceae, Acanthaceae, Lamiaceae, Roseaceae, Myrtaceae and Malvaceae are the major plant families comprising 30.8%, 23.1%, 11.5%, 9.6%, 5.6%, 7.7%, and 7.7% species respectively. The rest of the families are represented by one or two species. The growth forms of bee forage utilized by honeybees comprises herb (41.6%), shrubs (28.7%), trees (21.7%) and climbers (8%) Fig.1. The herbaceous flora is dominant bee forages comprising weeds (*Guizotia scabra*, *Hypoestes forskalii*, *Hypoestes triflora* and *Trifolium* spp) and some cultivated crops (*Brassica*, spp, *Vicia faba*, pulses, cereals and oil crops) greatly contribute to the honey production in the area.

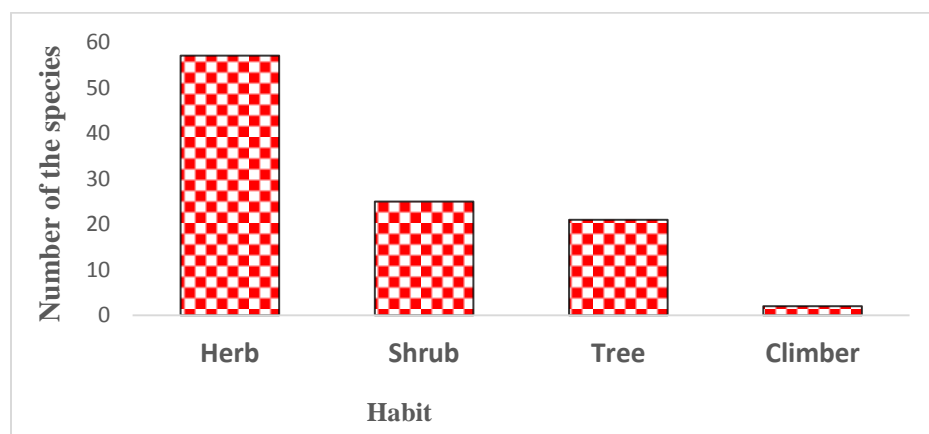


Figure 1: The growth habit of bee forages in the study area

### ***Abundance of bee forages***

From field inventory of bee forages 89 plant species was identified from inventory of bee forages and out of these 36 species are found to be the most frequent and abundant species in the area Table 1. These are *Eucalyptus* spp, *Croton macrostachyus*, *Justicia schimperana*, *Cordia africana*, *Vernonia* spp, *Guizotia scabra*, *Ageratum conyzoides*, *Achyrrathes aspera* , *Hypoestes* spp and *Coffea arabica*.

Table 1. The abundance of major bee forages of the study districts.

Plant Species	Wolisso	Amaya	Wonchi	Ejere	Gedo	Bako	Tolay	Gomma	Omonada
<i>Croton macrostachyus</i>	5.5	23	3.2	9.4	17.5	12.1	5	5	57
<i>Eucalyptus camaludensis</i>	12	21.5	0	14	1.4	38.3	28	25	3.6
<i>Eucalyptus globulus</i>	0	0	5	26.1	0.94	0	0	0	
<i>Justicia schimperiana</i>	26.1	0	0	35		27.3	3.5	0	16
<i>Solanum spp</i>	11	8.5	0	0	0	23.	0	0	0
<i>Erica arborea</i>	0		36	0	0	0	0	0	0
<i>Ocimum urticifolium</i>	6	0	0	2.9	0	12		0	2.7
<i>Cordia Africana</i>	0	18	3.6	0	0	8.9	2.2	0	4.2
<i>Maytenus spp</i>	31.3	0.7		7.5	4	4.76	0	0	1.4
<i>Vernonia amygdalina</i>	3.2	8.8	6	12.8	6	14	6.3	0	10
<i>Vernonia auriculifera</i>	12	14	9	32	12	17	22	0	10.6
<i>Coffea Arabica</i>	20.92	19	7.7	0	0	24.	56	45	
<i>Acanthus sennii</i>	1.36	28	23.6	0	0	36	0	0	11
<i>Calpurea aurea</i>	2.7	12	3.5	16		4.93	3.6	0	0
<i>Euphorbia turucalli</i>	0	3.7	0	0	0	37	0	0	9.3
<i>Syzygium guineese</i>	0	6.1	0	4.73		4.25	0	0	0
<i>Albizia gummifera</i>	1.29	1.7	0	0	0	1.9	2.8	2.7	0
<i>Bersama Abyssinia</i>	1.08	0	0	0	0	0.49	4.06	0	0
<i>Clausena anisata</i>	5	0	5.8	0	0	3.5	6.8		2.1
<i>Pavonia urens</i>	0.8	0	0	4.5	4.6	3.4	0	33.7	0
<i>Rumex spp</i>	6.25	0	0	2.8	2.8	2.7	0	0	0
<i>Terminalia schimperiana</i>	0	0	0	0	0	2.9	0	0	0
<i>Acacia spp</i>	12.5	4.5	2.2	0	0	2.2	0	0	0
<i>Brucea antidysenterica</i>	12	0	0	0	0	0	0	1.9	0
<i>Echinops macrochaetus</i>	14.2	0	0	0	0	0	0	0	0
<i>Carrisa edulis</i>	8.3	0	0	24.7	10	0	0	0	0
<i>Caesalpinia decapetala</i>	7.4	12.5	6.6	0	0	0	0	0	0
<i>Ekbergia capensis</i>	0.39	0	0	0	0	0	0	0	4.9
<i>Premania schimperi</i>	2.89	0	0	4.4		0	0	0	0
<i>Maesa lanceolata</i>	0	0	0	5.6	2.1	0	7.4	7.8	2.6

Plant Species	Wolisso	Amaya	Wonchi	Ejere	Gedo	Bako	Tolay	Gomma	Omonada
<i>Rubus steudneri</i>	0	0	0	7.7	7	0		5.6	0
<i>Persea Americana</i>	0	0	0	0	0	0	3.5	3.4	46
<i>Ageratum conyzoides</i>	6.7	1	6.8	0		1.2	0.33	1.67	3.7
<i>Bidens pilosa</i>	0	8	14.2	50	1.2	2.01	0	2.7	20
<i>Guizotia scabra</i>	8.8	1.14	1.9	1.16	1.5	1.22	0	3.3	3.6
<i>Hygrophilia auriculata</i>	7.5	1.5	0	13	23	2.1	0	8.8	50

### ***Pollen load analysis of major bee forages in southwest and west shoa zone***

From the analysis of pollen load collection, the most frequently visited for more than 50 days which include *Bidens prestiniana*, *Eucalyptus camludensis*, *Euclyptus globulus*, *Guizotia scabra*, *Plantago lanceolata*, *Maesa lanceolata*, *Trifolium* spp, and *Vernonia* spp. On the other hand plant species moderately visited for pollen by honeybees for 30-45 days include *Echinops macrochaetus*, *Maytenus obscura*, *vicia faba*, *Apodytes dimidiata*, *Caylusea abyssinica*, *Hagenia abyssinica*, *Terminalia schimperiana*, *Zea mays* and the rest of the species are rarely visited by honeybees below 30 days (Table 2).

Table 2. Foraging length of the major bee forages in southwest and west Shoa zones

plant species	Families	starting	ending	Foraging Day
<i>Andropogon abyssinica</i>	Poaceae	Dec	Feb	25
<i>Apodytes dimidiata</i>	Iciniaceae	Jan	Feb	30
<i>Bidens prestiniana</i>	Asteraceae	sept	Nov	70
<i>Brassica carinata</i>	Brassicaceae	sept	Dec	20
<i>Caylusea abyssinica</i>	Resedeaceae	Jan	Mar	35
<i>Cleome usambarica</i>	Cappriadeae	Jan	February	28
<i>Crambe abyssinica</i>	Brassicaceae	Jun	August	20
<i>Cyperus longus</i>	Cyperaceae	Mar	May	20
<i>Datura arborea</i>	Solanaceae	May	Oct	50
<i>Dombeya aethiopica</i>	Stericulaceae	Dec	Jan	20
<i>Echinops macrochaetus</i>	Asteraceae	Dec	Feb	45
<i>Eucalyptus camldensis</i>	Myrtaceae	Mar	Jun	65
<i>Erica arborea</i>	Ericaceae	sept	Dec	20
<i>Eucalyptus globulus</i>	Myrtaceae	May	Jun	70
<i>Guizotia scabra</i>	Asteraceae	Nov	Jan	95
<i>Hagenia abyssinica</i>	Roseaceae	Oct	Feb	35
<i>Hypoestes triflora</i>	Acanthaceae	Dec	Jan	25
<i>justicia heterocarpa</i>	Acanthaceae	Nov	Dec	25
<i>Maytenus obscura</i>	Calestarceae	Mar	Aug	45
<i>Mesea lanceolata</i>	Mrisenaceae	July	Dec	50
<i>Ocimum bucilucim</i>	Lamiaceae	Jun	Aug	20
<i>Pavonia urens</i>	Malvaceae	Dec	Mar	25

plant species	Families	starting	ending	Foraging Day
<i>Plantago lanceolata</i>	Plantaginaceae	Oct	March	115
<i>Prunus Africana</i>	Roseaceae	Jun	July	20
<i>Rumex nervosus</i>	Polygonaceae	July	march	87
<i>scheffleria abyssinica</i>	Araliaceae	Mar	April	30
<i>Schinus molle</i>	Anacardiaceae	sept	Oct	20
<i>Sesamum indicum</i>	Pedilaceae	Nov	Dec	20
<i>Syzygium guineese</i>	Myrtaceae	January	Mar	20
<i>Terminalia schimperiana</i>	Combretaceae	Dec	Jan	30
<i>Trifolium spp</i>	Fabaceae	Sept	Dec	59
<i>Vernonia amygdalina</i>	Asteraceae	Dec	Feb	57
<i>vicia faba</i>	Fabaceae	Aug	sept	45
<i>Zea mays</i>	Poaceae	Jun	Aug	30

### ***Pollen load analysis of major bee forages from Jimma zone***

Among the identified pollen source plants, *Ageratum conizoides*, *Guizotia scabra*, *Cordia africana*, *Datura arborea* *Plantago lanceolatum*, *Rumex nervosus*, and *Justicia schimperina*, were the most frequent pollen source plants and foraged for more than 50 days. The moderately foraged plant species were *Croton macrostachys*, *Carica papaya*, *Gravillea robusta*, *Clematis simensis*, *Rhus vulgaris*, *Rosa abyssinica*, *Vicia faba* for foraged for 25-45 days and the rest of the species were least foraged by honeybees Table 3.

Table 3: Foraging length of the major bee forages in Jimma zone.

<b>Plant species</b>	<b>Families</b>	<b>starting</b>	<b>ending</b>	<b>days</b>
<i>Acacia spp</i>	Fabaceae	Oct	Dec	54
<i>Ageratum conizoides</i>	Asteraceae	Feb	Aug	108
<i>Albizia spp</i>	Fabaceae	Oct	Nov	54
<i>Carex spp</i>	Cyperaceae	May	Jun	54
<i>carica papaya</i>	Caricaceae	Oct	Nov	30
<i>Clematis simensis</i>	Ranuclaceae	Dec	Jan	30
<i>Coffea Arabica</i>	Rubiaceae	Feb	March	27
<i>Cordia Africana</i>	Boraginaceae	Aug	Oct	60
<i>Croton macrostachys</i>	Euphorbiceae	April	May	40
<i>Datura arborea</i>	Solanaceae	Feb	May	54
<i>Echionpoes spp</i>	Asteraceae	Feb	March	54
<i>Eucalyptus camaldunsis</i>	Myrtaceae	May	Jun	35
<i>Grevillea robusta</i>	Proteaceae	March	April	43
<i>Guizotia abyssinca</i>	Asteraceae	Oct	Nov	33
<i>Guizotia scabra</i>	Asteraceae	Oct	Dec	144
<i>Hypoestes spp</i>	Acanthaceae	Oct	Nov	18
<i>Justicia schimperina</i>	Acanthaceae	Nov	Feb	56



Plant species	Families	starting	ending	days
<i>Mangifera indica</i>	Ancardiaceae	July	Aug	58
<i>Persea Americana</i>	Luraceae	May	Jun	60
<i>Phaseolous vulgaris</i>	Fabaceae	Aug	sept	30
<i>Plantago lanceolatum</i>	Plantaginaceae	May	Oct	60
<i>Ranunculas muitifidus</i>	Ranunculaceae	Aug	Oct	60
<i>Rhus vulgaris</i>	Anacardiaceae	sept	Oct	45
<i>Rosa abyssinica</i>	Roseaceae	July	Aug	42
<i>Rumex nervosus</i>	polygonacea	July	Aug	60
<i>Trifolium spp</i>	Fbaceae	Feb	Aug	48
<i>Vernonia amygdalina</i>	Asteraceae	Nov	Jan	35
<i>Vicia faba</i>	Fabaceae	Aug	Oct	42
<i>Zea mays</i>	Poaceae	Mar	May	25

### ***Seasonal availability pollen in the districts***

The fresh weight of pollen collected at different months of the year indicated that significant amount of pollen was collected during September, October to November but decline of fresh weight of pollen was recorded during, February and March in Wolliso, Amaya and Wonchi districts Figure2a. There was slight increase in fresh weight of pollen during the months of April and May which is known as minor honey flow period in the three districts. On the other hand, very low pollen weight was recorded during months of February and, July. Similar pattern of pollen weight collection was also recorded for Ejere, Bako-Tibe except for Challiya district in which the peak fresh weight of pollen was recorded from November to February (Fig.2b).

Regarding the monthly pollen weight collection for Jimma zone (Tolay, Gomma and Omonada), the highest pollen weight was recorded during the months of October, November and December. Relatively better amount of pollen was also collected during January and February in Gomma district due to flowering of *Vernonia amygdalina* and *Coffea arabica*. On top of this higher pollen weight was recorded during April to May due availability of *Eucalyptus spp* and *Croton macrostachyus* flowers in Gomma and Omonada as indicated in Figure 2c. The scarcity of pollen or very low pollen collection was recorded during February and July in Boter-Tolay and Omonda districts due to drought in February and extended rainy period in July Figure 2C.

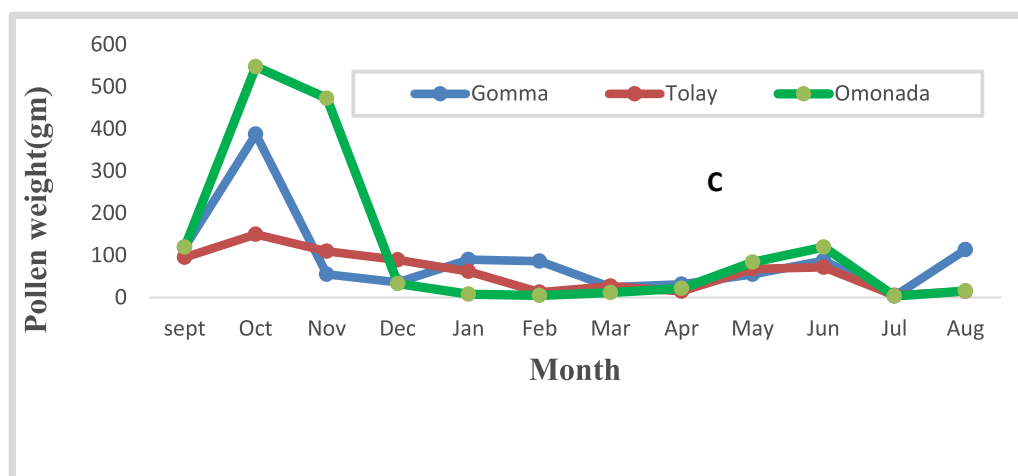
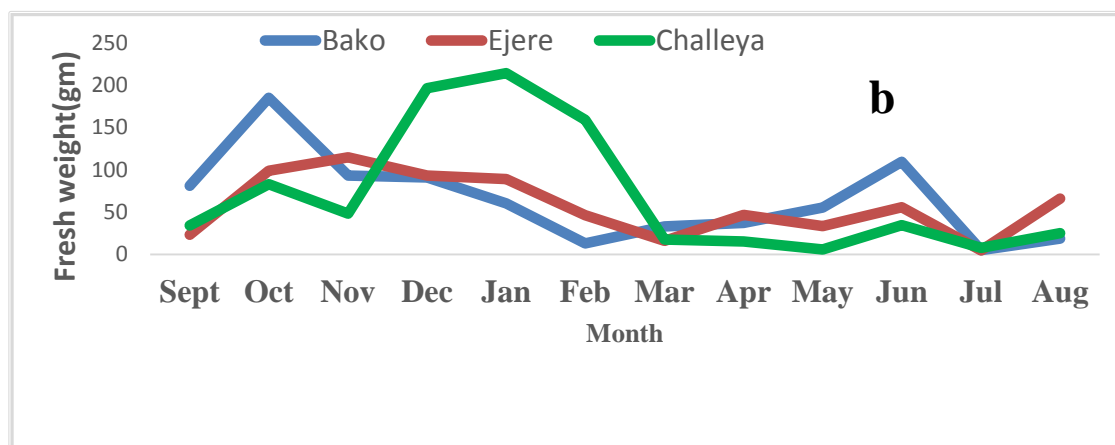
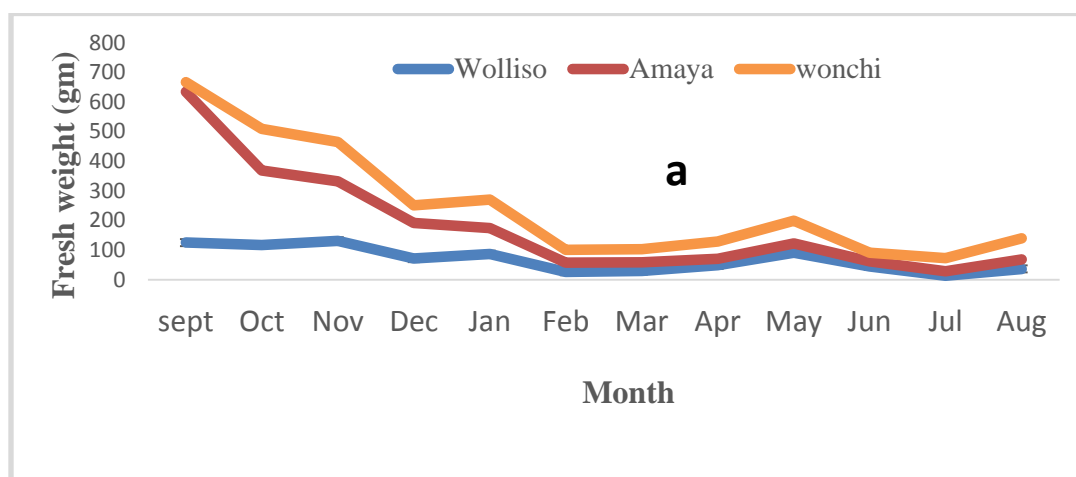


Figure 2 (a-c). Monthly pollen collection in southwest, west shoa and Jimma zones

**Flowering period of major bee forages in south west and west shoa zones**

All identified major bee forages share similar pattern of flowering in both zones due to similarity of agro-ecology and habitat of the plant. About 40.4% of the bee forage species flowers from September to November which include (*Guiztia scabra*, *Andropogon abyssinica*, *Brassica carinata* and *Bidens spp*), 32.3% flower from December to February (*Bersama abyssinica*, *Bidens pilosa*, *Climatis hirusta*, *Rumex*

*nervosus* and *Echinopes macrostachyus*). On the other hand, 17% plant species flower during March to May (*Eucalyptus globulus*, *Eucalyptus Camaludensis*, *Croton macrostachyus*) while 10.3% flowers June-August (*Maesa lanceolata*, *Zea mays*, *Vicia faba*) as indicated in Figure 3. Following the flowering period of bee forages, two honey flow periods were identified in both zones starting from Mid-November to December and May to June. However exceptional minor honey flow period was also documented in Challeya district during January to February due to availability of *Vernonia amygdalina*, *Rubus studneri*, *Olinia roechetiana* but beekeepers are harvesting once a year due lack of awareness on the calendar of bee forages.

Plant species	S	O	N	D	J	F	M	A	M	J	J	A
<i>Acacia polycantha</i>							■	■	■			
<i>Andropogon abyssinica</i>		■	■	■	■							
<i>Bersama abyssinica</i>			■	■	■	■						
<i>Bidens patula</i>	■	■										
<i>Bidens pilosa</i>			■	■	■	■						
<i>Bidens spp</i>	■	■										
<i>Climatis hirsuta</i>				■	■	■						
<i>Echinopes macrostachyus</i>			■	■	■	■						
<i>Eculayptus camludensis</i>							■	■	■			
<i>Erica arborea</i>	■	■										
<i>Eucalyptes globulus</i>							■	■	■			
<i>Guizotia scarba</i>		■	■	■	■							
<i>Maytenus obscura</i>	■	■	■	■								
<i>Maesea lanceolata</i>											■	■
<i>Pavonia urens</i>	■	■	■	■								
<i>Plantago lanceolata</i>			■	■	■	■						
<i>Rumex nervosus</i>			■	■	■	■						
<i>Syzygium guineese</i>			■	■	■	■	■					
<i>Trifolium spp</i>	■	■										
<i>Vernonia spp</i>					■							
<i>Vicia faba</i>												■
<i>Zea mays</i>											■	■

Fig 3. Flowering chart of major bee forages in south west and west Shoa zones

### **Flowering calendar bee forages in Jimma zone**

The majority of bee plant species flowers in the area starting from mid-September to December reaching peak in November. About 30.6 % bee plant species flowers during spring (September to November) which includes *Ageratum conizoides*, *Andropogon abyssinica*, *Bidens spp*, *Cordia africana* and *Guizotia sabra*. Approximately 44.4 % of the species flowered during the dry season (December to February) including *Vernonia amygdalina*, *Coffea arabica*, *Bersama abyssinica* and *Clematis spp*. About 13.5% of the plant species flower during the small rainy season (March-May) which include *Eucalyptus camalduensis*, *Syzygium guineese*, *Mangifera indica* and *Croton macrostachyus* while 11.1% flowered during the big rainy season June-August (*Vicia faba*, *Zea mays* and *Maesa lanceolata*). Following the analysis of flowering period of the bee forages, and seasonal pollen weight collection, three honey flow

season were identified in Gomma district starting from mid-November to December from *Guizotia scabra* honey. The minor honey flow period was identified at end of January and March mainly from *Vernonia amygdalina* and *Coffea arabica*. On the other hand, *Croton macrostachyus* and *Eucalyptus* spp honey were harvested during months of May and June around Gomma, Omonada, and Tolay. The overall flowering period of major bee forages in Jimma zone was indicated in Figure 4.

plant species	S	O	N	D	J	F	M	A	M	J	J	A
<i>Bidens spp</i>	■	■										
<i>Acacia spp</i>								■	■			
<i>Ageratum conizoides</i>		■	■	■								
<i>Albizia schimperiana</i>	■	■										
<i>Andropogon abyssinica</i>	■	■	■	■	■	■						
<i>Aspilia Africana</i>						■	■					
<i>Bersama abyssinica</i>		■	■	■	■	■						
<i>Bidens pilosa</i>			■	■	■	■						
<i>Clematis simensis</i>				■	■	■	■					
<i>Coffea Arabica</i>						■	■	■				
<i>Cordia Africana</i>	■	■										
<i>Crassocephalum vitellinum</i>	■	■										
<i>Croton macrostachyus</i>									■	■	■	
<i>Cyperus spp</i>	■	■										
<i>Datura arborea</i>						■	■	■	■	■		
<i>Dombeya torrida</i>	■	■										
<i>Echionopes macrostachyus</i>						■	■	■	■			
<i>Eucalyptus camaludensis</i>							■	■	■	■		
<i>Gravelia robusta</i>	■	■										
<i>Guizotia scabra</i>		■	■									
<i>Justcia spp</i>	■	■	■	■	■							
<i>Mesea lanceolata</i>											■	■
<i>Mangifera indica</i>											■	■
<i>Persea Americana</i>								■	■	■		
<i>Pterolobium stellatum</i>	■	■	■									
<i>Ranunculas muitifidus</i>	■	■										
<i>Rhus vulgaris</i>				■	■	■						
<i>Rosa abyssinica</i>				■	■	■						
<i>Rumex nervosus</i>				■	■	■	■					
<i>Syzygium guinnese</i>						■	■	■				
<i>Terminalia schimperii</i>				■	■	■	■					
<i>Trifolium spp</i>	■	■										
<i>Vernonia adoensis</i>					■	■	■					
<i>Vernonia amygdalina</i>					■	■	■					
<i>Zea mays</i>										■	■	■

Figure 4: Flowering calendar of major bee forages in Tolay, Gomma and Omonada districts

### Honey pollen analysis

The honey pollen analysis indicated that 41 plant species were identified from the study districts Table 4. The predominant honey source plants representing > 45% pollen count were recorded for *Guizotia scabra*, *Eucalyptus spp*, *Erica arborea*, *Coffea arabica* and *Brassica carinata*. The secondary pollen source plants representing (16-45%) were *Plantago lanceolata*, Grass pollen, *Zea mays*, *Rosa abyssinica*, *Brassica carinata*, and *Echinops macrochaetus*. The minor important pollen source plants ((3-15%) were *Trifolium spp*, *Rumex spp*, *Hypoestes spp*, *Andropogon abyssinica*, and *Vernonia spp*. Among the predominate honey source plants, *Guizotia scabra* appeared the major honey source plant during October and November almost in all study districts while *Eucalyptus spp* is the second pre dominant honey source plants in May and June in Amaya, Wolliso, Gedo and Ejere districts.

Table 4. Predominate and secondary pollen source plants in south west, west shoa and Jimma zones of Oromia

District	Predominant pollen source (> 45%)	secondary pollen (16-45%)	Important minor pollen (3-15%)	Minor pollen source (< 3%)
Wolliso	<i>Guizotia spp</i> (87%)	<i>Eucalyptus spp</i> (31.5%) <i>Brassica spp</i> (37.1%)	<i>Trifolium spp</i> (7.9%) Grass pollen (3.5%)	<i>Croton macrostachyus</i> (2.2%) <i>Datura spp</i> (1.1%) <i>Bersama abyssinica</i> (1.7%)
Amaya	<i>Guizotia spp</i> (92%) <i>Eucalyptus spp</i> (53%)	<i>Eucalyptus spp</i> (24%) Grass pollen (42%) <i>Plantago lanceolata</i> (7.2%)	Grass pollen (16.5%) <i>Sesum indicum</i> 7.2%	<i>Hypotes forskalii</i> (1.9%) <i>Vicia faba</i> (1.4%)
Wondhi	<i>Erica arborea</i> (62%)	<i>Eucalyptus spp</i> (18.75)	<i>Vernonia spp</i> (6.25%) <i>Rumex spp</i> (6.25%) <i>Brassica spp</i> (8%) <i>Solanum spp</i> (6%)	<i>Justicia spp</i> (3%) <i>Bersama abyssinica</i> (1%) <i>Vernonia spp</i> (1%)
Challeya	<i>Guizotia scabra</i> (67%) <i>Eucalyptus spp</i> (95%)	<i>Echinops macrostachyus</i> (24%)	<i>Trifolium spp</i> (6%) Grass pollen (7%)	<i>Plantago</i> (2%) <i>Hypoestes spp</i> (1%) <i>Trifolium spp</i> (2%)
Bakotibe	<i>Guizotia spp</i> (78%) <i>Brassica spp</i> (47%)		<i>Hypoestes spp</i> (9%) <i>Eucalyptus spp</i> (6%) <i>Echinops spp</i> (9%)	<i>Rumex spp</i> (1%) <i>Vicia faba</i> (1.4%) <i>Sesum indicum spp</i> (5%)
Ejere	<i>Eucalyptus spp</i> (57%)	<i>Guizotia spp</i> (23%) <i>Eucalyptus camaluensis</i> (19%)	Grass(11%) <i>Echinops spp</i> (9%)	Grass spp (3%) <i>Echinops spp</i> (1%) <i>Hypotes forskalii</i> (1%)
Boter-Tolay	<i>Guizotia spp</i> (65%)	<i>Croton macrostachyus</i> (37%)	<i>Coffea arabica</i> (10%)	<i>Ageratum spp</i> (3%) <i>Acacia spp</i> (2%) <i>Terminalia spp</i> (1%)
Gomma	<i>Coffea arabica</i> (49%)	<i>Vernonia amygdalina</i> (35%) <i>Guizotia scabra</i> (15%)	<i>Schefflera abyssinica</i> (4%) <i>Hypoestes spp</i> (4%)	<i>Saturja spp</i> (2%) <i>Allophylus abyssinica</i> (1.2%) <i>Gravillea roubusta</i> (1.2%) <i>Olea africana</i> (2.17%)
Omonada	<i>Guizotia scabra</i> (65%)	<i>Croton macrostachyus</i> (10%) <i>Eucalyptus spp</i> (3%)	<i>Syzygium guineese</i> <i>Terminalia schimperi</i>	<i>Hypoestes spp</i> (1%) <i>Zea mays</i> (2%) <i>Solanum spp</i> (1.3)

### Monofloral honey

The classification of pollen percentage count using PCA clustering indicated that five monofloral honey types were identified, for the three zones. Accordingly, for south west and west shoa zones *Guizotia* spp, *Erica arborea* and *Eucalyptus* honey are the most dominant types of monofloral honey (Figure 6a, b). The *Guizotia* monofloral honey types cover large poroportion of cultivated areas of Amaya, Woilso, Bako Tibe and Tolay while *Eucalyptus* honey is found in Amaya, Wollisso and Ejere districts of west shoa and south west shoa zones of Oromia (Figure 6ab). On the other hand, *Coffe arabica* honey, with some degree of mix with *Vernonia amygdalina* and *Croton macrostachyus* honey, was found in Gomma district while *Croton macrostachyus* honey was found in Omonada and Chora-Boter-Becho forest in Jimma zone (Figure 6c). The pollen count dominance for monofloral honey ranges from 45 % to 100%. From the total number of species of pollen grains found in the monofloral honey, *Guizotia* honey is represented by 19 species, *Eucalyptus* honey by 10 plant species, *Coffea arabica* honey by five species, *Erica arborea* honey by six species and *Croton macrostachyus* honey by 9 species (Figure5ab).

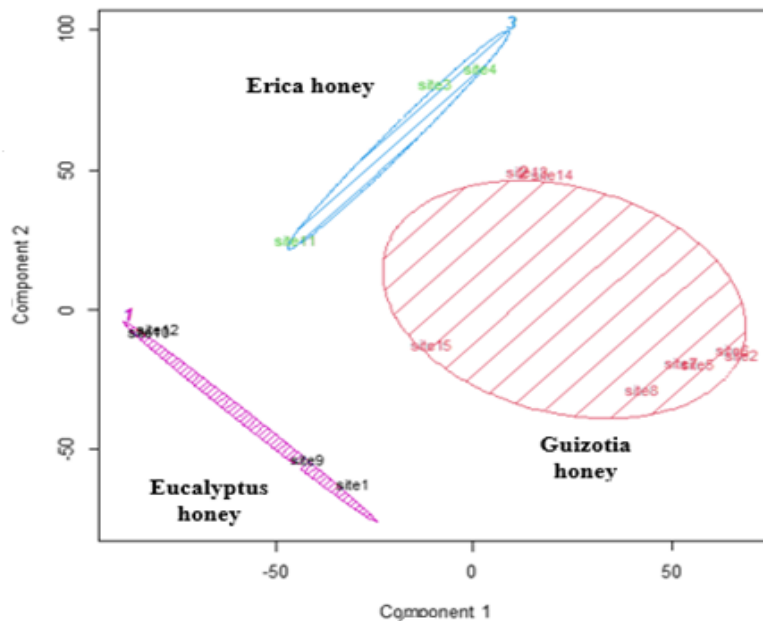


Figure 5a. Monofloral honey types from south west shoa zone

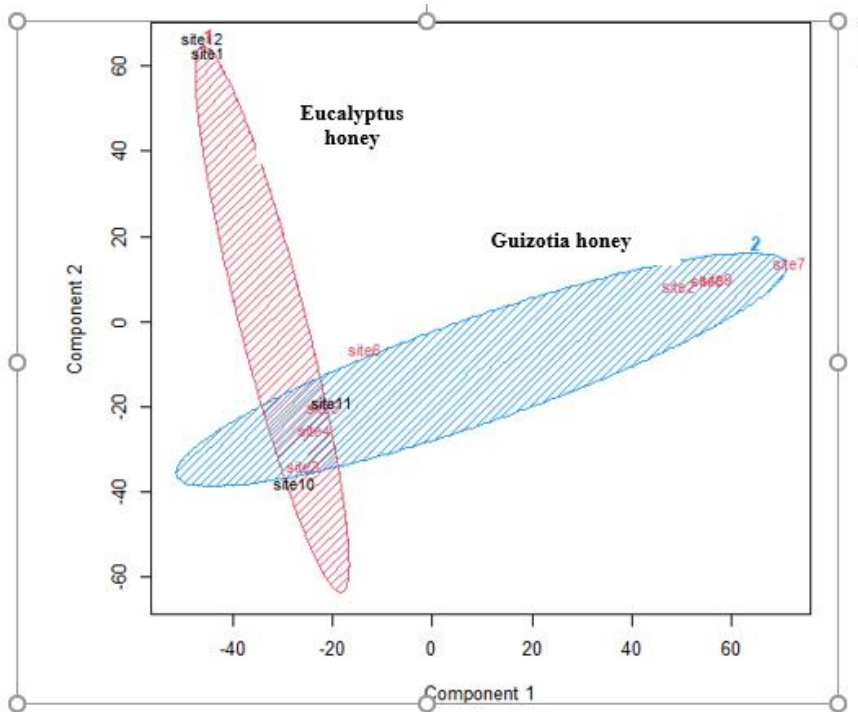


Figure 5b. Monofloral honey types from west shoa zone

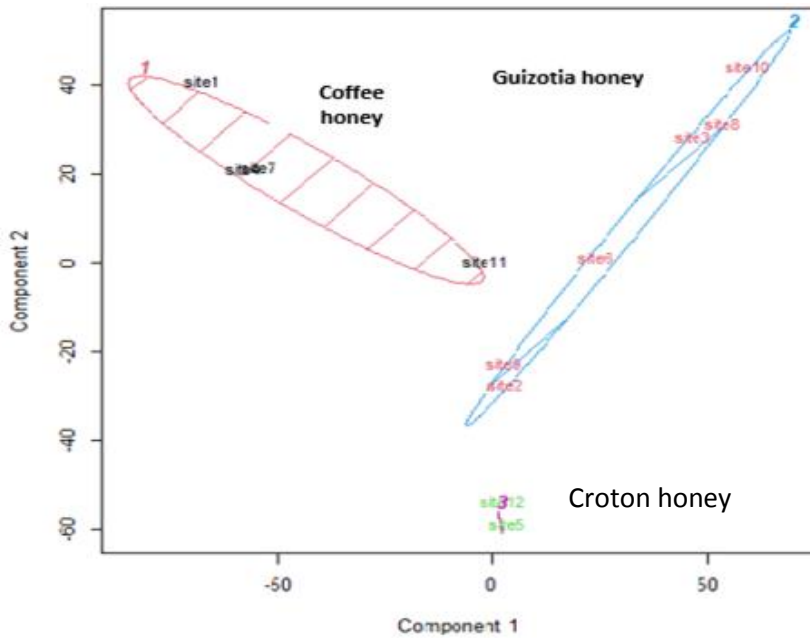


Figure 5c. Monofloral honey types from Jimma zone

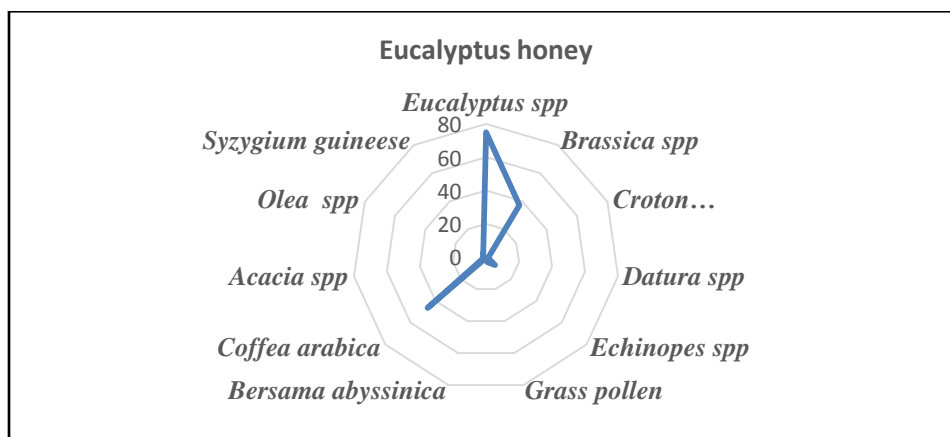
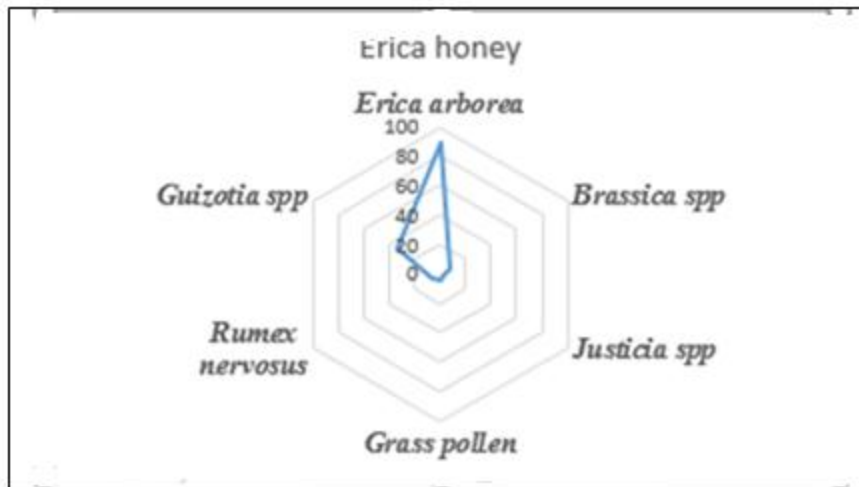
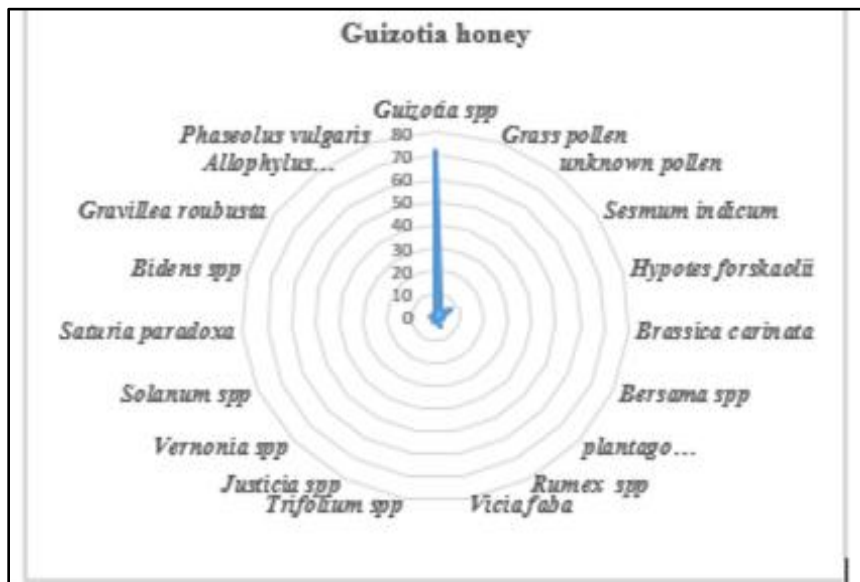


Figure 6a, Spider web distribution for Monofloral honey of *Guizotia scabra* honey, *Erica arborea* and *Eucalyptus spp*



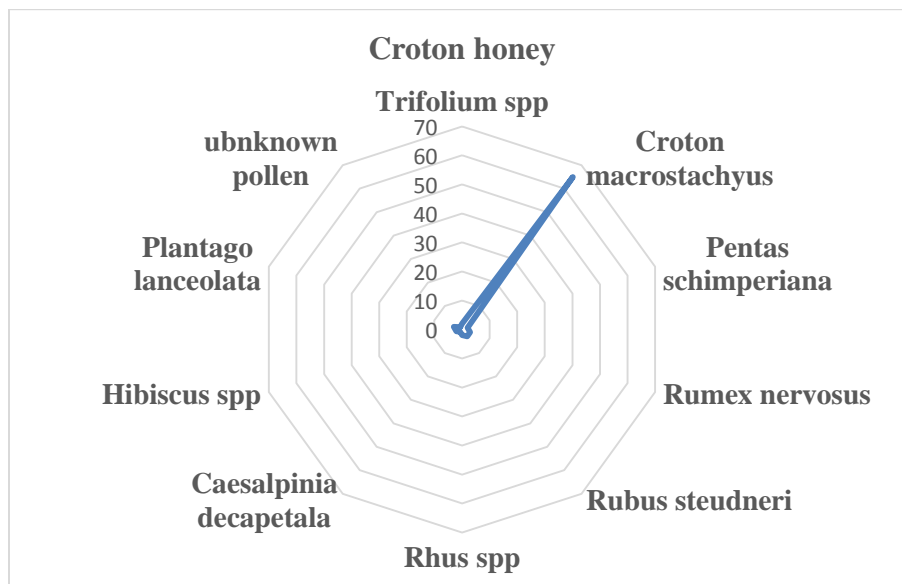
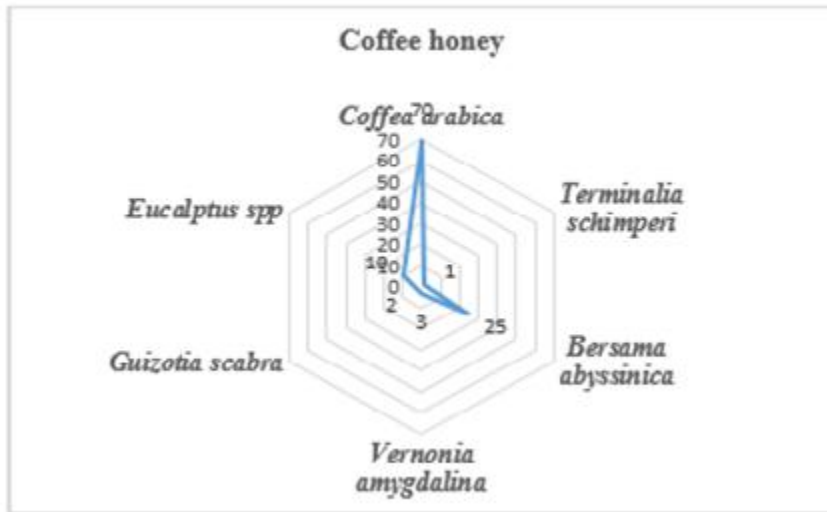


Figure 6b. Spider web distribution for Monofloral honey of *Coffea arabica* and *Croton macrostachyus*

## Discussions

From pollen load collection and honey pollen analysis, 101 plant species were identified belonging to 32 families. Among the plant families Asteraceae, Fabaceae, Acanthaceae and Lamiaceae are dominant families, comprising higher number of bee forage species composition in the area. These families are also reported to be the most species-rich families in the Flora of Ethiopia and Eritrea (Ensermu Kelbessa, 2014, Admassu et al. 2014). Moreover the Asteraceae is one of the pollinator dependent plant family and comprises various species of weeds, and cultivated oil crops. This agrees with (Admassu and Tura 2018), Asteraceae has attractive flower color to be pollinated by different insect pollinators including honeybees and favoring them to colonize wider ecological niches for honey production.

Analysis of growth habit of the plant revealed that the herbaceous flora contributes for 57% of the total species composition of the area. This could be due to removal of shrubs and trees from farmland for cultivation and forest encroachment favoring the growth of herbaceous flora. This is in agreement with Admassu and Tura 2017 agricultural practices favor for the growth various weeds and other herbaceous flora.

### ***Identification of major bee forages from pollen trap***

The identification of pollen loads from pollen traps is very important because it gives about the preferences and resource richness as the main sources pollen for honeybees. The identification of bee-collected pollen loads from pollen traps indicated that not only honeybees collected pollen from plant species but also it shows the relative importance of each plant species as source of pollen for honeybees. A total of 41 plant species were identified from pollen trap, however the large quantity of pollen came from only a few plant species such as *Bidens spp*, *Eucalyptus spp*, *Guizotia scabra*, *Plantago lanceolata*, *Maesa lanceolata*, *Trifolium spp*, *Vernonia spp*, *Terminalia schimperiana* and *Zea mays*. The contribution of each species as major source of pollen is due to abundance and potentiality of plant for nectar and pollen. This result is also supported by findings of Abebe *et al* 2013 and Debissa and Admassu 2008, who reported that honeybees forage on more productive and profitable plants that provide necessary nutrition nearby their hives.

### ***Seasonal pollen collection***

The analysis of monthly pollen weight for the different districts of the study area indicated that there was significance difference of pollen weight between the districts as well as seasons. The fresh weight of pollen collected at different months of the year indicated that significant amount of pollen was collected during September, October and November but decline of incoming pollen collection was recorded during, February and March in Wolliso, Amaya and Wonchi districts of the southwest shoa zone. Similar trend was observed for west Shoa zone particularly in Ejere and Bako districts. The highest fresh weight of pollen collection was occurred during active period (October and November) is due to availability of abundant bee forages including field crops, weeds and naturally growing shrubs and trees providing abundant pollen and nectar for honeybees (Admassu *et al.* 2014, and Abebe *et al.*, 2005). However, a significance decline of pollen weight was recorded during months of February and July in all districts except for Challeya district. The highest pollen collection occurred in Challeya district during December to February is due to availability of flowers of the different species of *Vernonia*, *Rubus studneri*, and *Olinia rochetiana*. On the other hand, in Jimma zone (Tolay, Gomma and Omonada) the highest fresh weight of pollen was collected during the months of October, November and December due to extended rainy season resulting in longer duration of plants in flower. In addition, there are some drought tolerant species such as *Vernonia amygdalina*, *Bersama abyssinica*, *Terminalia brownii* and *Coffea arabica*. This is agreeing with (Abera, 2017, Tura and Admassu 2019) who reported that *Vernonia Spp*, *Coffea arabica*, and *Croton macrostachyus*, are the major bee flora flowering both in dry period and small rainy season in the Gomma district. According to the same author *Vernonia amygdalina* is a very valuable honey source plant in the area. In this district low pollen collection was occurred during the months of March due to availability of low moisture in the soil resulting drying of flowers. Relatively higher pollen collection was recorded during the months of April, May and June for Gomma, Tolay and Omonada districts due to availability of *Croton macrosatchyus* and similar finding also reported by Abera *et al.*, 2017, Chala *et al.* 2012.

### ***Flowering calendar of the bee forages in west and south west shoa***

From field observation and pollen load collection the highest percentage of flowering bee forages occurred during September to November reaching peak flowering in October in all study districts. The availability of higher proportion of flowering species during October to November is due to occurrence of summer rains in June, July and August and at the end of this rainy period, majority of the plants come into flower. This finding agrees with Fichtl and Admassu 1994, Nuru *et al.* 2001 and Tessega, 2009, Tura and Admassu 2020, majority of bee forages species flower after the heavy rain in July through September known as “Kremt” or summer rain and most of the Ethiopian highlands are colored with golden-yellow flower of *Bidens* spp, indigenous oil crops, weeds including *Trifolium* spp. Similarly, April to May also one of the minor flowering period in Amaya, Woliso, Ejere and Challey districts due to availability *Eucalyptus* plantation around boundaries of farmland and homegraden. Similar finding also reported by Nuru *et al.* 2001, Amssalu Bezabeh 2005, Mesert Gemada 2010, Abera *et al.* 2015 who reported that *Eucalyptus* is one of the important source of honey in central highlands of Ethiopia. Relatively low number bee forages are available in March and July due to low moisture condition in March and high rainfall in July. This is supported with (Admassu and Debissa 2009) who stated that during the rainy season, low temperatures possibly inhibit growth and flowering of bee forages, whereas the higher temperature during dry period causes water deficiency in plants resulting in low nectar secretion and pollen production. Similar flowering pattern was observed for Tolay, Gomma and Omonada in which majority of bee forages flowering from October to December due to extension of summer rainfall creating availability of enough moisture until end of December. Exceptionally some plant species including *Vernonia amygdalina*, and *Coffea arabica* abundantly flowering from January to February and minor honey flow is expected in Gomma district. This study also agreement with Abera 2017, Tolera and Dajene, Chala 2012 indicated that *Vernonia Spp.*, *Coffea arabica*, *Schefflera abyssinica* and *Croton macrostachyus* were common honey bee floras in Jimma zone flowering from January to May.

### ***Pollen analysis of honey***

Pollen spectra of honey analysis revealed that a variety of pollen source plants were identified ranging from predominate, secondary pollen and minor important. The degree of the dominance of the bee forages depend on abundance, nectar and pollen potential of the plants. Based on honey pollen analysis the highest bee forage diversity of bee plants was found in the honey sample collected from Tolay and Ejere as compared to the rest of the districts since the study area is located in central highland which is known for high plant diversity. Similar finding by Bareke & Addi (2018) in the Borena Zone of Oromia, Ethiopia. The PCA analysis of honey samples of the study districts revealed that, *Guizotia spp*, contributed more than 72% of the pollen count. The dominance of *Guizotia* pollen from honey can be attributed to widespread distribution in agricultural lands, fallow land and since it is associated weed of major crops that grow in within and at the margin of field crops. This is agreeing with Debissa and Admassu 2009, Mesert 2018, Tura and Admassu 2019 who reported that *Guizotia scabra* was a predominant monofloral honey source plant in east and west shoa and Borena zones of Oromia. The identification of *Erica arborea* monofloral honey in wonchi district is due to availability of *Erica arborea* on the top of mountain nearby Lake Wonchi. The *Erica* monofloral honey is reported by Taye Beyene 2014 and Nuru and Admassu 2001 in west shoa zone. The third honey type identified was *Eucalyptus* mono floral honey which is wide spread in all study districts and separated as distinct honey types in PCA plot. The availability of *Eucalyptus* Monofloral honey have been reported by Abera 2015 in Addis Ababa, (Admassu and Debissa, 2008), in Menagesha suba and (Mesert Gameda 2010) in Ada berga

and Ejere districts in west shoa zone. The availability of coffee honey and croton honey in Jimma zone were reported by Abera , 2017 and Tura and Admassu 2020).

### **Conclusion and Recommendation**

Based on direct field observation, and pollen load collection 101 plant species were identified and herbaceous flora comprising the major portion of bee forages in the area. The higher proportion of the pollen weight was collected from a few plant species including *Guizotia* spp, *Vernonia* spp , *Eucalyptus camaludensis*, *Echinops macrostchyus*, *Bidens pilosa*, *Mesea lanceolata*, *Planatgo lanceolate*, *Echinopes macrostachus* . Majority of plant species flower two times a year during September to November and April to May in most districts. However, in Challeya and Gomma districts three peak pollen collection periods were recorded during October, January to February and May. Based on floral calendar, the scarcity of pollen collection was occurred in early march and July and feeding colonies may require. Thus creation of awareness and on farm demonstration about the important of floral calendar for appropriate management of honeybee colonies is recommended to increase frequency of honey harvest. Moreover *Masea lanceolata* was identified as major bee forages during rainy and further study on the rapid propagation methods is recommended.

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Appendix 1. Checklist of plant species identified from pollen load collection and pollen analysis of honey

plant species	Family	Habit	Food source	Flowering period
<i>Abutilon angulatum</i>	Malvaceae	shrub	N&P	Oct-Dec
<i>Acacia abyssinica</i>	Fabaceae	Tree	N&P	sept-Oct
<i>Acacia spp</i>	Fabaceae	Tree	N&P	sept- oct
<i>Ageratum houstonianum</i>	Asteraceae	Herb	N&P	Oct-Dec
<i>Albizia spp</i>	Fabaceae	Tree	N&P	Sept- Oct
<i>Andropogon abyssinica</i>	Poaceae	Herb	P	sept -Oct
<i>Apodytes dimidata</i>	Icniaceae	Tree	NP	sept- oct
<i>Aspilia Africana</i>	Asteraceae	Herb	P	Oct-Dec
<i>Bartsia trixago</i>	Schurophulaceae	Herb	NP	sept- oct
<i>Basilicum polystachyum</i>	Lamiaceae	Herb	NP	sept- oct
<i>Belpharis linariifolia</i>			NP	
<i>Blepharis brevicilata</i>	Acanthaceae	Herb		sept- oct
<i>Bersama Abyssinia</i>	Meliantaceae	Tree	NP	sept- Dec
<i>Bidens pachyloma</i>	Asteraceae	Herb	P	sept-Oct
<i>Bidens pilosa</i>	Asteraceae	Herb	P	sept- Dec
<i>Brassica carinata</i>	Brassicaceae	Herb	NP	Sept-Oct
<i>Brucea antidysenterica</i>	Simaroubaceae	Shrub	NP	Sept-Oct
<i>Cadaba glandulosa</i>	Capparidaceae	Shrub	P	Nov-Dec
<i>Caesalpinia decapetala</i>	Fabaceae	Climber	P	Nov-Dec
<i>Calpurnia aurea</i>	Fabaceae	Shrub	NP	Nov-Dec
<i>Carduus nyassanus</i>	Asteraceae	Herb	P	Sept-Nov
<i>Cassipourea malosana</i>	Rhizophoraceae	Tree	NP	sept-Oct
<i>Caylusea abyssinica</i>	Resedaceae	Herb	P	Oct- Dec
<i>Cicer arietinum</i>	Fabaceae	Herb	NP	Oct- Dec
<i>Cirsium schimperi</i>	Asteraceae	Herb	P	Oct-Dec
<i>Clematis simensis</i>	Ranunculaceae	Herb	N	Oct-Dec
<i>Cleome usambarica</i>	Capparidaceae	Herb	NP	Oct-Dec
<i>Coffea Arabica</i>	Rubiaceae	Shrub	NP	Jan- Feb
<i>Crambe abyssinica</i>	Brassicaceae	Herb	NP	Oct-Dec
<i>Crassocephalum vitellinum</i>	Asteraceae	Herb	P	Oct-nov
<i>Croton macrostachyus</i>	Euphorbiaceae	Tree	NP	May- June
<i>Cyperus spp</i>	Cyperaceae	Herb	P	Oct- Dec
<i>Datura arborea</i>	Solanaceae	Shrub	NP	Nov- Jan
<i>Discopodium peninervum</i>	Solanaceae	Shrub	NP	Nov- dec
<i>Dombeya torrida</i>	Sterculiaceae	Tree	NP	sept-Oct
<i>Echinops macrochaetus</i>	Asteraceae	Herb	P	Oct-Dec
<i>Ecualyptus camaldulensis</i>	Myrtaceae	Tree	NP	Mar- May
<i>Ekebergia capensis</i>	Meliantaceae	Tree	NP	Jan- Feb
<i>Erica arborea</i>	Eriaceae	Shrub	NP	Sept-Oct
<i>Eucalyptus globulus</i>	Myrtaceae	Tree	NP	Mar- May

plant species	Family	Habit	Food source	Flowering period
<i>Euclea schimperi</i>	Ebenaceae	Shrub	NP	sept-oct
<i>Glycine weightii</i>	Fabaceae	Herb	NP	sept-oct
<i>Gouinia longispicata</i>	Rhamnaceae	Climber	NP	sept- Dec
<i>Grevillea robusta</i>	Proteaceae	Tree	NP	sept-oct
<i>Guizotia spp</i>	Asteraceae	Herb	NP	sept-oct
<i>Guizotia scabra</i>	Asteraceae	Herb	NP	Oct-Nov
<i>Hagenia abyssinica</i>	Roseaceae	Tree	NP	sept-oct
<i>Helianthus annuus</i>	Asteraceae	Herb	NP	sept-oct
<i>Heliotropium cinerascens</i>	Boraginaceae	Herb	P	sept-oct
<i>Helminthotheca echioides</i>	Asteraceae	Herb	NP	sept-oct
<i>Hibiscus spp</i>	Malvaceae	shrub	NP	Oct-Nov
<i>Hypoestes spp</i>	Acanthaceae	Herb	NP	Oct-Nov
<i>Hypoestes triflora</i>	Acanthaceae	Herb	NP	sept-oct
<i>Impatiens hochstetteri</i>	Lamiaceae	Herb	N	sept-oct
<i>Isoglossa laxa</i>	Acanthaceae	Herb	NP	sept-oct
<i>Justicia heterocarpa</i>	Acanthaceae	Herb	NP	sept-oct
<i>Justicia schimperana</i>	Acanthaceae	Herb	NP	sept-oct
<i>Lens culinaris</i>	Fabaceae	Herb	NP	sept- Dec
<i>Leonotis ocyimifolia</i>	Lamiaceae	Herb	NP	sept-oct
<i>Maesa lanceolata</i>	Myrsinaceae	Shrub	P	Aug- Oct
<i>Malva verticillata</i>	Malvaceae	Herb	NP	sept-oct
<i>Maytenus arbutifolia</i>	Clestraceae	Shrub	NP	Oct-Nov
<i>Maytenus obscura</i>	Clestraceae	Shrub	NP	Oct-Nov
<i>Melitous spp</i>	Fabaceae	Herb	NP	sept-oct
<i>Musa paradisca</i>	Musaceae	Herb	NP	sept-oct
<i>Ocimum spp</i>	Lamiaceae	Herb	NP	sept-oct
<i>Ocimum basilicum</i>	Lamiaceae	Herb	NP	sept-oct
<i>Ocotea kenyensis</i>	Rubiaceae	Shrub	NP	Sept-Oct
<i>Olea europa sub spp africana</i>	Oleaceae	Tree	NP	May- June
<i>Parkinsonia aculeate</i>		shrub	NP	sept-oct
<i>Pavonia urens</i>	Malaveae	Shrub	NP	sept-oct
<i>Phyllanthus reticulate</i>	Malavaeae	Herb	NP	sept-oct
<i>Phytolacca dodecandra</i>	Phytolaceae	Herb	NP	Sept-Dec
<i>Pisum sativum</i>		Tree	NP	sept-oct
<i>Plantago lanceolata</i>	Plantaginaceae	Herb	P	Sept-Dec
<i>Prunus Africana</i>	Roseaeae	Tree	NP	sept-oct
<i>Psidium guajava</i>	Myrtaceae	shrub	NP	sept-oct
<i>Rhamnus perinodes</i>	Rhamanceae	Shrub	NP	sept-oct
<i>Rosa abyssinica</i>	Roseaeae	Shrub	P	sept-Dec
<i>Rubus steudneri</i>	Roseaeae	Shrub	P	Oct-Dec
<i>Rumex nepaleniss</i>	Polygonaceae	Herb	P	Oct-Dec
<i>Rumex nervosus</i>	Polygonaceae	Herb	P	Oct-Dec

<b>plant species</b>	<b>Family</b>	<b>Habit</b>	<b>Food source</b>	<b>Flowering period</b>
<i>Salix subserata</i>	Saliceae	Shrub	P	sept-oct
<i>Saturja paradoxa</i>	Lamaiceae	Herb	NP	sept-oct
<i>Schefflera abyssinica</i>	Araliaceae	Tree	N	March-April
<i>Schinus molle</i>	Anacardaceae	Tree	P	sept-oct
<i>Sesamu indicum</i>	Pedaliaceae	Herb	NP	sept-Dec
<i>Silybum marianum</i>	Asteraceae	Herb	P	sept-oct
<i>Solanium spp</i>	Solanaceae	Herb	P	sept-oct
<i>Sorghum bicolor</i>	Poaceae	Herb	P	sept-oct
<i>Syzygium guinnese</i>	Myrtaceae	Tree	NP	Feb -Mar
<i>Tagetes minuta</i>	Asteraceae	Herb	P	sept-oct
<i>Terminalia schimperiana</i>	Combretaceae	Tree	NP	Dec-Jan
<i>Trifolium spp</i>	Fabaceae	Herb	NP	Sept-Oct
<i>Unknown Grass pollen</i>	Poaceae	herb	P	Sept-Dec
<i>vernonia amygdalina</i>	Asteraceae	Shrub	NP	Dec-Jan
<i>Vernonia leopoldii</i>	Asteraceae	Shrub	NP	Dec-Jan
<i>Vicia faba</i>	Fabaceae	Herb	NP	Aug- Oct
<i>Zantedeschia aethiopica</i>	Aracieace	Herb	NP	Aug- Oct
<i>Zea mays</i>	Poaceae	Herb	P	Aug- Oct



# Nectar secretion dynamics and honey production potential of some herbaceous plants in South West Shoa Zone

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## Abstract

*Honey production potential of bee plants is varied from species to species. Thus, this study was conducted to quantify the nectar secretion dynamics, and honey production potential of *Hygrophila auriculata*, *Salvia leucantha* and *Pavonia urens*. One day before nectar collection, five inflorescences were enclosed with mesh wire to measure accumulated nectar volume. Additionally, nectar volume and concentration, temperature, and humidity were measured from flowers covered with mesh bag at interval of 3 hours. One way Anova and linear regression model were used for data analysis. Accordingly nectar secretion dynamics of the three species were significantly varied ( $p < 0.05$ ) at different times of the day. The nectar volume became available between the two consecutive measurements (3hr intervals) varied from 2.24 to 5.79  $\mu\text{l}/\text{flower}$ , 1.5 to 2.5  $\mu\text{l}/\text{flower}$ , and 4.1 to 5.5  $\mu\text{l}/\text{flower}$  for *P. urens*, *H. auriculata* and *S. leucantha*, respectively. This variation reflects the differences in the dynamics of nectar secretion between the species. Temperature was positively correlated with nectar concentration for all species. However, temperature was negatively correlated with nectar volume of *P. urens* and *H. auriculata* whereas almost at equilibrium for *S. leucantha*. Humidity was negatively correlated with nectar concentration for all species whereas it was positively correlated with nectar volume for *P. urens* and *H. auriculata*. However, the relationships between humidity and nectar volume were almost found at equilibrium for *S. leucantha*. Based on the mean amount of nectar sugar secreted by the plants, the mean honey production potentials of the species were estimated to be 35 kg/ha, 29.88 kg and 60.2 kg/ha depending on the size of the plants for *P. urens*, *H. auriculata* and *S. leucantha*, respectively. Therefore, further propagation and in-situ conservation of these species are also recommended for sustainable honey production.*

**Keywords:** Nectar, Honey, Bee plant, Sugar, Humidity

## Introduction

Ethiopia has suitable environmental conditions for the existence of diversified natural resource which is suitable for beekeeping. There are abundant flowering plants, most of them are honeybee plants (Addi et al., 2014). Honeybee plants are those plant species that provide bees food in the form of pollen and nectar (Bareke & Addi, 2019). The contribution of honeybee plant species for honey production depend on the floral morphology, anatomical structure of flowers (Kim et al., 2017), flowering duration, quality and quantity of nectar secreted (Alqarni et al., 2015). The quality and quantity of nectar is affected by environmental factors (Adgaba et al., 2017). Since nectar is exposed to evaporation under the influence of temperature, humidity (Bareke et al., 2021), wind speed and solar irradiation (Dafni, 1992). It is necessary at least to convoy every nectar measurement with the measurements of temperature and humidity.

For boosting of honey production potential of bee forages is very essential to estimate the honey bee colony carrying capacity of a given vegetation type without negatively affecting honey production

capacity of single colony (Bareke et al., 2021a). The honey production potential of bee plants is varied from plant species to species (Bareke et al., 2021b). In every geographical region there are very few important honey source plants that provide honey to the world depending on nectar and pollen potential of the plants (Adgaba et al., 2017). Hence, it has supreme importance to characterize them according to their degree of honey production capacity.

Nectar secretion dynamics and honey production potential of different bee forage plants have been done by different authors. Accordingly, the honey production potential were known for *Acacia gerrardii* (Alqarni et al., 2015); for *Otostegia fruticosa* and *Ziziphus spina-christi* (Adgaba et al., 2017); for *Croton macrostachyus* and *Schefflera abyssinica* (Bareke et al., 2020a,b) and for *Coffea arabica* (Bareke et al., 2021b). In Ethiopia, the knowledge of nectar secretion dynamics and potential for honey production is relatively a new impression for the bee forage evaluation. For many important bee forage plant species, nectar secretion capacity and its contribution to honey production have not yet documented. These important plant species include one of the most important melliferous species, *Pavonia urens* Cav.; *Hygrophila auriculata* (Schum.) Heine, and *Salvia leucantha* Cav.

*Pavonia urens* is one of an important honey source plants (Addi et al., 2014). It is a perennial herb growing up to 2 m high and it belongs to the Malvaceae family. It grows along edges, paths and clearings in upland and riverine forests, secondary forest and scrub, abandoned cultivations and as a weed at altitudes between 1500 and 3000 m throughout the Ethiopian highlands (Fichtl & Adi, 1994). *P. urens* is a medicinal plant that used to treat Pneumonia and stomachache (De Boer et al., 2005) as well as it provides nectar and pollen for honeybees (Addi et al., 2014).

*Hygrophila auriculata* is an erect sparingly branched perennial or annual herb with square stems and elliptic leaves that belongs to Acanthaceae family (Fichtl and Admassu, 1994). It grows at altitudes as low as 500 m above sea level in areas waterlogged for part of the year, and up to 2800 m in grassland and along paths and forest edges. It is eaten by cattle and the seeds can produce semi-drying oil. The fleshy root has a cooling, slimy taste and a marshy smell. *H. auriculata* gives flower from August to February; honeybees collect pollen and nectar from the flowers frequently. The plant is an important honey source in lower parts of the Ethiopia (Fichtl and Admassu, 1994).

*Salvia leucantha* is a perennial ornamental herb, growing up to 40-100 cm and much branched, densely pubescent with white hairs. It flowers whitish pink or pale violet. It is cultivated as an ornamental and often naturalized in the wetter regions of the Flora area, at altitudes between 1800 and 2800 m, and also in upland Eritrea and elsewhere but native in Mexico. *Salvia leucantha* gives flowers after rainy season and it is an excellent source of bee forage during dry period (Addi et al., 2014). However, nectar secretion dynamics and honey production prospective of these plant species have not been known. Therefore, the main objective of this study was to identify the nectar secretion dynamics and estimate honey production potential of *Pavonia urens*; *Hygrophila auriculata* and *Salvia leucantha*.

## **Materials and Methods**

### ***Study area***

The experiment was carried out in south west Shoa Zone, Ethiopia. Study sites were selected based on the availability and abundance of the species. Moreover the study species were selected based on their

ecological adaptation range, foraging intensity of honeybees and accessibility of the flower for nectar measurement.

***Estimating number of flowers per plant and plant per area***

Ten plots were randomly taken to determine the number of plants per plot from each site. Plot size was 2m\*2m for *Pavonia urens* and *Salvia leucantha* and 1m\*1m for *Hygrophila auriculata*. In addition plants per plot were taken to count the number of flowers per plant. Accordingly, average number of flowers per hectare= average number of plants/ha \* average flowers/plant.

***Nectar volume and solute concentration measurement***

Prior to nectar removal the inflorescences were covered with fine mesh for 24 hours to prevent removal of nectar by visitors. Flowers were marked at random from different inflorescence parts (Wyatt, Broyles, & Derda, 1992). Accumulated nectar for 24 hours was taken from 20 flowers at random per day for three days (Esteves, Villadelrey, & Rabajante, 2014).

***Identifying the nectar secretion duration***

Time of opening of flowers, pollen release and nectar secretion were taken. Fifteen (15) individual flowers were measured daily from the start to the end of nectar secretion to determine the nectar secretion duration of the plants (Bareke, Kumsa, & Addi, 2020).

***Determining nectar secretion dynamics***

Data of nectar volume, nectar concentration, temperature and humidity were taken at three hours intervals from 6:00-18:00 hours simultaneously (Wyatt et al., 1992) depending the nectar secretion durations of plant species. For each plant and sampling time and the nectar volume was measured from an average of 5 individual flowers at a time (Esteves et al., 2014).

***Calculation of sugar amount in nectar per flower***

Amount of sugar found in nectar was calculated from the nectar volume, concentration and sucrose density. Nectar concentration was converted to sucrose density using Pry-jones and Corbet (1991) equation

$$P=0.003729/C + 0.0000178C^2 + 0.9988603,$$

Where, p=the sucrose density for a given value of C and C is the refractometer reading.

Amount of sugar was calculated using (Dafni, 1992) equation.

$$\text{Nectar volume (mg) per flower} = \frac{\% \text{ sugar reading in the refractometer}}{100} \times (\text{Nectar volume (}\mu\text{l)/flower}) \times (\text{Density of sucrose at the observed concentration})$$

***Estimation of honey production potential (HPP)***

Honey production potential of the plants was estimated as the following: Average amount of sugar per hectare = Average number of flowers per ha \* average amount sugar per flower \* nectar secretion days. Average amount of sugar per m<sup>2</sup> was converted to hectares (Dafni, 1992; Masierowska, 2003; Kim et al., 2017). The mean amount of sugar per hectare was converted to honey.

At the international market the average moisture content of the honey is 18% from 1kg while 82% is sugar. This was used to convert the mean amount of sugar produced per hectare per flowering season to honey. The honey estimated from the sugar was the honey production potential of the plants (Bareke, Kumsa, & Addi, 2020).

### Data analysis

Data was analyzed using descriptive statistics and One-way ANOVA. Tukey used for mean separation among the treatments. In addition to this, a linear regression model was computed using R-software to see the effect of temperature and humidity on nectar volume, concentration and calculated sugar.

### Results

The mean numbers flowers/ha were the highest for *S. leucantha* whereas the lowest for *P. urens* (Figure 1 a). The more branched plant species provide the highest number of flowers per plant. On the other hand, the nectar secretion durations of *P. urens* were longer than the two species (Figure 1b). This difference happened due to the nature of the plants species. The mean nectar secretion longevity of *P. urens* was for 10 days with a range of 9-12 days whereas 7-10 for *H. auriculata* and *S. leucantha*.

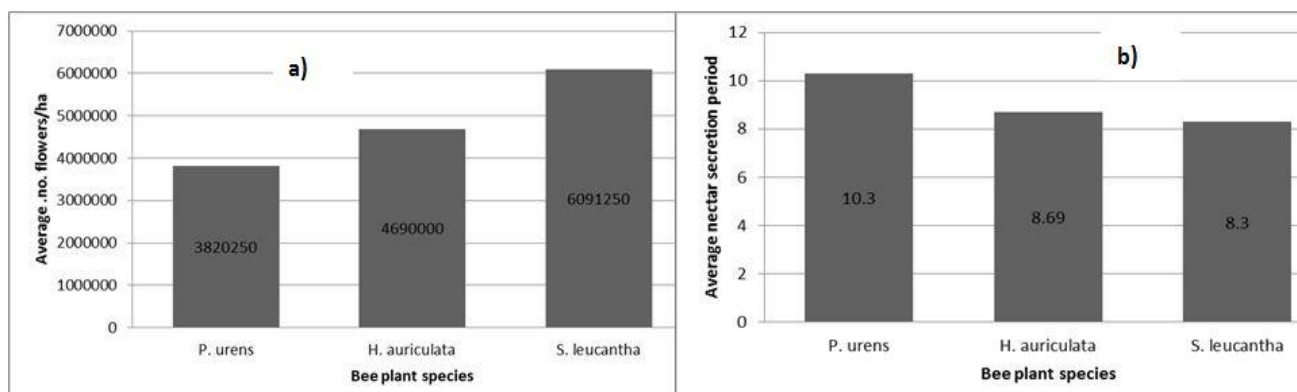


Figure 1: Mean no. of flowers/ha and mean nectar secretion length of flower/season for *P. urens*, *H. auriculata* and *S. leucantha*

### Nectar secretion dynamics

The nectar volume was significantly different ( $p < 0.05$ ) in different times of the day with the highest value at 6 hour whereas the lowest nectar volume was obtained at 15 hour (Table 1) for *P. urens*. In the morning (6:00 hour) there was the highest humidity in the area. Due to this the highest nectar volume was obtained in the morning. On the other hand, the nectar concentration has significant variation in different hours of the day. The highest nectar concentration was obtained at 15hour of the day while the lowest was obtained early in the morning (Table 1) for *P. urens*.

For the *P. urens* the main nectar secretion time was from 6:00-12:00 hours and the plant closed the flower after 12:00 hour under normal condition without shade plant (Figure 2). However, under shade condition the flowers were also left open and provide nectar to the honeybees up to 15:00 hour of the day.



Figure 2: Flowers of *P. urens* with nectar and the closed flowers at the afternoon

The highest nectar volume of *H. auriculata* was obtained at 18 hour while the lowest was at 9 hour (Table 1). Due to the nature this plant (Figure 3), the lowest nectar volume was found early in the morning until 9:00 hours. On the other hand, the highest nectar concentration (33%) was recorded at 15 hour while the lowest was at 6 a.m early in the morning.

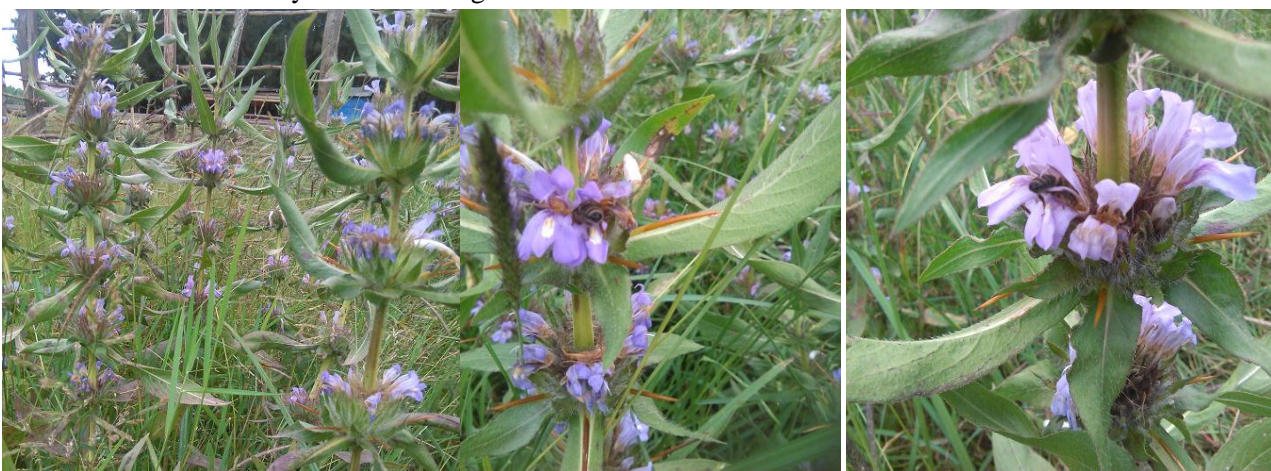


Figure 3: Flowers of *H. auriculata* and when honeybees collect nectar

Nectar volume and concentration of *S. leucantha* were significantly different ( $p < 0.05$ ) in different times of the day. The highest nectar volume (5.5 $\mu$ l) of *S. leucantha* was found at 15 hour whereas the lowest was early in the morning (Table 1). For many plant species, the nectar volume is the highest early in the morning due to high humidity and low temperature of the local area. However, due to the nature of *S. leucantha* and its flower morphology (Figure 4) the utmost nectar volume was obtained at the maximum humidity and temperature values of the study area. On the other hand, the uppermost nectar concentration was obtained from 12-18 hours of the day while the lowest was at 6 hour.



Figure 4: Flowers of *S. leucantha* and when honeybees collect nectar

Table 1: Mean nectar concentration (%) and volume ( $\mu$ l) per flower at 3 hours intervals with  $\pm$  (SE) of the 3 species at 6:00 to 18:00 hours.

	<i>P. urens</i>		<i>H. auriculata</i>		<i>S. leucantha</i>	
Time (h)	Volume $\pm$ SE	Conc. $\pm$ SE	Volume $\pm$ SE	Conc. $\pm$ SE	Volume $\pm$ SE	Conc. $\pm$ SE
6:00	5.79 $\pm$ 0.39 <sup>a</sup>	16.89 $\pm$ 1.1 <sup>d</sup>	1.8 $\pm$ 0.42 <sup>ab</sup>	11 $\pm$ 2.14 <sup>d</sup>	4.1 $\pm$ 0.3 <sup>b</sup>	24.8 $\pm$ 0.46 <sup>c</sup>
9:00	4.49 $\pm$ 0.42 <sup>ab</sup>	22.28 $\pm$ 1.14 <sup>c</sup>	1.5 $\pm$ 0.2 <sup>b</sup>	16.6 $\pm$ 2 <sup>c</sup>	4.3 $\pm$ 0.4 <sup>b</sup>	28.1 $\pm$ 0.45 <sup>b</sup>
12:00	3.18 $\pm$ 0.21 <sup>bc</sup>	29.38 $\pm$ 1.16 <sup>b</sup>	2.2 $\pm$ 0.18 <sup>ab</sup>	27 $\pm$ 1.5 <sup>b</sup>	4.27 $\pm$ 0.4 <sup>b</sup>	30.98 $\pm$ .51 <sup>a</sup>
15:00	2.24 $\pm$ 0.21 <sup>c</sup>	38.63 $\pm$ 0.94 <sup>a</sup>	2.17 $\pm$ 0.24 <sup>ab</sup>	33 $\pm$ 0.7 <sup>a</sup>	5.5 $\pm$ 0.52 <sup>a</sup>	31 $\pm$ 0.70 <sup>a</sup>
18:00	0.00	0.00	2.5 $\pm$ 0.3 <sup>a</sup>	30.1 $\pm$ 1.86 <sup>ab</sup>	4.81 $\pm$ 0.37 <sup>ab</sup>	31.7 $\pm$ 1.00 <sup>a</sup>
Mean	4.3 $\pm$ 0.2	24.43 $\pm$ 0.85	2.0 $\pm$ 0.13	23.5 $\pm$ 1	4.59 $\pm$ 0.18	29.3 $\pm$ 0.35

Note: Treatments with the same letter are not significantly different along column.

#### ***Effect of temperature and humidity on nectar volume and concentration***

Temperature ( $^{\circ}$ C) has direct relationship with the nectar concentration of *P. urens*. This means as temperature increased the values of nectar concentration increased. However, the temperature has less effect on the amount of nectar concentration (Figure 5a). The highest nectar concentration values were secreted between 20 and 30 $^{\circ}$ C for *P. urens*. On the other hand, temperature has indirect relationships with nectar volume. This means as temperature increased the nectar volume decreased and vice versa. However, temperature has no significant effect on secretion of nectar volume for *P. urens*. The highest nectar volume was obtained between 15 and 25 $^{\circ}$ C (Figure 5b).

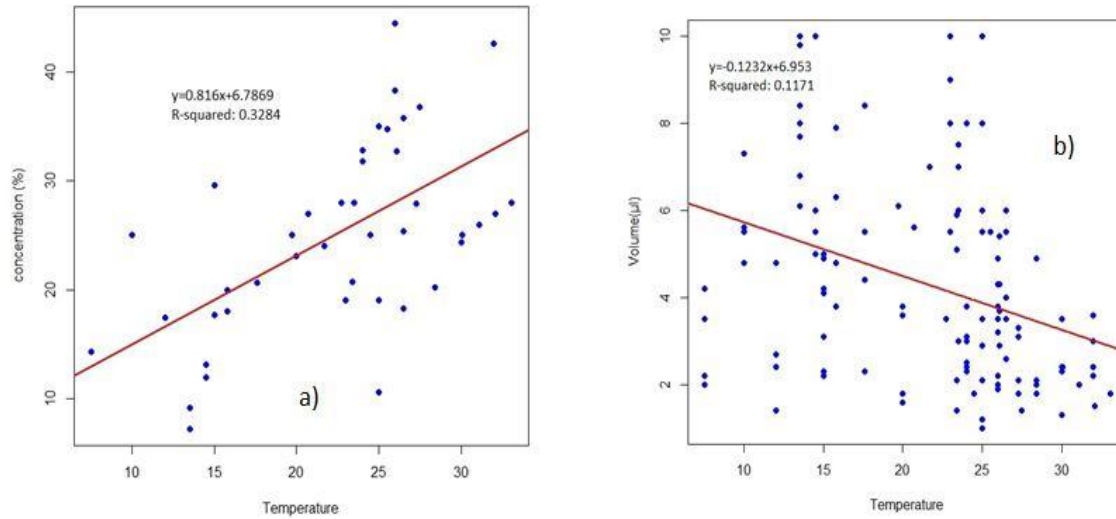


Figure 5: Effect of Temperature on nectar concentration (a) and volume (b) of *P. urens*

Humidity of the study area has direct relationships with nectar volume (Figure 6a). As a result the nectar volume was increased as the air humidity increased. Air humidity influences the amount of nectar volume by 30.94%. The highest values of the nectar volume were found between 40 and 70% of air humidity whereas the lowest values of nectar volume were obtained at <40% of air humidity. Humidity was negatively correlated with the nectar concentration of *P. urens* (Figure 6b). This indicates that at the lower values of air humidity the highest nectar concentration values were obtained and vice versa. Humidity influences the values of nectar concentration by 50.88%.

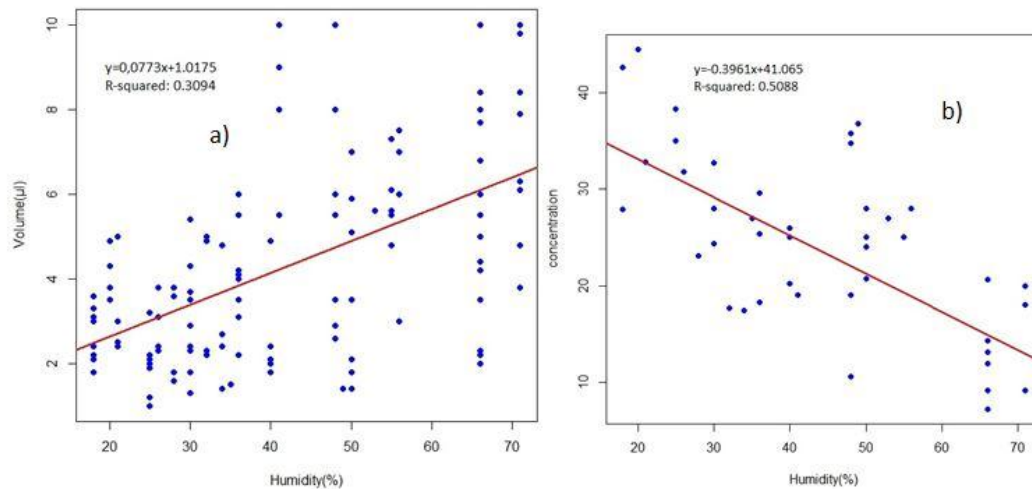


Figure 6: Effect of air humidity on nectar volume and concentration of *Pavonia urens*

Temperature ( $^{\circ}\text{C}$ ) was positively correlated with nectar concentration of *H. auriculata*. However, the effect of temperature on the nectar concentration was insignificant (Figure 7 a). The highest nectar concentration values were secreted between 20 and 25 $^{\circ}\text{C}$  for *H. auriculata*. On the other hand, temperature was negatively correlated with nectar volume. This means as temperature increased the nectar volume decreased and vice versa. However, temperature has no significant effect on the nectar volume of *H. auriculata* (Figure 7b).

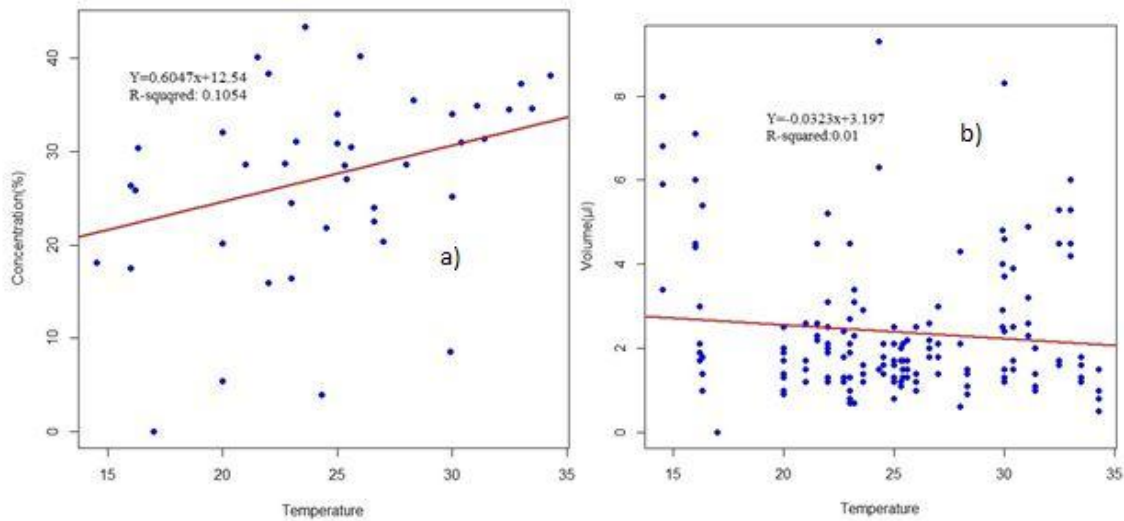


Figure 7: Effect of Temperature on nectar concentration (a) and volume (b) of *Hygrophila auriculata*

Humidity was negatively correlated with the nectar concentration of *H. auriculata* (Figure 8 a). This indicates that at the lower values of humidity the highest nectar concentration values were obtained and vice versa. The influence of humidity on the values of nectar concentration was insignificant. Humidity of the study area has direct relationships with nectar volume (Figure 8b). The highest values of the nectar volume were found between 45 and 70% of humidity whereas the lowest values of nectar volume were obtained at <25% of humidity.

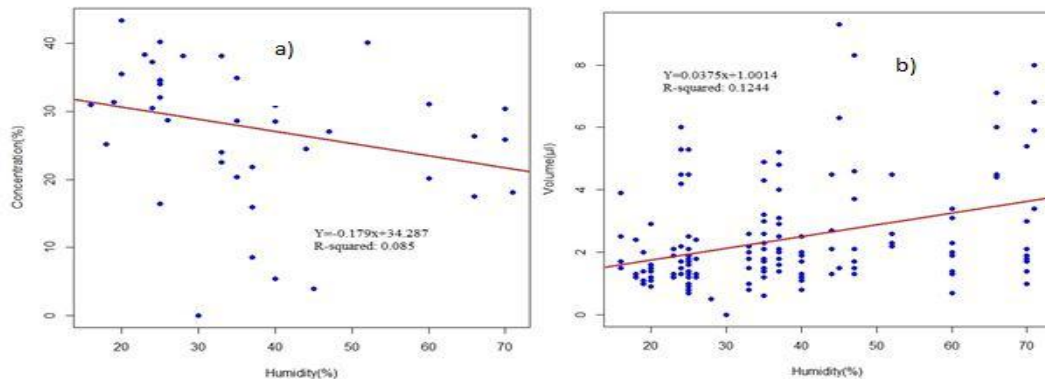


Figure 8: Effect of air humidity on nectar concentration (a) and volume (b) of *Hygrophila auriculata*

Temperature was positively correlated with nectar concentration of *S. leucantha* (Figure 9a). It influences the nectar concentration by 30.67%. The peak nectar concentration was obtained between 25°C and 30°C. The relationships between nectar volume and temperature of the local area were almost found at equilibrium (Figure 9b). This indicates whether the temperature of the local area increased or decreased it has no effect on nectar volume of *S. leucantha*.



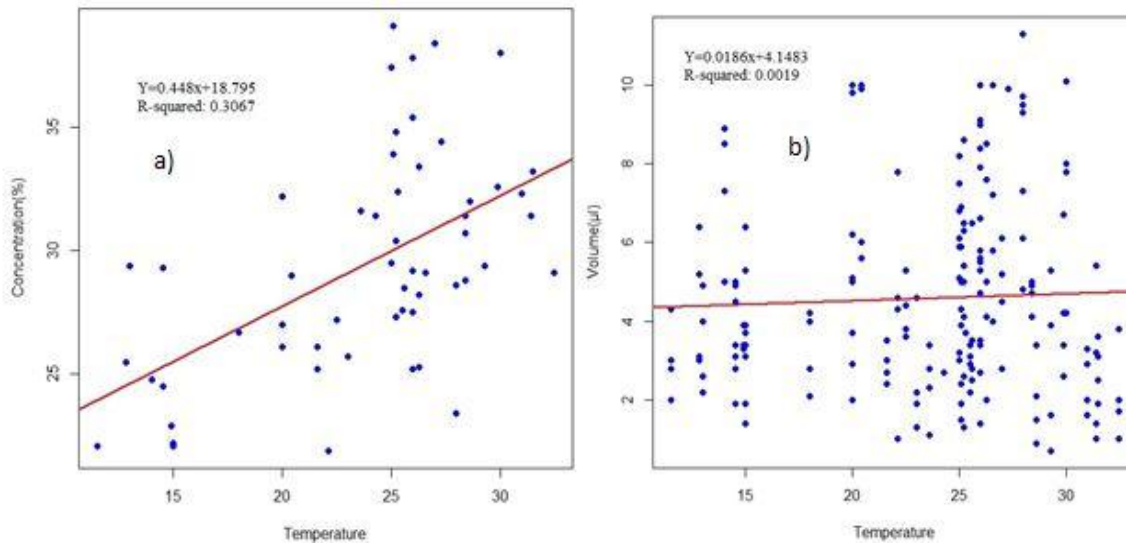


Figure 9: Effect of Temperature on nectar concentration (a) and volume (b) of *S. leucantha*

Humidity of the study area was negatively correlated with nectar concentration of *S. leucantha* (Figure 10a). At the lowest of the humidity the highest nectar concentration values were obtained. The relationship between humidity and nectar volume of *S. leucantha* was almost found at equilibrium (Figure 10b). Hence, whether the values of humidity increased or decreased, it has no effect on nectar volume of *S. leucantha*.

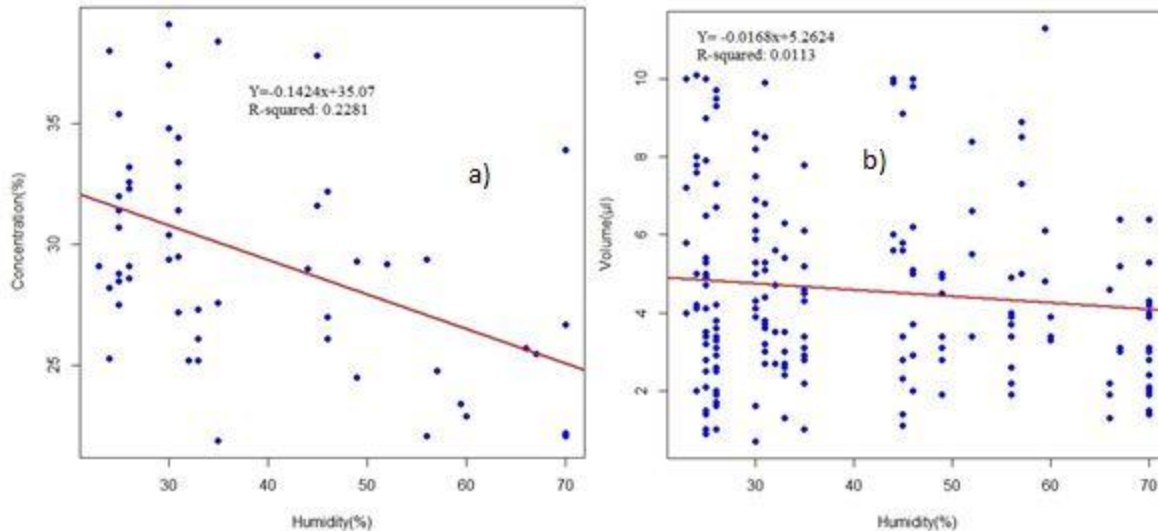


Figure 10: Effect of air humidity on nectar concentration (a) and volume (b) of *S. leucantha*

### ***Estimating amount of sugar and honey production capacity***

The mean number of flowers of *P. urens* per hectare was 3820250 (ranges from 170000 to 11880000 based on the flower branches of the plant) (Figure 1). This was used to estimate the amount of sugar per hectare by multiplying with sugar per flower. The mean sugar per flower was 7.5 mg (ranges from 2.24 to 19.53 mg/flower depending on the age of the flower). The mean sugar amount per hectare was 28.7 kg/ha (ranges from 0.42 to 232 kg/ha) and when converted to honey it was 35 kg/ha (ranges from 0.51 kg to 282.93 kg/ha) of honey per hectare of *Pavonia urens* plants.

The mean number of flowers/ha of *H. auriculata* were 4690000 (ranges from 630000 to 14400000) (Figure 1) and it was used to estimate sugar per ha. The mean sugar per flower was 5.22 mg (ranges from 0.47 to 23.02 mg/flower). Accordingly, the mean sugar per ha was 24.5 kg (ranges from 1.54 to 281.2 kg/ha). When it was converted to honey, the mean amount of honey per ha was 29.88 kg (ranges from 1.88 to 342.9 kg/ha).

The mean number of flowers per ha of *S. leucantha* were 4892000 (ranges from 800,000 to 13500, 000) (Figure 1). The mean sugar per flower was 10.09 mg (ranges from 1.83 to 30.45 mg/flower). The amount of sugar per ha was 49.36 kg (ranges from 1.46 to 411 kg/ha depending on the size of the plants). When the above mentioned amount of sugar converted to honey it was 60.2 kg/ha (ranges from 1.78 to 501.2 kg/ha depending on the size of the plants).

## Discussions

The mean number of flowers per plant was varied due to the variation in size and age of the plants. Similar study was conducted on *Croton macrostachyus* and *Schefflera abyssinica* indicated that the variations between number of flowers per plant could be attributed to the variations in their ecological distribution and climatic factors such as temperature, rainfall and wind (Bareke et al., 2020).

### *Nectar secretion dynamics and effect of weather conditions*

The opening of the flower was early in the morning and completely closed after 15 hour for *P. urens*. It provides nectar for 7hr. at normal condition whereas it continues up to 9 h under shade of the big tree. The nectar secretion duration of *H. auriculata* and *S. leucantha* were the whole day times. However, the nectar secretion of *H. auriculata* was varied depending on the weather condition of the area, during windy period it was started the secretion after 9hr, during normal weather it was started at 6hr. Microclimate determine the chances of changes in nectar volume and concentration; patterns of daily and or seasonal changes in nectar volume and pollinator behavior which include time or activity, frequency, and duration of visits and foraging behavior (Dafni, 1992). Nectar production is differed between the growing seasons due to environmental variables such as air temperature and humidity which can highly affect the nectar secretion and concentration of sugars (Denisow et al., 2018).

The nectar secretion duration is varied from the plant species to species. For *Antigonon leptopus* and *Thevetia peruviana* 6 hr. to 19 hr. (Adjaloo et al., 2015); *Lavandula dentata* and *Lavandula pubescens* 6 hr. to 18 hr. (Adgaba et al., 2015); *Ziziphus spina-christi* 6hr. to 18 hr. (Adgaba et al., 2012) and Pear cultivars 8hr. to 19hr (Farkas & Orosz-Kovács, 2003). Floral durability plays an important role in reproductive ecology, influencing the total number of visits by honeybee and other pollinators as well as the honey production potential of the plants (Adgaba et al., 2015).

Temperature was positively correlated with nectar concentration for all species. However, temperature was negatively correlated with nectar volume of *P. urens* and *H. auriculata* whereas almost at equilibrium for *S. leucantha*. Humidity was negatively correlated with nectar concentration for all species whereas it positively correlated with nectar volume for *P. urens* and *H. auriculata*. However, the relationships between humidity and nectar volume were almost found at equilibrium for *S. leucantha*. This indicates that every plant species has its own optimum humidity and temperature for nectar secretion. Similar study was conducted by Adgaba et al., (2015) on *Lavandula dentata* and *Lavandula pubescens* also showed that

the two species have different optimum humidity and temperature levels for the secretion of maximum nectar.

### ***Honey production potential***

Under natural condition, the amount of honey obtained per hectare of land was 35 kg/ha (ranges from 0.51 kg to 282.93 kg/ha), 29.88 kg (ranges from 1.88 to 342.9 kg/ha) and 60.2 kg/ha (ranges from 1.78 to 501.2 kg/ha) depending on the size of the plants for *P. urens*, *H. auriculata* and *S. leucantha*, respectively. This variation shows the honey production potential of bee plants is varied from plant species to species. This variation occurred due to nature of the plant, habit of the plants as well as weather condition of the study area. In addition to this, many authors have been reported that the honey production potential of bee plants is varied from plant species to species. For example: *Lavandula dentata* and *Lavandula pubescens* provide honey 51 kg/ha and 24.1 kg/ha respectively Adgaba et al. (2015), *Coffea arabica* 125 kg of honey/ha (Bareke et al., 2021b) and *Schefflera abyssinica* 1791 kg honey/ha (Bareke et al., 2020).

### **Conclusions and recommendation**

Based on the dynamics and the amounts of nectar secreted per flower and per plant, for three species can be considered as potential honey source plants for the study area. However, the honey production potential of *S. leucantha* is better than that of two species. In general, the importance of the species is significant not only in terms of serving as sources of honey but also in terms of their ecological values. Therefore, further research on how to propagate and *in-situ* conservation of these species are also recommended for sustainable honey production.

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# Establishing floral calendar in West Hararghe Zone of Oromia Region, Ethiopia

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## Abstract

*Identification of nectar and pollen source and the establishment of flowering calendar are important steps in beekeeping development program. The study was conducted in selected districts of West Hararghe Zone with the objective of characterizing and documenting the major bee forages contributing for honey production and to establish appropriate floral calendar for effective bee management in different agro-ecological condition of the area. Bee forage inventory was made using transect methods in a plot size of 20mx20m, for woody plants and 2mx2m for herbs. Pollen traps having 16% pollen trapping efficiency was fitted at the entrance of beehives for pollen load collection. Honey pollen analysis procedure was also followed to determine the botanical origin of honey.*

*Based on assessment of bee plants and with the respondents are similar with the honey plants identified through pollen analysis collected from the hives by pollen traps. From analysis of respondent interview it was possible to identify two honey-harvesting seasons in the area. The major honey-harvesting season begins in West Hararghe zone from October to December as well as from May to June as major and minor honey flow season respectively. A total of 60 honey bee plants belonging to 29 families were identified and comprising trees, shrubs, herbs, grasses and cultivated crops in the zone. The Pollen analysis of honey showed that *Cordia africana*, *Guizotia scabra*, *Croton macrostachyus* and *Vernonia amygdalina* are the major honeybee source plants. From pollen load analysis and honey pollen analysis, many plant species were flower from October to December and also active season/honey flow season of the zone. Generally, it is recommended, to conserve the identified bee plant species to boost honey production and determination of total carrying capacity of major bee forages in the study area.*

**Keywords:** floral calendar, Bee pollen, bee plants, Honey pollen analysis, West Hararghe

## Introduction

Ethiopia is endowed with natural and cultivated flora and diverse agro-ecological and climatic condition that are well-suited for beekeeping (Fichtl & Admassu, 1994; Admassu et al., 2014). Oromia region is characterized by high plateau and very limited low land areas. The altitude of the region ranges from 900masl at the rift valley to 4377masl at Mt. Tullu Dimtu in Bale Zone. Out of the 58 National Forest Priority Areas of the Country, 49 are found in Oromia. The region has virgin forest of rich biodiversity like Harena, Yayu, Dindin, Anfarara, Munessa, Jibat, Chilimo and Menagesha-Suba which are highly potential for beekeeping. The Region also comprises cultivated crops such as oil and horticultural, and pulses all of which that can augment the beekeeping development further. This makes the region one of the leading regions accounting 55% of the apiculture resources of the country with annual honey production volume of 24.8 thousand tones out of the total 54 thousand tones.

Honeybee plants are those plant species that provide bees with food sources in the form of nectar and pollen or both (Fichtl & Admassu, 1994; Admassu et al., 2014). Not all bee plants are equally important

to bees and honey production (Nuru et al., 2017). Only about 16% of the world's flowering plant species contribute to honeybees as food sources (Crane, 1990).

In order to boost the production of honey from natural resources of the region, identification and documentation of economic bee forages and documentation of economic bee forages and establishing their flowering calendar is critical for the sub-sector development. Though identification of bee forages and establishing floral calendar is not exhaustively done in the region for planning bee management operation, thus establishing floral calendar is a critical tool for planning various beekeeping management operations such as hive super adding and to predict the frequency and period of honey flow in a given area. There are strong associations between the seasonal cycles of honeybee colonies and calendar of bee plants in such way that it will be applied in practical seasonal colony management. Timing of management operations corresponding to phenological pattern of bee plants of the area is critical in building up colony populations before the main nectar flow. Even though bees naturally build up their population during periods when resources are available, the beekeeper must ensure that peak population size attained before or during the nectar flow.

The assessment of bee forages of the zone and its floral calendar are not adequately documented and their correlations with seasonal colony management plan are not established to the required level. Moreover areas with unique production potential are not identified that will contribute to the economies of local beekeepers. Therefore, assessing the availability of bee forage and establishing flowering at the different agro-ecology of West Hararghe Zone for that enable effective seasonal colony management. Therefore this study was conducted to identify, document and prepare flowering calendar of nectar and pollen sources bee forage that can be applied for practical bee management operation in different agro-ecological condition of West Hararghe Zone.

## **Materials and Methods**

### ***2.1 Field Survey***

The study was conducted at Darolabu, Oda Bultum and Gemechis districts of West Hararghe zone comprising low land, midland and highland agroecologies of the area districts. From each district three kebeles were selected and for each kebele four honey bee colonies were established and data like pollen and honey samples were collected from established bee hives. In addition, a total of 81 beekeepers were interviewed from each district with their respective kebele to collect information on status of honey bee production, bee plant availability, and their floral calendar. Moreover Participatory Rural Appraisal (PRA) techniques through focused group discussion were conducted with experts, community groups, development agents and farmer beekeepers were carried out to generate relevant information.

### ***2.2 Bee forage inventory***

For plant inventory each district was classified into three agro-ecologies (Highland, mid land and low lands) and from each agro-ecology three kebeles were selected. Based on this agro ecological stratification, four transect lines were laid out from apiary sites to North, South, West and East within 2 Km radius following GPS. Apiary sites were selected systematically within 2 km distance from one to the other in order to avoid redundancy. Along these transects plots of 20 m x 20 m were laid out within 400 m interval between the sample plots. In order to retain accuracy, five (5) sub plots measuring 2 m x 2 m (4 m<sup>2</sup>) were laid out within the larger plot to capture herbs and grasses. All the plant species encountered

in each sample plots were recorded and percentage cover of each species was estimated visually. For those plant species which could not identified in the field, sample of the specimens were collected using the standard Herbarium techniques and identified at Holeta Bee Research center following the relevant literature and published flora books. Plant inventory was also conducted in all study area and different plant specimens were collected and identified.

### ***2.3 Honey sample collection and Laboratory analysis***

Fresh honey samples of 500gm were collected at different seasons from different agro-ecologies of the districts for laboratory analysis. All samples were kept in sealed glass jars and frozen at -20<sup>0</sup>C until analysis. The pollen analysis was made following the methods adopted by Louvuex *et al.* (1978) for determination of botanical composition and frequency of pollen grains in the sample.

### ***2.4 Colony establishment for pollen collection and seasonal dynamics of honeybee population***

A total of 32 honeybee colonies were established in 8 kebeles of the three districts. For each site 4 honeybee colonies were established (two for pollen trapping and two for honey harvesting). For pollen collection honeybee colonies were fitted with pollen trap having 16% pollen trapping efficiency and pollen loads were collected every seven days interval and frozen in the refrigerator until analysis were made using the prepared reference data base and identified to the generic or species level using the pollen Atlas (Nuru, 2002).

## **Results and Discussions**

### ***3.1 Honey bee plant species and their flowering calendar***

Floral calendar is a time-table that indicates to the beekeeper; the approximate date and duration of the flowering periods of the important nectar and pollen source plants (Diver, 2002). Accordingly the honey bee plants of the study area were composed of trees, shrubs, herbs, grasses, and cultivated crops. Moreover, the species diversity and population density varies widely from area to area. Based on survey result, 60 honeybee plant species belonging from 29 families were identified in the zone. Fabaceae Poaceae, Asteraceae and Myrtaceae Solanaceae and Anacaridaceae were the dominant family comprising higher species diversity in the study area (Fig. 1). The flowering time of common bee flora species in the study area based on response of beekeeper households and key informants indicated that; about 61.04% flowers from Sept- Nov, 19.48% from Dec-Jan, 12.99% from Mar-May and 6.49% from June-August. The identified flowering plants in the study area have been presented in Appendix 1, 2 and 3 for each district. Most of these plant species mentioned by respondents during the survey were similar to those identified through plant inventory and pollen analysis through pollen load collection. This has indicated that all results supported each other and indigenous knowledge of the farmers is dependable. The distribution and type of honeybee plants, as well as their flowering duration, vary from one place to another place due to variation in topography, climate and farming practices. Variation in seasonal availability of honeybee forage species was observed in the zone and the same species have difference in flowering length and season with different agro ecology.

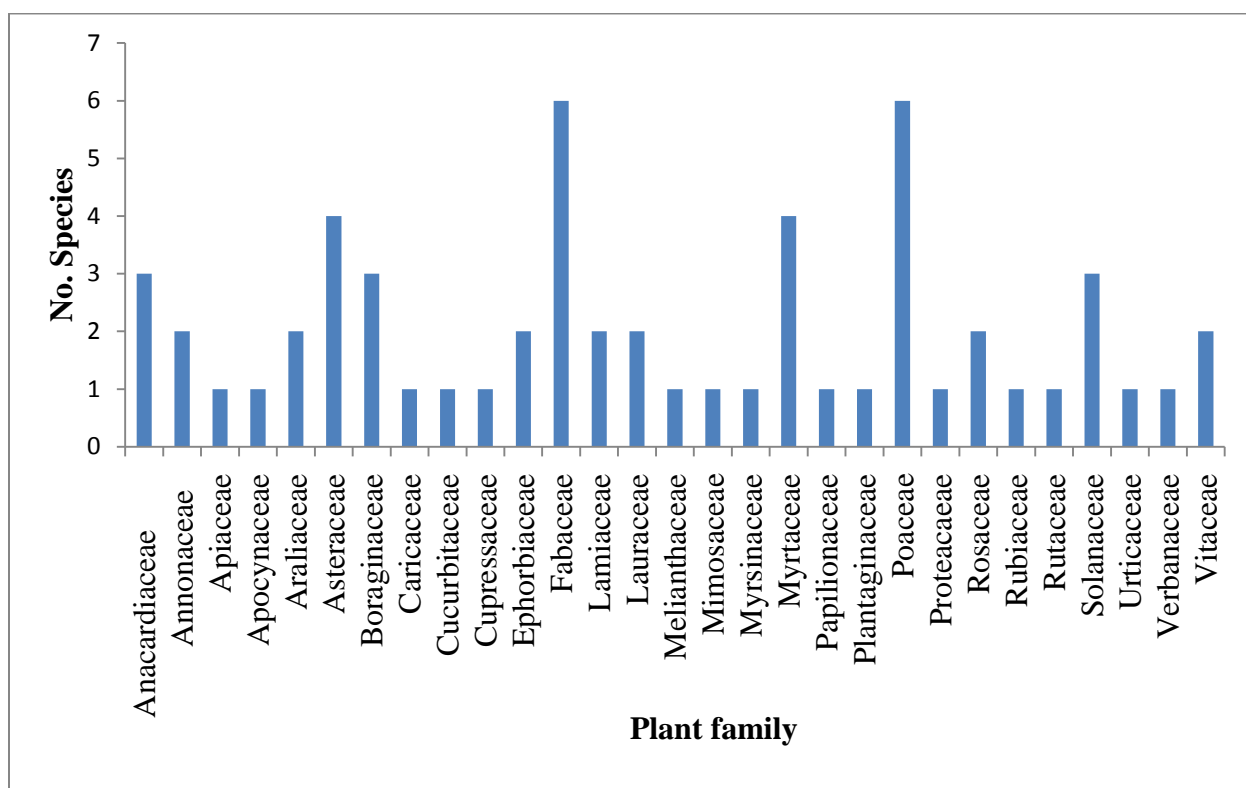


Figure 1 Major Families of honeybee plants in West Hararghe zone

Table 1 Flowering calendar of major bee plants in west Hararghe zone

Botanical name	Family	Food Source	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
<i>Delonix regia</i>	Fabaceae										■	■		
<i>Helichrysum schimperi</i>	Asteraceae											■		
<i>Malus saylvestris</i>	Roseaceae										■	■		
<i>Bersama abyssinica</i>	Melianthaceae	N and P									■			
<i>Carica papaya</i>	Caricaceae			■	■				■	■				
<i>Cissus petiolata</i>	Vitaceae										■	■		
<i>Citrus sinensis</i>	Rutaceae				■	■	■	■	■	■	■	■		
<i>Ehretia cymosa</i>	Boraginaceae			■	■	■	■	■	■	■	■			
<i>Erythrina brucei</i>	Fabaceae							■						
<i>Eucalyptus camaldlensis</i>	Myrtaceae	N and P	■	■	■	■	■	■	■	■	■	■	■	■
<i>Hygenia abyssinica</i>	Rosaceae	N and P						■	■	■	■	■		
<i>Ocimum hamiifolium</i>	Lamiaceae		■	■	■	■	■	■	■	■	■	■	■	■
<i>Pterolobium stellatum</i>	Fabaceae	P and N									■	■		
<i>Trifolium Spp</i>	Fabaceae									■	■	■	■	■
<i>Acacia albida Del.</i>	Fabaceae	N and P	■	■	■									
<i>Acacia etbaica</i>	Fabaceae						■	■	■	■	■	■		





<i>Vernonia amygdalina</i>	Asteraceae	N and P	□	□	□										
<i>Viciafaba</i>	Papilionaceae	N and P													□
<i>Zea mays</i>	Poaceae	P							□	□	□				

### 3.2 Bee flora Species Diversity in Relation to Agro-Ecology

The Shannon diversity indices for the common bee flora species in the study area was calculated (Table 2). Accordingly, bee flora species diversity at Gemachis (highland) (2.16) was relatively lower than both Oda bultum (midland) (2.49) and Daro Lebu (lowland) (2.21). In this study species richness (S) was computed as, the observed number of bee flora species for each agro ecology (Table 2). As a result, the number of species observed in Daro Lebu district was higher in terms of number. The Shannon diversity indices for the common bee flora species in the study area were calculated and there was no significance difference between different sites.

Table. 2. Shannon Diversity Index for Bee Flora Species in West Hararghe Zone

Bee flora species diversity index	Districts		
	Gemachis(highland)	Odabultum(midland)	Daro labu(lowland)
Observed number of species (S)	38	32	40
Shannon diversity (H')	2.16	2.49	2.21
Shannon evenness (E)	0.593	0.718	0.599

Table 3. Identified bee plants species from trapped pollen in West Hararghe zone along different agro ecology

Plant species	Agro ecology	Foraging length											
		S	O	N	D	J	F	M	A	M	J	J	A
<i>Bidens Spp</i>	Highland		X	X									
<i>Rumex 2</i>				X									
<i>Eucalyptus comldens,</i>			X	X	X				X				
<i>Plantago</i>			X	X	X	X							X
<i>Giuzotia Scarba,</i>			X	X	X								
<i>Phyllanthus Reticaltor</i>				X	X	X							
<i>Hypostes Spps</i>					X								
<i>Rumex</i>					X	X							
<i>Hypostes Spps,</i>			X	X									
<i>Giuzotia Scarba</i>			X		X								
<i>Echinops Spp,</i>			X	X									
<i>Phyllanthus</i>			X	X									
<i>Coffe Arbica (Domins),</i>			X										
<i>Vernonia spp.</i>				X	X			X					
<i>Grass Spp,</i>				X	X								
<i>Guizotia Spp</i>					X								
<i>Phonix</i>					X								
<i>Rumex Spp.</i>					X								
<i>Achyranthes aspera</i>								X					X

<i>Terminalia</i>						X								
<i>Justicia spp.</i>							X							
<i>Eucalyptus</i>				X										
<i>Andro Pogon</i>		X												
<i>Coffee</i>		X												
<i>Plantago Lalaca,</i>				X										
<i>Guizotia scalar,</i>			X											
<i>Trifolium spp,</i>			X											
<i>Awxii Xiqaa</i>			X											
<i>Acation Spp,</i>			X											
<i>Maize</i>														X
<i>Saturejapara</i>														X
<i>Androposon abyssinca</i>	X													
<i>Zea maize</i>	Midland											X	X	
<i>Guizatia Spp.</i>			X	X										X
<i>Bidens</i>		X		X										
<i>Eucalyptus</i>				X								X		
<i>Phyllanthus</i>			X									X		
<i>Guizotia Scarbus</i>		X	X											
<i>Andro Pogon abbyssinica,</i>		X												
<i>Reticaltor Spp</i>		X												
<i>Unkown Pollen</i>		X												
<i>Phyllanthus Reticaltor</i>		X												
<i>Grass 2</i>			X											
<i>Plango</i>			X											
<i>Eucalyptus comldens</i>		X												
<i>Achyranthes aspera</i>				X	X									X
<i>Banana</i>														X
<i>Grass spp.</i>														X
<i>Isoglossa laxa</i>												X		
<i>Achyranthes</i>				X								X		
<i>Achyranthes aspera</i>				X	X							X	X	
<i>Juistica cufodonti</i>				X										
<i>Rumex</i>					X									
<i>Parkinsunia acweatu</i>						X								
<i>Guizotia abyssinica</i>														X
<i>Varnonia Spp,</i>					X									
<i>Echinops Spp,</i>					X									
<i>Bidens Spp</i>			X		X									X
<i>Coffe spp.</i>				X										
<i>Scarba</i>				X										
<i>Vernonia</i>						X								
<i>Trifolium spp</i>	Lowland			X	X	X								
<i>Vernonia,</i>				X										
<i>Guizotia spp</i>				X										
<i>Coffe spp</i>				X										
<i>Isoglossa laxa</i>				X										
<i>Vernonia spp.</i>				X	X									

<i>Schinus molle</i>				X								
<i>Brasica</i>			X									
<i>Guizotia scarab</i>		X	X									
<i>Eucalptus</i>				X					X			
<i>Terminalia</i>					X	X						
<i>Grass Spp</i>										X	X	
<i>Andro Pogon abyssinica</i>	X									X	X	
<i>Eucalyptus Spp</i>		X										
<i>Inisica Spp</i>		X										
<i>Trifolium</i>		X										
<i>Plantago</i>			X					X				
<i>Lanceolata</i>			X					X				
<i>Eschinops</i>			X									
<i>Guizotia</i>	X							X				
<i>BidenSpp</i>	X									X	X	
<i>Maize</i>										X		
<i>Eucalptus</i>										X		
<i>Aloa spp</i>										X		
<i>Eucalyptus comldens</i>		X										
<i>Visia Fiba</i>		X										
<i>Andro Pogn</i>		X										
<i>Rumex</i>		X										
<i>Sunflower</i>		X										
<i>Echinops spp</i>			X				X					

### 3.3. Major colony dynamics in the study area

For practical beekeeping application it is very important to identify honey plant flowering seasons in their area in relation to honeybee colony dynamics in order to provide bees with additional feed during the drought period. In study area the major colony dynamics such as pick time of brood rearing, colony swarming, migration, honey flow and dearth period season were identified. Accordingly January to March was the peak time of dearth period in the west Hararghe Zone and also the colony swarming, brood rearing and honey flow season listed by respondents were September, October to November and October to December., From these seasons September to October is the major ones and October to December is the minor honey flow periods of the study area (Fig.1). The variation of the honey colony dynamic due to climatic condition, variation in forage abundance and flowering period of the plants.

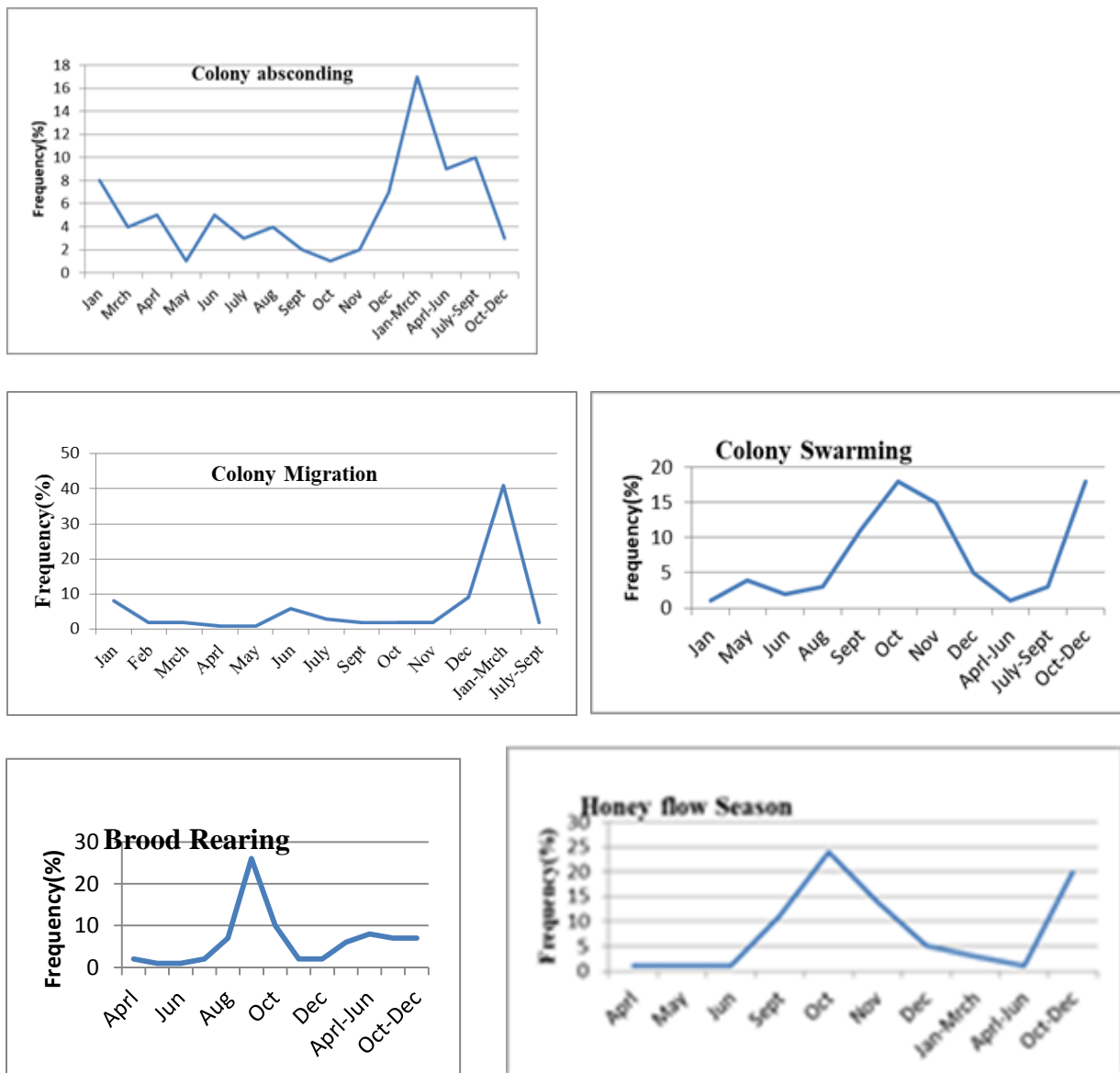


Figure 2 Major colony Dynamics of the area

### 3.4. Extraction of pollen from honey

Mono floral honey is where the bees have been foraging predominantly on one type of plant, and is named according to that plant. As the result of pollen analysis of honey, two types of mono floral honey types were identified in the area and their relative pollen count for species contributing for mono floral honey. The dominance of pollen from the *Guizotia spp* and *Hibiscus spp* can be attributed to widespread distribution in the area and high pollen and nectar potential of the plants.

**Table. 4. Pollen extracted from honey**

Districts	Year	Plant species	No pollen count	Total pollen count	%
Oda Bultum	2012	<i>Eleusine floceifolia</i>	5	40	12.5%
		<i>Guizotia spp</i>	35		87.5%
	2011	<i>Eleusine floceifolia</i>	10	65	5.4%
		<i>Guizotia spp</i>	5		7.7%
		<i>Hibiscus spp</i>	50		76.9%
Gemechis	2011	<i>Apodytes dimidiata</i>	2	47	4.3%
		<i>Guizotia spp</i>	45		95.7%
	2012	<i>Guizotia spp</i>	40	45	88.9%
		<i>Grass spp</i>	5		11.1%
	2011	<i>Guizotia spp</i>	44	45	97.8%
		<i>Grass spp</i>	1		2.2%
Daro Labu	2012	<i>Guizotia spp</i>	40	60	66.7%
		<i>Trifolium spp</i>	20		33.3%
	2011	<i>Cleusinefoi ceifolia</i>	3	41	7.3%
		<i>Guizotia spp</i>	38		92.7%

### Conclusion and Recommendation

Knowledge of honeybee plants, and proper understanding of relationship between seasonal management of honeybees and floral calendar of the plant species is very important to improve the productivity of beekeeping. In West Hararghe Zone experienced beekeepers also familiar with honeybee plants that give good honey, and duration of flowering. Two honey flowering season was identified. Majority of bee forages in the zone flowered from September to November and March to May and very few plant species flowers from August to December. Colony migration, absconding and shortage of bee forage were also seen from January to March in all selected districts of the Zone.

The , herbaceous honeybee forage species were the dominant honey source plants during September to November. However, in March to May majority of honey source plants are trees and shrubs species, Among the identified plant species. *Guizotia sppa*, *Eucalyptus spp*, and *Vernonia spp* are dominate honey source plants in all selected districts s both in social survey and from pollen analysis. Based on the result of the study, beekeepers ; should be awared in line with the flowering calendar, of the area to manage the honeybee colonies to boost honey production .Further study should be conducted on determination of total carrying capacity for potential of flowering plants identified in the study area

## **Acknowledgment**

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# Investigating antioxidant contents of stingless bee (*M. beccarii*) honey from different agro-ecologies of Oromia, Ethiopia

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## Abstract

Stingless bees are form a large group of bees that lack sting apparatus and spread throughout the tropical and subtropical areas of the globe. We analyzed the antioxidant contents of stingless bee honey (*M. beccarii*) collected from Gera, Gomma, Bacho, Didu, Alle, Tokke Kuttaye, Chalia and Wolemera located in different agro-ecologies of Oromia. Antioxidant contents: radical scavenging activity, antioxidant potential assay, total phenolic and flavonoid contents were determined using UV spectrophotometry. The botanical origin and colour of collected samples were tested through pollen analysis and colour grading respectively.. The pollen analysis indicates that among plants contributed to the production of honey, 15.4% of them were categorized under families of Asteraceae and Fabaceae. The mean total phenol, total flavonoid and DPPH of stingless bee honeys was  $995.73 \pm 18.1$  mg GAE / 100 g,  $511.64 \pm 1.38$  mg QE /100 g and  $1317.24 \pm 3.7$  mg/100 g, respectively. Gera stingless bee honey has the highest total phenol content ( $995.73 \pm 18.1$  mg GAE / 100 g). In general the results of this study indicated that stingless bee honey of the study areas contain high level of antioxidants. Thus use of stingless bee honey from these areas is rich in antioxidants.

**Keywords:** Agro-ecologies, Antioxidant, Stingless bee honey

## Introduction

The stingless bees are a vast monophyletic class of highly eusocial bees commonly found in abundance in warm humid forests around the world (Michener, 2007) . Stingless honey like honey bee honey has its own properties: taste, aroma, more fluid texture, and slow crystallization. Due to this particular characteristics stingless bee honey has gained attention in recent years (Biluca et al., 2017).

Stingless honey has resistance to 5-HMF formation even when subjected to elevated temperatures (Biluca et al., 2017). This feature increases the interest of pharmaceutical and food companies as it has no health risks due to excess 5-HMF. Previous study showed that stingless bee honey act as anti-inflammatory, anticancer, antimicrobial and possessed antioxidant properties (Batista et al., 2016). Antioxidant inhibits oxidation; counteract the deterioration of stored foods, removes potential damaging oxidizing agents in a living organism.

Free radicals of oxygen are a natural product of metabolism within the organism; they cause cellular damage and breakdown the structure of DNA (Khalil et al., 2010). Antioxidant components get rid of harmful by-products of normal metabolic functions by inhibiting destructive chemical reactions in our bodies. According to Alzahrani et al. (2012), the antioxidant capacity of honey is due to the phenolic and flavonoids compounds it contains. Antioxidants protect the body from the harmful effect of the free



radicals (Azab et al., 2017) and there is a high correlation between polyphenols and honey antioxidant capacity.

The most traditional practice of using stingless bee honey has great potential as an added value in modern medicine and are considered to have a higher medicinal value than other bee species honey (Rajab, 2018). In Ethiopia, honey produced by stingless bees is considered important in traditional treatments of wound, respiratory ailments, surface infection, diarrheal and various other diseases in line with treatments with other honey (Yalemwork et al., 2013). Stingless bee honey has high market demand, achieving higher prices than the honey produced by other honeybee species.

Rural and urban communities attach high therapeutic value to stingless bee honey. In Ethiopia, recently different studies have been conducted in the areas of indigenous knowledge of stingless bee keeping (Jemberie et al., 2020; Kidane et al., 2021), stingless bee domestication (Alemayehu & Zewdu, 2021), stingless bee honey physicochemical characterization (Alemayehu et al., 2021), antibacterial activity (Ashenafi, 1994).

Although the use of stingless bee honey has been of great importance in Ethiopia, there is little or no information on the characteristics of the antioxidant content of stingless bee honey, especially in potential areas of the Oromia region. In addition, there is not enough research done and information on the antioxidants properties of stingless bee honey of different agro-ecology of Ethiopia. Thus this study was initiated to determine and quantify antioxidants contents, free radical scavenging activity and botanical origin of stingless bee honey collected from different potential areas of Oromia Regional State of Ethiopia.

## Materials & Methods

### *Study area*

This stingless bee honey samples collected from eight representative districts (Gera, Gomma, Bacho, Didu, Alle, Tokke-Kutaye, Chalia and Wolemera) of Jimma, Ilu Aba Bora, West Shoa and Special Zone around Finfinnee zones of Oromia Regional State. These areas were selected purposively based on their potential of stingless bee honey.

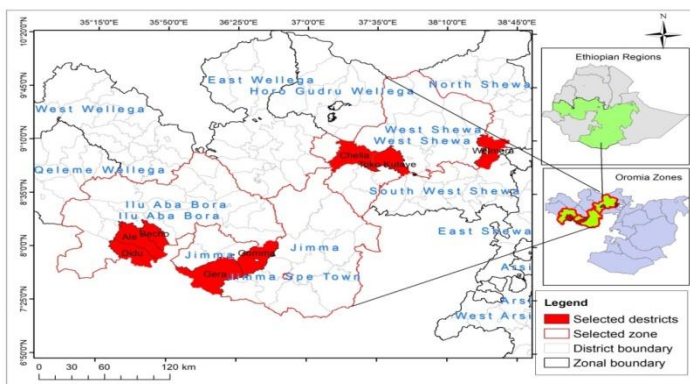


Figure 1. Map showing study areas

### ***Stingless bee honey sample collection***

Forty two stingless bee honey samples were collected from eight districts of four selected zones during 2019 to 2020. Sealed honey pots were selected from cerumen pots from stingless bee nests and fresh and pure honey was harvested using disposable syringes. Honey samples were stored in contaminant-free honey containers (plastic bottles) at about 4 °C in refrigerator till laboratory analyses.



***Figure 2. Stingless honey sample collection***

### ***Pollen analysis***

The botanical origin of honey was identified and quantified using the method of melissopalynology elaborated and published in 2015 (Louveaux et al., 2015). Glycerine jelly, distilled water, centrifuge, vortex, microscope, heating plate, sterile slides, and cover slips were used in the process of analysis. Accordingly, 10 g of honey placed in a 50ml centrifuge tube and dissolved in 20 ml of distilled water. Then, the solvent centrifuged for 10 min at 1000 rpm. After 10 minutes, the supernatant liquid decanted and the remaining solid content transferred to a sterile slide, placed on a heating plate, spread evenly and left to dry for a day. Finally to identify plant spp and quantify the contribution of each plant, glycerol was added to the prepared sample, covered with a cover slip and examined under 40X magnification power of camera fitted computerized microscope.

### ***Honey colour analysis***

Honey samples were placed in clean and clear glass bottles and observed against the colour grading chart and colour intensity was given a rank according to USDA Honey Colour Grading Chart (USDA, 1985).

The relationship between honey colour and other honey parameters reported in this study was explored by conducting Pearson Correlation Tests.

### ***Antioxidant content and activity***

#### ***Determination of total phenolic content (natural compounds)***

The total phenolic content of honey samples were analyzed using folin-ciocalteu reagent based on the method described by Meda et al. (2005) with some modifications. Accordingly, 0.5 ml of honey solution (0.5g/ml) was mixed with 2.5 ml of folin–ciocalteu reagent for 5 minutes. Then, 2 ml of sodium carbonate solution (75 g/l) was added and incubated for 2 hours at room temperature. After this, 0.8 ml of 7.5% sodium carbonate was added and the mixture agitated for 30 minutes in the dark and then centrifuged for 5 minutes at 3300 g. The absorbance of the honey samples and a prepared blank were measured at 765 nm using a spectrophotometer. The concentration of total phenolic compounds in the honey samples was expressed as milligram of gallic acid equivalents (GAE) per 100 g weight of honey using linear equation obtained from the standard gallic acid calibration curve.

#### ***Determination of total flavonoid content***

There are two different spectrophotometric methods in determining the total flavonoid contents. Both measure the formation of colored complex substances quantitatively (Chua et al., 2013). The difference between the two methods is the compound used to react with flavonoid. One uses aluminum ion (III), usually from aluminum chloride ( $\text{AlCl}_3$ ), while the other uses 2, 4-dinitrophenylhydrazine (DNP). However, certain flavonoids such as flavones and flavonols could not react with DNP, thus suggesting that the former method is preferred in determining the total flavonoid content. The total flavonoid content of honey samples was determined based on the method described by (Chua et al., 2013) with some modifications. For each sample, 5 ml of honey solution (0.5 g/ml) was mixed with 5 ml of 2% aluminum chloride ( $\text{AlCl}_3$ ) and incubated for 10 minutes at room temperature. The formation of the Flavonoid-Aluminum complex was measured using spectrophotometry at 415 nm using Uv-vis spectrophotometer. A total concentration was calculated using quercetin standard curve (REF), and expressed as rutin equivalent/100 g of honey.

#### ***Determination of free radical scavenging activity***

The free radical scavenging activity (RSA %) of honey was determined by the 1, 1-diphenyl-2-picrylhydrazyl (DPPH) and it is used in a radical scavenging assay that is based on electron-transfer. this assesses the scavenging potential of a given substance. The assay produces an intense violet solution that is stable at room temperature. When a DPPH solution is mixed with a test compound that is able to donate a hydrogen atom, color of the compound is changed to colorless or light yellow. The free radical scavenging activity of honey was determined by the 1, 1-diphenyl-2-picrylhydrazyl assays described by Chua et al. (2013) with minor modifications. The DPPH solution (20 mg/l) was prepared by dissolving 2 mg of DPPH in 100 ml methanol. A 0.75 ml of methanolic honey solution at different concentrations, ranging from 2.5 to 40 mg/ml, was added to 1.5 ml of DPPH solution. The mixtures were incubated for 15 minutes at 25 °C and the absorbance was measured at 517 nm. DPPH radical scavenging activity of each extract was determined at the various concentrations.

## Statistical analyses

All analyses were performed in triplicates and data were presented as mean standard error. Differences in performance between individual/group of honey samples were analyzed using General Linear Model. Correlation between studied parameters was analyzed by SPSS 20 version statistical software.

## Result and Discussion

### Botanical origin

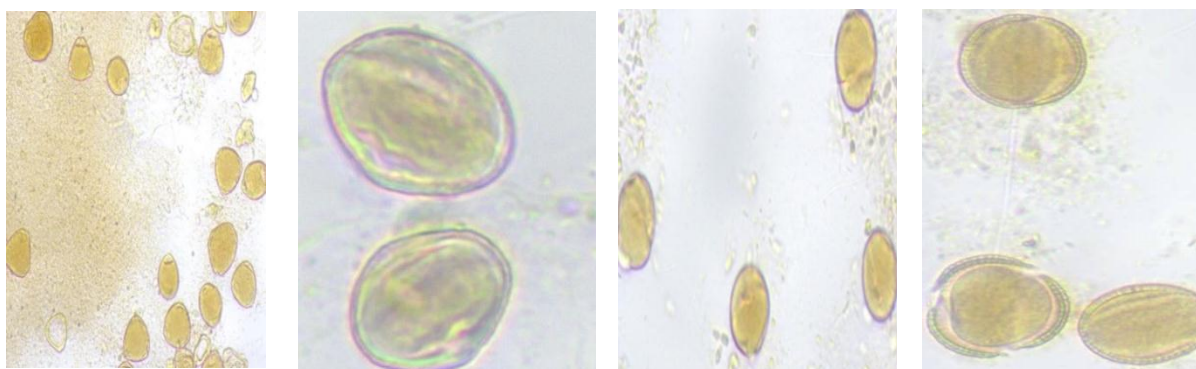
The result of pollen analysis indicated that, the predominant pollen recorded in study samples were *Coffea arabica*, *Eucalyptus globulus*, *Eucalyptus camaldulensis*, *Guizotia scabra*, *Maesa lanceolata*, *Trifolium semipilosum*, *Schefflera abyssinica*. Among these predominant pollen source plant species *Eucalyptus Spps* and *Guizotia spp*s are the predominant pollen source of stingless bee honey collected from five districts. *Eucalyptus spp*s was found predominant pollen source of stingless bee honey in Tokke, Gedo, Wolmera, Becho and Gera, while *Guizotia spp*s was the major source of stingless bee honey in Tokke, Gedo, Wolmera, Didu and Goma. In general these two plant spp are major source of stingless bee honey in West shoa and some parts of southwestern (Ilu Ababor and jimma). On the other hand, *Schefflera abyssinica* was predominant pollen source of singles bee honey obtained from Becho, Didu and Alle of Ilu Ababor. The predominance of coffee was found only in Gomma district.

From the identified pollen sources, 15 % was from families of *Asteraceae* and *Fabaceae* (Figure. 6). The dominant and the diversity of plants identified in this study was in line with the findings of previous study ( Alemayehu et al., 2021)

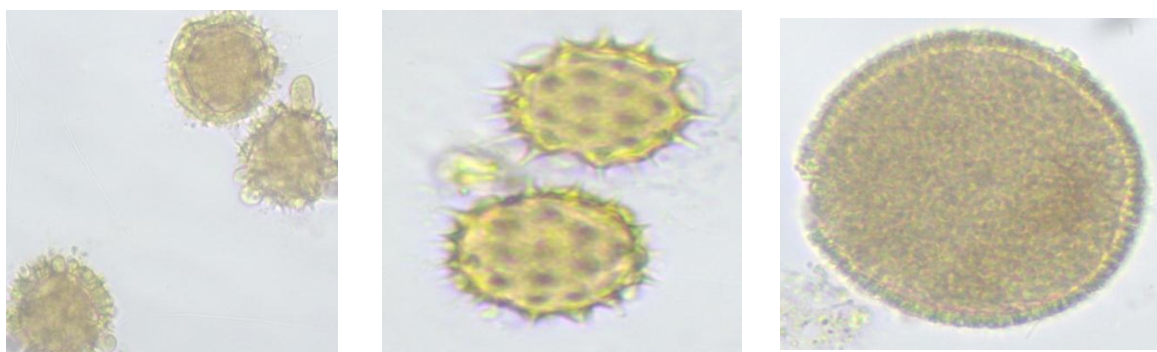
Table 3. The botanical origin of stingless bee (*M. bacarii*) honey from study areas

District	Predominant pollen source (> 45%)	Secondary pollen source (16-45%)	Important minor pollen source (3-15%)	Minor pollen source (< 3%)
<b>Tokkee</b>	<i>Eucalyptus globulus</i> <i>Guizotia scabra</i>		<i>Grass spp</i>	<i>Plantago lanceolata</i>
<b>Gedo</b>	<i>Eucalyptus glubulus</i> , <i>Trifolium semipilosum</i> and <i>Guizotia scabra</i>		<i>Brassica carinata</i>	<i>Coriodrum sativam</i> and <i>Plantago lanceolata</i>
<b>Wolemera</b>	<i>Eucalyptus globules</i> and <i>Guizotia scabra</i>		<i>Acacia abyssinica</i>	<i>Brassica carinata</i>
<b>Alle</b>	<i>Schefflera abyssinica</i>		<i>Coffea Arabica</i> , <i>Croton macrostachys</i> and <i>Vernonia amygdalina</i>	
<b>Didu</b>	<i>Guizotia scabra</i> , <i>Schefflera abyssinica</i> and <i>Maesa lanceolata</i>	<i>Coffea arabica</i>	<i>Eucalyptus camaldulnesis</i> and <i>Vernonia amygdalina</i>	
<b>Bacho</b>	<i>Schefflera abyssinica</i> and <i>Eucalyptus camaldulensis</i>	<i>Maesa lanceolata</i>	<i>Guizotia scabra</i>	<i>Croton macrostachys</i> and <i>Vernonia amygdalina</i>
<b>Gera</b>	<i>Vernonia amygdalina</i> and <i>Eucalyptus camaldulensis</i>	<i>Coffea arabica</i>	<i>Schefflera abyssinica</i> and <i>Croton macrostachys</i>	<i>Verpris dainelli</i>

<b>Goma</b>	<i>Guizotia scabra</i> <i>Coffea Arabica</i>		<i>Vernonia amygdalina</i> and <i>Croton macrostachys</i>
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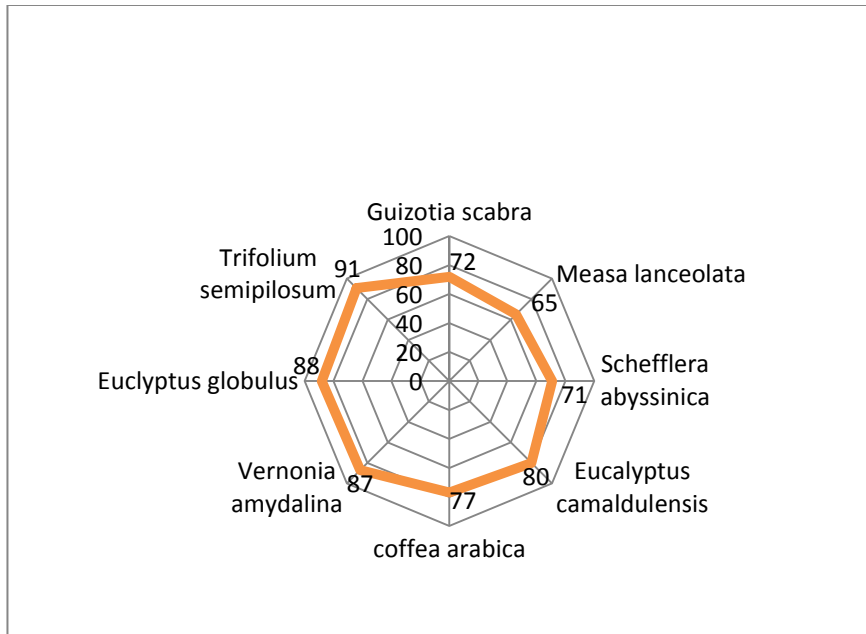
*Eucalyptus globulus*    *Maesa lanceolata*    *Trifolium semipilosum*    *Brassica carinata*



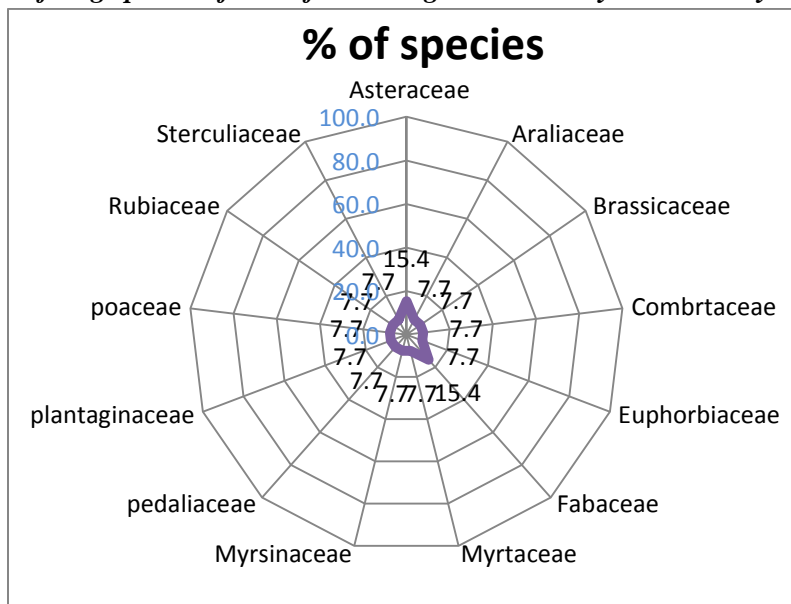
*Croton macrostachys*    *Vernonia amygdalina*    *Guizotia scabra*

Figure 3. Major pollen identified from the stingless bee honey samples

Fifteen pollen source plants species and 13 plants families were identified in the collected stingless bee honey samples. The result of pollen analysis shown that, the predominant pollen types noted may be able to alter the anti-oxidant content(AOC) levels of the stingless bee (*M. bacarii*) honey.



**Figure 4. Major bee forage plants of monofloral stingless bee honeys in the study area**



**Figure 5. Percentages of plant species from different botanical families identified in the samples collected from the different zones of the Oromia region**

**Color of the honey**

The colour of the stingless bee honey collected from study areas was in the range of Extra amber and amber (Table 2). Honey color varies due to many factors including the type of vegetation from which bees forage, soil and associated minerals, age of honey, storage factors and honey processing (Khalil et al., 2010). The present results indicated that the color grade of stingless bee honey range from 4 to 11.5 mm Pfund scale (Table: 2).

Table 4: Colour of the honey samples

Districts	Colour	Color Pfund range, mm
Tokke Kutayye	Extra light amber, light amber and Amber	4.5-8.3
Chaliya	Light amber and Amber	7-11.2
Wolmera	Light amber and Amber	6.8-10
Allee	Extra light amber	4-4.2
Didu	Light amber and Amber	4.4-10
Bacho	Amber	9.8-11.2
Gera	Amber	9.7-10.2
Goma	Amber	11-11.5

### *Antioxidant contents and activities*

#### *The total phenol content (TPC)*

The total phenolic content of the honey samples ranged from 213 to 995mg GAE / 100 g and significantly varied ( $P < 0.05$ ). The stingless bee honey samples from Gera districts had the highest total phenol content ( $995.73 \pm 18.1$  mg GAE / 100 g) with the ranges from of 949.40 to 1042.06 milligram of gallic acid equivalents (GAE) per 100 g and followed by Goma and Becho. On the other hand stingless honey samples from Tokke Kuttaye had the lowest total phenol ( $213.10 \pm 28.5$  mg GAE/ 100g) with the range of 153.25 to 272.94 milligram of gallic acid equivalents (GAE) per 100 g weight of honey content (table 3).

Table 5. Total phenol, total Flavonoid content and radical scavenging activity M. beccari honey

Sampling Districts	TPC mg of Gallic acid equivalents (GAE)/ 100 g of honey	TFC mg quercetin equivalent (QE) /100 g of honey	RSA mg of Ascorbic acid equivalent /100g of honey
Tokke Kutayye	$213.10 \pm 28.5^e$	$290.05 \pm 18.1^{cd}$	$713.02 \pm 74.4^d$
Chaliya	$321.62 \pm 15.1^d$	$287.08 \pm 15.5^{cd}$	$797.26 \pm 28.9^d$
Wolmera	$468.56 \pm 39.3^c$	$419.15 \pm 26.3^{ab}$	$848.15 \pm 76.3^d$
Alle	$473.97 \pm 5.65^c$	$290.24 \pm 28.3^{cd}$	$1112.46 \pm 27.6^{bc}$
Didu	$486.18 \pm 26.8^c$	$331.31 \pm 45.3^{bc}$	$1091.47 \pm 29.4^c$
Bacho	$633.23 \pm 21.9^b$	$191.57 \pm 44.2^{de}$	$1317.24 \pm 3.7^a$
Gera	$995.73 \pm 44.18^a$	$177.96 \pm 1.6^e$	$1303.98 \pm 5.5^{ab}$
Goma	$701.32 \pm 2.16^b$	$511.64 \pm 1.38^a$	$1114.00 \pm 6.2^{bc}$

*Different letters in the column showed significant difference ( $p < 0.05$ )*

*TPC = Total phenol content, TFC = Total Flavonoid content RSA = radical scavenging activity*

The difference in phenolic level arises from botanical and geographical origins of the honey. This concurs with several studies that indicated the phenolic contents in honey are greatly affected by the nectar source. (Biluca et al., 2017; Shamsudin et al., 2019; Silva et al., 2013). Harvest season, weather, and processing also affect the level of phenolic level. TPC level is not significantly different among honey samples collected from Wolmera, Alle, and Didu (Table 2). This may be attributed to similarity in predominant source of the honey (table 2). The TPC level in all our samples is higher than that of the Malaysia

(27.33 and 55.86 mg GAE/100 g) (Shamsudin et al., 2019) and Amazon stingless honey (0.6 mg/100 g)( Bastos *et al*, 2009).

### ***Total Flavonoid Content (TFC)***

There was statistical significant difference in level of total flavonoid among samples of singles bee honey ( $P < 0.05$ ). Stingless bee honey from Wolmera has the highest flavonoid ( $511.64 \pm 1.38$  mg quercetin equivalent (QE) /100 g), while the lowest was recorded in stingless bee honey from Gera ( $177.96 \pm 19.6$  mg quercetin equivalent (QE) /100 g) (table 3). This variation in the total flavonoid content may be due to high diversity of floral and agro ecologies difference of the study areas. Level of flavonoid in all our samples much higher than Tanzania ( $44.82$  mgRE/100g) (Muruke, 2014).

### ***The Antioxidant activity***

The results indicated that the levels of DPPH or scavenging activity range from 713.02 to 1317.72 mg/100g (Table 3) and it is significantly varied ( $P < 0.05$ ). The value of DPPH, in honey from Bacho exhibited the highest DPPH with ranges of 1196.12-1438.38 and lowest is found in honey samples from Toke Kutayye, cheliya and Alle. However the overall DPPH level of stingless honey from Oromia is higher than that of stingless bee honey from Tanzania ( $4.19$  mg/ml) (Tuksitha et al., 2018)

### ***Correlation***

There was significant positive correlation between the total phenol content (TPC) and the antioxidant activity ( $r = 0.675$ ,  $p = 0.01$ ) of stingless bee honey, whereas there was low negative correlation between antioxidant activity and total flavonoid content (TFC) ( $r = -0.255$ ,  $p = 0.05$ ). The antioxidant capacity of honey is the result of the combined activity of a wide range of compounds such as Maillard reaction products, organic acids, amino acids, protein and ascorbic acid in honey. Antioxidant properties contributed to the antioxidant activity via synergistic interaction of phenolic and flavonoid compounds in honey (Alvarez-Suarez et al., 2010). Colors also have low negative correlation ( $r = -0.278$ ,  $p = 0.05$ ) with TPC. This indicates that the substances which give colour to honey (phenolic, minerals, 5-HMF and products of Maillard reaction) influence the antioxidant activity. However, there was no significant correlation happened between the TFC ( $r = -0.092$ ) and the colour of the honey, indicating that the antioxidant activity is not solely dependent on flavonoids content. This means the colour is not influenced by the TFC in Oromia stingless bee honey; and the same result reported by (Shamsudin et al., 2019).

### **Conclusion and Recommendations**

Recently there is grown interest in the use of natural antioxidants as a form of protection against oxidative damage in the human body. The consummation of stingless bee honey increases antioxidant defenses against cancer-causing agents. However contents of stingless bee honey related to health issues has not been studied in Ethiopia. The present study for the first time indicates that stingless bee honey produced in Ethiopia in general and Oromia in particular is rich in antioxidants compared to that of Tanzania and Brazil. Thus stingless bee honey of Ethiopia has high medicinal value. *Eucalyptus* Spps., *Guizotia* spp and *Schefflera abyssinica*, were the major source of stingless bee honey in the study areas. Expanding Stingless bee keeping in the country is very important to utilize the product as medicine and income generation. Furthermore, further study is required to test the medicinal value of singles bee honey in



other parts of Ethiopia and set appropriate dosage to exploit the medicinal value of the honey in human health,

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## Appendices

### Appendix 1. Honeybee flora of Oda Bultum district

Botanical name	Local name	Family	Food Source	Flowering month
<i>Carissa edulis</i>		Apocynaceae		April
<i>Bersama abyssinica</i>	Asaamiroo	Melanthaceae	N and P	June, april, march
<i>Persea Americana</i>	Avokaadoo	Lauraceae		March-april
<i>Viciafaba</i>	Baaqela	Papilionaceae	N and P	Sept
<i>Croton macrostachyus</i>	Bakkanisa	Ephorbiaceae	N and P	Sept, april, aug
<i>Datura stramonium</i>	Banjii	Solanaceae	N and P	Sept-oct
<i>Capsicum annum</i>	Barbare	Solanaceae	N and P	July, aug,
<i>Eucalyptus camaldlensis</i>	Bargamo	Myrtaceae	N and P	Dec-jan
<i>Salviani lotica</i>	Bassobila	Lamiaceae		April
<i>Zea mays</i>	Boqollo	Poaceae	P	June, July
<i>Coffea Arabica</i>	Buna	Rubiaceae	N	March, April
<i>Citrus sinensis</i>	Burtukana	Rutaceae		March-april
<i>Cynodon dactyl</i>	Citaa	Poaceae	P	Sept-oct
<i>Agrocharis Sp</i>	Cogoge	Apiaceae		Sept
<i>Vernonia amygdalina</i>	Ebicha	Asteraceae	N and P	Jan , feb, march
<i>Acacia albida Del.</i>	Garbi	Fabaceae	N and P	Dec-march
<i>Schefflera abyssinica</i>	Gatame	Araliaceae	P an N	Jan, feb
<i>Annona senegalensis</i>	Giishxaa	Annonaceae		April
????	Giraavillaa			All year round
<i>Rosa abyssinica</i>	Gora	Rosaceae	P	Sept, oct
<i>Guizotia scabra</i>	Hada	Asteraceae	P an N	Sept, oct, nov
<i>Entada abyssinica</i>	Halanqabesa			April ,dec feb, march
<i>Commelina benghalensis</i>	Laaluu	Commelinaceae	P	April
<i>Mangifera indica</i>	Mango	Anacardiaceae	P an N	March ,april
<i>Boswellia papyrifera</i>	Muka Ixaanaa	<i>Burseraceae</i>	P an N	Sept-nov
<i>Guizotia abyssiniea</i>	Nugi	Asteraceae	N and P	Sept,, oct
<i>Cicer arietinum L.</i>	Shumbura	<i>Fabaceae</i>	N and P	Oct-nov
<i>Lycopersicon esculentum</i>	Timaatima	Solanaceae	N and P	April-aug
<i>Ehretia cymosa</i>	Ulaagaa	Boraginaceae	N and P	June-july
<i>Cordia Africana</i>	Wadessa	Boraginaceae	P an N	Sept, july, aug, apr
<i>Allophylus rubifolius</i>	Xaaxessa	Sapindaceae		March
<i>Psidium guajava</i>	Zaytuna	Myrtaceae		April, may

### Appendix 2. honeybee flora of Gemechis District

<b>Botanical name</b>	<b>Family</b>	<b>food source</b>	<b>Flowering period</b>
<i>Carissa edulis</i>	Apocynaceae	P an N	Sept-oct
<i>Malus saylvestris</i>	Roseaceae		sept, oct
<i>Bersama abyssinica</i>	Melanthaceae	N and P	Sept
<i>Persea americana</i>	Lauraceae	P an N	Jan
<i>Croton macrostachyus</i>	Ephorbiaceae	N and P	Aug-sept
<i>Viciafaba</i>	Papilionaceae		Sept
<i>Eucalyptus camaldlensis</i>	Myrtaceae	N and P	Year round
<i>Podocarpus falcatus</i>	Podocarpaceae		April
<i>Zea mays</i>	Poaceae	P	July, aug
<i>Coffea Arabica</i>	Rubiaceae	N and P	April
<i>Cynodon dactyl</i>	Poaceae	P	Sept-oct
<i>Justica schimperiana</i>	Acanthaceae		Sept-oct
<i>Salanum tuberosum</i>	Solanaceae		July-aug
<i>Cucurbita pepo</i>	<i>Cucurbita pepo</i>		Oct
<i>Vernonia amygdalina</i>	Asteraceae	N and P	March
<i>Juniperus procera</i>	Cupressaceae		Oct
<i>Annona senegalensis</i>	Annonaceae		
<i>Rosa abyssinica</i>	Rosaceae	P	jan, feb,sept, oct
<i>Guizotiascabra</i>	Asteraceae		sept, oct, nov
<i>Phytolacca dodecandra</i>	Phytolacaceae	N and P	nov-feb
<i>Embelia schimperii</i>	Myrsinaceae		dec, jan, feb
<i>Catha edulis</i>	Celastraceae	N and P	Jul
<i>Dovyalis caffra</i>	Flacourtiaceae	P and N	jan-mar
<i>Sorghum bicolor</i>	Poaceae	P	
<i>Trifolium spp</i>	Fabaceae	P and N	sept, oct
<i>Carthamus tinctorius L</i>	Asteraceae	P and N	sept, oct
<i>Lantana camara</i>	Verbanaceae		jan, feb
<i>Cordia Africana</i>	Boraginaceae	P and N	,,,,
<i>Erythrina brucei</i>	Fabaceae	P and N	
<i>Rhus spp</i>	Anacardiaceae		sept, oct
<i>Psidium guajava</i>	Myrtaceae		sept, oct

### Appendix 3. honeybee flora of Daro Lebu district

<b>Scientific Name</b>	<b>Family Name</b>	<b>Food Source</b>	<b>Flowering month</b>
<i>Perseaamericana</i>	Lauraceae	N and P	oct
<i>Croton Macrostachyus Huchst.Ex Del</i>	Ephorbiaceae	N and P	, mar,,jun
<i>Helichrysum schimperii</i> (Sch.Bip.ex	Asteraceae		oct

A.Rich.) Moeser			
<i>Eucalyptus camaldlensis</i>	Myrtaceae	N and P	all the year
<i>Zea mays</i>	Poaceae	P	aug, sept
<i>Coffea Arabica</i>	Rubiaceae	N and P	jan, feb,
<i>Lagenaria abyssinica (Hook.F.) C. Jeffrey</i>	Cucurbitaceae	P and N	sept, oct, nov, may
<i>Citrus sinensis</i>	Rutaceae		oct,aug, march, apr, may
<i>Caesalpinia decapetala</i>	<i>Caesalpinioideae</i>		
<i>Agrocharis Sp</i>	Apiaceae		Sept,oct, jun, jul, aug,may
<i>Delonix regia</i>	<i>Fabaceae</i>		sept, ct
<i>Acacia etbaica</i>	Fabaceae		jun,sept, oct, may,
<i>Vernonia amygdalina</i>	Asteraceae	N and P	jan, feb,
<i>Acacia albida Del</i>	<i>Fabaceae</i>	N and P	aug
<i>Annona senegalensis</i>	Annonaceae		
<i>Entada abyssinica</i>			jan
<i>Embelia schimperii</i>	Myrsinaceae		May
<i>Pterolobium stellatum</i>	Fabaceae	P and N	Sept, oct,
<i>Hygenia abyssinica</i>	Rosaceae	N and P	jul, aug, oct
<i>Ocimum hamiifolium</i>	Lamiaceae		All the year
<i>Prunus persica</i>	Rosaceae	P and N	
<i>Acacia Sp.</i>	Mimosaceae		jan,
<i>Sorghum bicolor</i>	Poaceae	P	sept,aug oct, nov
<i>Cissus petiolata</i>	Vitaceae		sept,oct
<i>Carica papaya</i>	Caricaceae		feb,march, jul,aug
<i>Trifolium Spp</i>	Fabaceae		aug, sept, oct, nov, dec
<i>Lantana camara</i>	Verbanaceae		sept, oct, jul aug,march, apr
<i>Lycopersicon esculentum</i>	Solanaceae		march, apr
<i>Ehretia cymosa</i>	Boraginaceae		feb,march, jun, apr, sept, may

# Stingless bee species diversity and ecology in Oromia

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## Abstract

*Stingless bees play a key role in natural resource conservation and enhancing human economy because of their pollination services and high value products respectively. However, information on their nesting habitats, nest architectures and nest bionomics are scarce almost for all Ethiopian stingless bee species. To gain an insight into nest ecology and biology of stingless bees in the country, different habitats from 11 districts were assessed and general nest properties, such as entrance dimension, shape and entrance tunnel length, nest size, nesting substrate and internal nest architecture were characterized. A total of 49 natural nests of two stingless bee species belonging to two genera were found: *Meliponula* and *Liotrigona*. 47 nests of *Meliponula baccarii* (*M. baccarii*) found under the ground at the average depth of  $50.50 \pm 9.32$  cm in three different soil types, while the two nests of *Liotrigona bottegoi* (*L. bottegoi*) was found in the cavities of *Albizia shimperiana* trunks at the heights of 2.8 and 3.0 m above the ground. Although there was no an exposed nest under natural condition for both species, the nest architecture and nest biology were highly varying between the species but similar within the species. Although the basic nest architectures and bionomics were similar in the analyzed parameters within the species, the natural nests were quantitatively varied. The present results can be used for the management and conservation of the species. Finally, the data on nesting substrates, nest architectures and nest biology are more helpful for future designing of suitable bee hives for keeping and conservation of both species.*

**Keywords:** *stingless bee, nesting habitat, nest architecture, nest biology, nesting substrate*

## Introduction

Stingless bees which belongs to the family Apidae and sub family Meliponinae are a group of eusocial small to average sized honey producing bees with vestigial stings. About 56 genera of stingless bees containing over 600 species are found in most of the tropical and subtropical regions of the globe (Cortopassi-Laurino et al., 2006; Eardley, 2004; Roubik, 1979). In Africa, only about 20 stingless bee species have been described (Eardley, 2004). In Ethiopia, stingless bees are represented by about five species (Pauly and Zewdu, 2013) and some of them have a high demand for their honeys in traditional medicine and all of them playing a major role in pollinating natural plants and cultivated crop although, their contribution is not noticed yet.

The diversity of stingless bees varies among different ecologies. The most diverse habitat is natural forests, followed by secondary and utilized forests, and farm lands (Njoya et al., 2018; Suriawanto et al., 2017). In general, lower altitude environments are more diverse in stingless species than higher altitudes (Salim et al., 2012). Although stingless bees are considered generalist in nest site selection, they are still exhibiting a degree of plasticity in their nesting habitats and nesting substrates (Njoya et al., 2019). The majority of the species construct their nests in abandoned nests of other social insects, such as ants and termite nests (Roubik, 1980). For instance, stingless bee species, such as *M. baccarii* (Gribodo,

1879) is known as forest species nesting in underground cavities and *Meliponula bocandei* (Spinola, 1853) is also known as forest species nesting in underground cavities or in the tree trunk cavities (Njoya et al., 2018). Many other species solely nest in the cavity of tree trunks or in underground cavities.

Stingless bee species inhabiting tropical and subtropical regions of the world considerably differ in colony size, body size and colour (Michener, 2007). Typically, perennial colonies of stingless bees consist of a few dozens to more than 20,000 workers, single fertile queen and drones (Alves et al., 2019; Danaraddi et al., 1996; Njoya et al., 2018; Viana et al., 2015). Stingless bees also vary significantly in their nest architecture with different characteristics of nest entrances and diverse designs in brood cells arrangements (Njoya et al., 2019). Brood cells are arranged either in horizontal or vertical cells with full combs or semi-combs or in clustered cells (Kajobe, 2007). The nest entrance of stingless bees also varied in shape, length and colour, which can be used for taxonomic studies of stingless bee species with other nest characteristics (Njoya et al., 2019; Suriawanto et al., 2017). It has also been confirmed that stingless bees nest characteristics are useful in taxonomic studies particularly in tropical African countries (Kajobe and Roubik, 2006) like in Ethiopia, where a few have been studied. In Ethiopia, some studies like indigenous knowledge of stingless bee keeping (Wondmeneh et al., 2020; Amenay et al., 2021), stingless bee domestication (Alemayehu and Zewdu, 2021), stingless bee honey physicochemical characterization (Alemayehu et al., 2021), antibacterial activity (Mogessie, 1994), and antioxidant activity of stingless bee honey (Meseret et al, unpublished data) have been carried out.

However, nesting habitats, nest biology and nest architectures of stingless bee species have not been so far well described in Ethiopia in general and in Oromia in particular. Knowledge about ecology and nest biology of stingless bees is vital for underpinning their adaptation and helps for the development and implementation of proper conservation strategies of the species (Siqueira et al., 2012; Viana et al., 2015). Information on ecology and nest biology of stingless bees is also important to develop and improve management practices in order to exploit the stingless bees both for pollination and production of stingless bee products (Danaraddi et al., 1996). Hence, investigating the nesting habitats and nest biology of native stingless bee species in Ethiopia where little studies have been conducted is more vital to develop proper stingless bee management practices and conservation. As a result, it is felt valuable to undertake a comprehensive investigation on stingless bees' nesting ecology and nest biology. Therefore, the aim of this work was to contribute inclusive information on nesting ecology, nest architecture and nest biology of the stingless bees in Oromia Regional State of Ethiopia through systematic investigation of wild stingless bee colonies.

## **Materials and methods**

The study was conducted in six different Zones of Oromia Regional State (Oromia Special Zone Around Finfine, West Shewa, Buno Bedele, Ilubabor, Jimma and East Guji) (Figure 1). The zones, districts and peasant associations were purposively selected as sampling areas based on the presence of local stingless bee honey marketing and potentiality for the diverse wild stingless bee nests. The sampling areas include different nesting habitats, such as natural vegetation, plantations, protected forests, woody shrubs along crop fields, grass lands and even crop fields, as the bees can use different habitats for their nesting. Nesting sites and natural stingless bee nests were found mainly based on information provided by local stingless bee hunters, farmers and beekeepers of the respective peasant associations. Once the nests located, the coordinates for the locations were obtained using GPS (Global Positioning System). Then, nesting habitat type, soil type (for

subterranean nesting stingless bees), tree species, position on the tree and height of the entrance above the ground (for tree trunk cavity nesting bees), and detailed external entrance characteristics were carefully recorded. The surroundings of each nest were cleaned and their entrance height above the ground and entrance tube diameter were measured using digital caliper with the precision of 0.01 mm. For the subterranean nests, nest excavation was done following our previous method (Alemayehu and Zewdu, 2021). The lateral part of each nest was excavated to expose the nest. The dimension (width and height), length of the entrance tunnel above the nest to the ground level were measured using a measuring tape. External entrance height above the ground, diameter of the entrance and thickness of the tube were also measured using digital caliper.

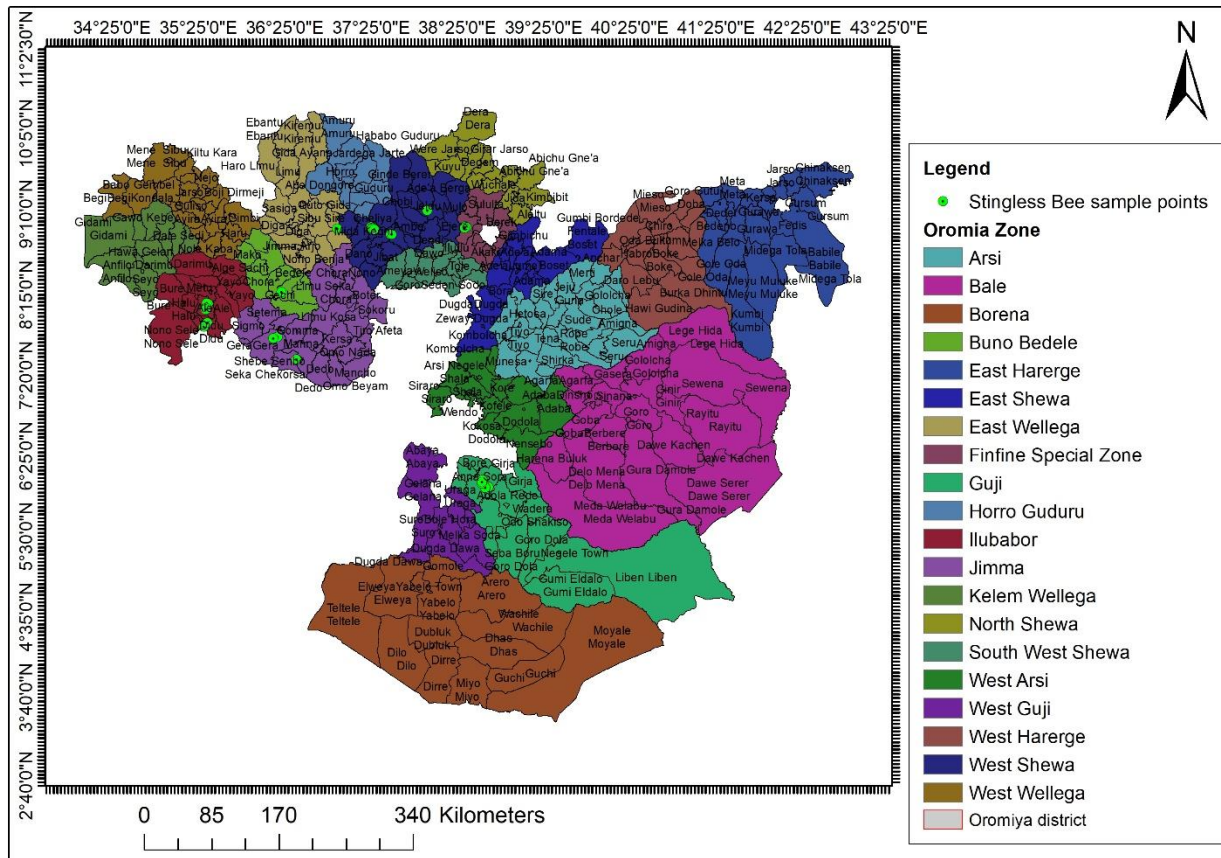


Figure 1. Map of Oromia showing the 11 sampling districts in the six zones.

After opening layers of a nest involucrum, number of brood combs per nest, number of queen cells per nest, number of cells per 4 cm<sup>2</sup> (an area of 20 mm x 20 mm) were quantified, and the presence or the absence of queen bee was visually checked by separating brood combs (cluster of brood cells). The shape of brood combs and storage pots, their arrangements and color, as well as their positions in the nest were carefully observed and recorded especially for the ground nesting one. The diameter of brood combs, comb thickness, pillar height, pillar thickness, worker cell depth and width (by measuring width of 10 cells dividing the result by 10) were measured for some colonies. This close examination of colonies was made only for some colonies, as the method is a destructive.

Finally, stingless bees from nests were taken and killed by drowning them in soapy water in plastic bottle. Dead bee specimens were placed in a labeled plastic sampling bottles containing 70% ethanol and brought to



Holeta Bee Research laboratory for taxonomic study purpose. Samples were identified using morphological features, such as colour, body size, scutle surface and head setae using key described (Pauly and Zewdu, 2013) and compared with reference specimens at Holeta Bee Research Center.

### ***Data Analysis***

All data on altitude, latitude, longitude, nesting habitat type, nesting substrate, nest architecture, and internal parameters measured were entered into Microsoft excel. Frequency analysis was used to obtain the proportion of nests across habitat type. Descriptive analysis was run to characterize internal and external nest sizes. Moreover, Pearson correlation was computed to determine if there is a significant correlation between number of brood combs and number of queen cells counted per nest at  $\alpha = 0.05$  level of significance using IBM SPSS Statistics version 20.

## **Results**

### ***Study area and nesting habitats***

Stingless bee nests were sampled from 11 districts of six different Zones of Oromia Regional State, Ethiopia (Figure 1). The altitudes in which the nests were found ranged from 1758 to 2543 meter above sea level (m.a.s.l) with an average of  $2256.56 \pm 218.73$ m.a.s.l(N = 48). During the survey, 49 nests of Meliponini bees were found in the study areas, belonging to two genera: (*Meliponula* and *Liotrigona*). The genera *Meliponula* and *Liotrigona* were each represented by one species: *M.baccarii* and *L. bottegoi*. These bee species occupied two distinct nesting substrates. Forty seven 47 nests of *M. baccarii* found nesting in the ground in three different soil types, while the two nests of *L. bottegoi* were found in the cavity of *Albizia shimperiana* trunk at the heights of 2.8 and 3.0 m above the ground (Table 1, Figure 2A). *Meliponula baccarii* was the species most commonly found in all sampled sites and covering the entire range of nesting habitats, while the two nests of *L. bottegoi* found in the cavity of the tree trunks in the natural coffee forest. However, the preference for the nesting habitat of *M. baccarii* was greatly varied among the six nesting habitats. The highest number of *M. baccarii* was obtained from cultivated lands adjoining forest areas (17 nests), followed by protected forest (13 nest), and grass land (5 nests) and woody shrubs along field side (5 nests) (Figure 3).

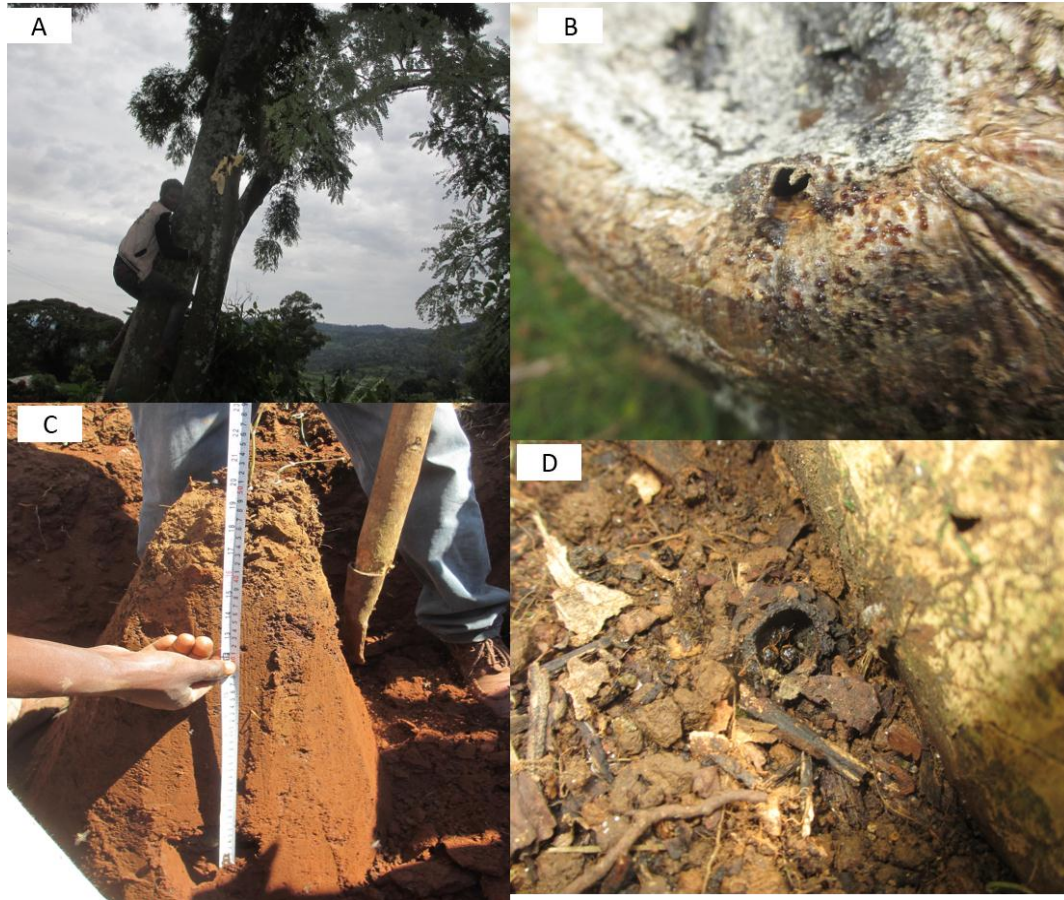


Figure 2. Stingsless bee nests in the tree trunk and under the ground

Nest of *L. bottegoii* in the trunk of *Albizia shimperiana* at the heights of 3.0 m above the ground (A), nest entrance of *L. bottegoii* with its sticky resin around the entrance (B), nest of *M. baccarii* at the depth of about 53.0 cm below the ground (C), and external entrance of *M. baccarii* with guard bees (D).

Table 1. Localities of *M.baccarii* and *L. bottegoii* in the six Zones of Oromia Regional State (altitude, nesting habitat type, nesting substrate used and entrance tube shape).

Nest code	Zone	District	Altitude	Habitat type	Nesting substrate (Soil type/Tree spp.)	Entrance tunnel shape
1	OSZAF	Walmara	2397	1	Red	Round
2	OSZAF	Walmara	2397	1	Red	Round
3	OSZAF	Walmara	2392	2	Red	Round
4	West Shewa	Jeldu	2400	3	Red	Round
5	West Shewa	Chaliya	2269	3	Red	Round
6	OSZAF	Walmara	2397	2	Red	Round
7	West Shewa	Chaliya	2282	2	Red	Round
8	West Shewa	Chaliya	2280	4	Red	Round
9	West Shewa	Chaliya	2353	4	Red	Round
10	West Shewa	Toke Kutaye	2506	3	Red	Round

11	West Shewa	Toke Kutaye	2543	2	Loamy	Round
12	OSZAF	Walmara	2395	2	Red	Round
13	West Shewa	Chaliya	2317	3	Red	Round
14	West Shewa	Chaliya	2254	5	Red	Round
15	West Shewa	Chaliya	2525	4	Red	Round
16	West Shewa	Chaliya	2477	3	Red	Round
17	West Shewa	Chaliya	2475	4	Red	Round
18	West Shewa	Chaliya	2437	4	Red	Round
19	West Shewa	Chaliya	2477	4	Red	Round
20	West Shewa	Chaliya	2408	4	Red	Round
21	West Shewa	Chaliya	2356	4	Red	Round
22	West Shewa	Chaliya	2452	1	Loamy	Round
23	West Shewa	Chaliya	2359	5	Loamy	Round
24	Ilubabor	Alle	1758	6	Red	Round
25	Ilubabor	Alle	1779	6	Red	Round
26	Ilubabor	Alle	1862	1	Albizia trunk	Round
27	Ilubabor	Didu	1820	6	Loamy	Round
28	Ilubabor	Didu		3	Red	Round
29	Ilubabor	Alle	1899	6	Albizia trunk	Round
30	Ilubabor	Alle	1913	3	Red	Round
31	Ilubabor	Alle	1910	3	Red	Round
32	Ilubabor	Alle	1934	5	Red	Round
33	B/Bedele	Gachi	2108	3	Red	Round
34	Jimma	Gera	1920	6	Loamy	Round
35	Jimma	Gera	1951	6	Loamy	Round
36	Jimma	ShebeSenbo	2130	4	Loamy	Round
37	East Guji	ArdaJila-Me'eBoko	2236	3	Red	Round
38	East Guji	ArdaJila-Me'eBoko	2265	4	Red	Round
39	East Guji	ArdaJila-Me'eBoko	2258	1	Red	Round
40	East Guji	Anna Sorra	2384	4	Loamy	Round
41	East Guji	Anna Sorra	2311	4	Red	Round
42	East Guji	Anna Sorra	2313	3	Red	Round
43	East Guji	Anna Sorra	2430	3	Red	Round
44	East Guji	Anna Sorra	2407	3	Red	Round
45	East Guji	Anna Sorra	2329	3	Loamy	Round
46	East Guji	ArdaJila-Me'eBoko	2260	3	Loamy	Round
47	East Guji	ArdaJila-Me'eBoko	2262	4	Sandy	Round
48	East Guji	Anna Sorra	2358	3	Red	Round
49	East Guji	Anna Sorra	2370	3	Red	Round

Nesting habitat type: Grass land (1), Woody shrubs along field side (2), cultivated land (3), protected forest (4), Eucalyptus plantation (5) and Natural coffee forest (6).

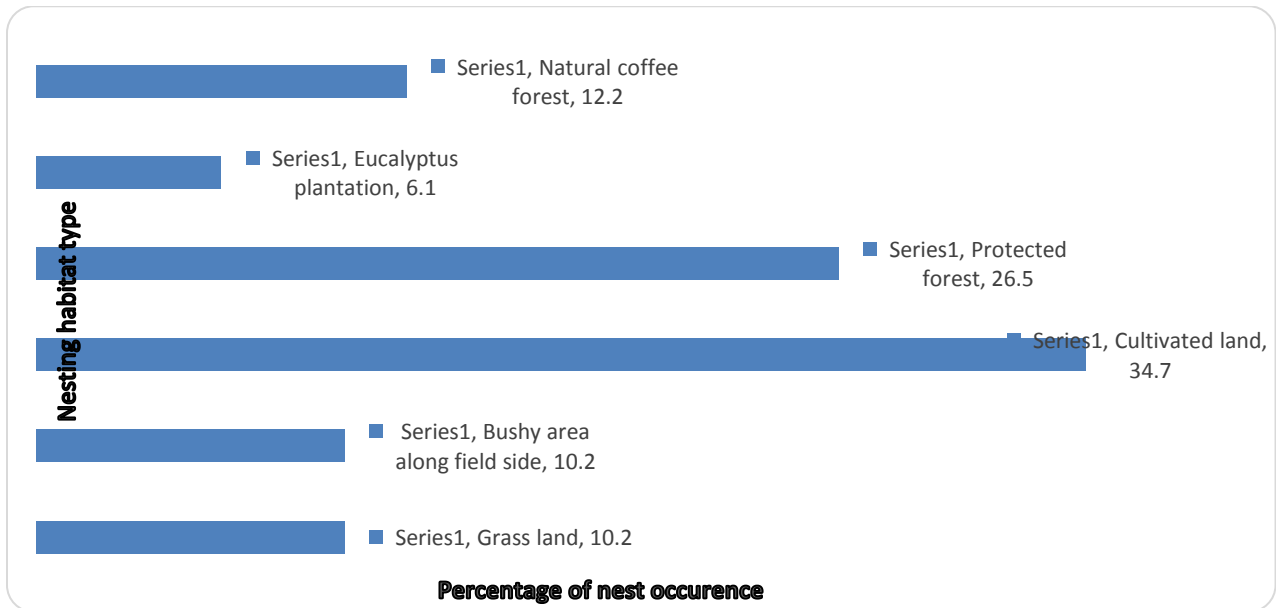


Figure 3. Percentage of stingless bee nests found across nesting habitats in the study areas.

### *External characteristics of the nests*

The external entrance tubes of *L. bottegoi* and *M. baccarii* were assessed. The entrance of *L. bottegoi* was horizontally arranged and faced in the eastwards direction. It was also surrounded by dot like rim of sticky material (Figure 2B). However, the nest entrance of *M. baccarii* was up right and the area closer to the tube was kept clean. *M. baccarii* nests were hardly visible, with rounded shapes. The entrance and opening of the nests of *M. baccarii*, were not varied in shape. The external entrance tubes were short and soft in rigidity, built from dark cerumen, opening into long and hard brittle internal tunnel. The external entrances of colonies were raised on average 0.85 cm above the ground. The diameter of the entrance orifice, nest entrance tunnel thickness and number of guard bees on the internal part of the entrances are presented in Table 2. Unfortunately, it was difficult to measure the diameter and length of nest entrances and the number of guard bees on the entrance of *L. bottegoi*, as the entrances were destroyed while cutting the trunks.

Table 2. Results of measurements recorded for *M.baccarii* nests from different zones of Oromia Regional State

Parameters	N	Minimum	Maximum	Mean	Std. Deviation
Altitude	48	1758.00	2543.00	2256.56	218.73
Entrance tunnel length above the nest (cm )	47	14.00	43.00	28.64	5.92
No. of guard bees on the entrance	35	0.00	13.00	4.97	2.93
Nest entrance height above the ground (cm)	33	0.00	1.67	0.85	0.40
Nest entrance tunnel diameter (cm)	47	0.70	1.30	0.99	0.20
Nest entrance tunnel thickness (cm)	7	0.12	.20	0.16	0.03
Nest width (cm)	47	11.00	35.00	23.59	5.04
Nest Height (cm)	47	10.00	37.00	20.09	6.80
Nest depth below the ground (cm)	47	34.00	74.00	50.50	9.32

### *Internal characteristics of the nests*

The internal tunnel length above the nest to the ground level, the nest depth below the ground to the bottom of the nest, and width and height nest of *M. baccarii* are depicted in Table 2. The inner surface of the entrance tube was decorated with a black cerumen. Nests of the colonies were situated at different depths. All the nests were surrounded by a dark brown to black brittle batumen layer connected through short pillars protruding from a wall of the nests to the soil (Figure 4A).

The nests contained food storages (honey and pollen pots) and brood zones inside their batumen layers. The food pots were frequently located lateral to the brood nest, with honey pots at a periphery of a nest and pollen pots closer to the boundaries of brood nests. Next to food pots, several brown layers of involucre were arranged separating storage zone from the brood zone (Figure 4C). Honey pots were oval in shape and built with thick dark to brown cerumen connected to the batumen layer through pillars. The honey pots were slightly darker than the pollen pots. The height and diameter of the honey pots ranged from 8 – 20 mm and 13 – 23 mm with mean value of  $13.79 \pm 3.70$  mm and  $23.00 \pm 6.12$  mm, respectively. The volume of honey in a pot varied from 5.00 to 9.00 ml with mean value of 7.57 ml.

Unlike that of *M. baccarii*, the internal entrance tube of *L. bottegoi* was inconspicuous. Like in *M. baccarii*, food pots were larger than brood cells in *L. bottegoi*, however, both food pots and brood cells were smaller than that of *M. baccarii*. Food pots of *L. bottegoi* were oval in shape, yellowish in colour and arranged in clusters. Honey pots were located at the periphery of a nest and some were intermixed



Figure 4. Nest architecture of *M. baccarii*: general view of the nest in the ground cavity (A), nest entrance tunnel above the nest (B), storage zone (C), brood combs enclosed in layers of involucrum (D).

### ***Brood nest***

Brood nest of *M. baccarii* situated at the center of the nest surrounded by several sheaths of involucrum. The brood nest contained horizontally arranged brood combs. The horizontal brood combs were usually round in shape. The size and number of brood combs varied among nests from 38.36 – 90.40 mm in diameter and 5 – 12 in number, with mean values of  $65.93 \pm 13.38$  and  $8.91 \pm 1.95$ , respectively. The average thickness of brood comb was  $4.52 \pm 1.01$  mm with a range of 2.82 – 7.00 mm. Brood combs were arranged one over the other in all the nests assessed except in one colony, which were united in a spiral shape. The average distance (pillar height) between two successive combs was  $4.28 \pm 0.77$  mm. Combs were supported by many pillars, with thickness varying from 1 – 3 mm and an average of  $1.59 \pm 0.43$  mm. Brood combs containing eggs and larvae were dark brownish in colour, while combs with old pupae and emerging bees were yellowish and creamy white in colour respectively (Figure 5A, Band C). Unlike honeybees, *M. baccarii* cells were vertically constructed and old comb from which young bees emerged was destroyed by worker bees (Figure 5B). Worker brood cells had an average depth of  $6.72 \pm 0.72$  mm and average width of  $3.36 \pm 0.24$  mm. Number of cells per  $\text{cm}^2$  for *M. baccarii* was varied from 9 – 10 cells, with mean of  $9.60 \pm 0.34$  cells.

Large queen cells were detected at the periphery of brood combs in all the colonies except in two colonies, however, only single queen per colony was found in 14 assessed *M. baccarii* nests (Figure 5C and D). The number of queen cells per colony ranged from 0 – 12 with an average of  $7.92 \pm 3.84$ . The

queen cells were pale in colour and connected to brood combs with short pillars (Figure 5C). The correlation between number of queen cells and number of brood combs was found to be significant ( $p < 0.05$ ).

Unlike in *M. baccarii*, layer of involucrum around the brood was absent in *L. bottegoi*. Brood cells were oval in shape and creamy white in colour. The amorphously clustered brood cells were located at the center of the nest with some spaces which enabled easy movement of the bees within the cluster of brood cells (Figure 5E). The clustered brood cells were surrounded by pollen and honey pots (Figure 5G).

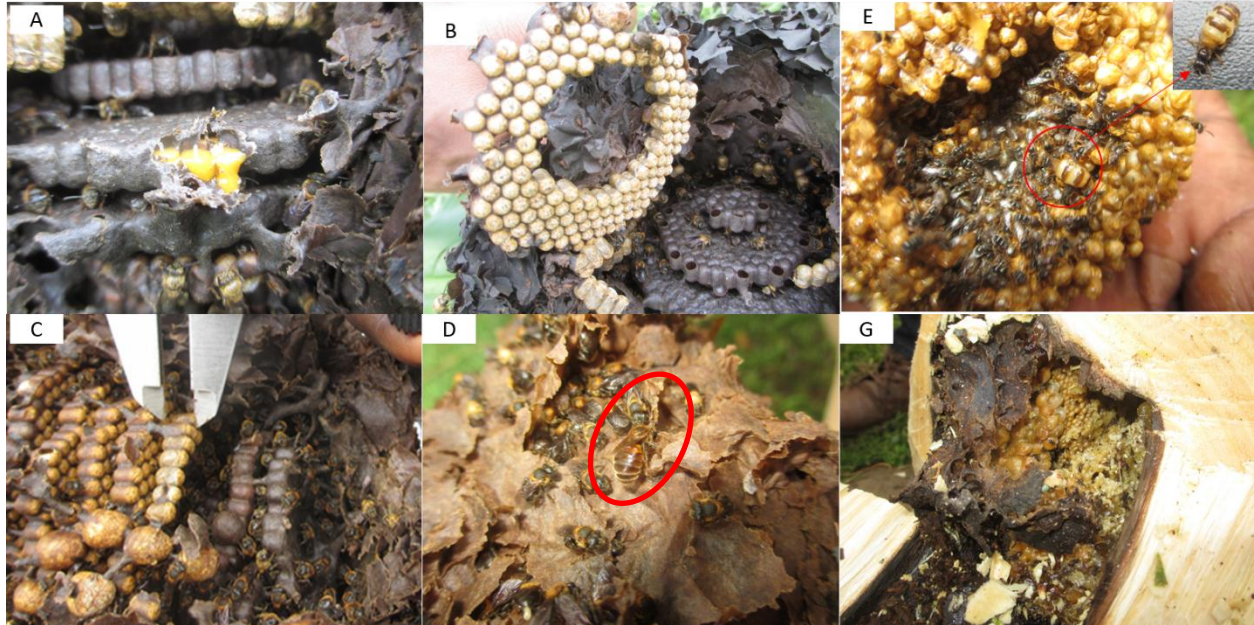


Figure 5. Brood nests of *M. baccarii* and *L. bottegoi*: young brood comb cells with stored larval food and eggs (A), old brood comb from which new worker bees started emerging at its center (B), young and old brood combs with queen cells at the edge (C), queen bee of *M. baccarii* (D), amorphously clustered brood cells with queen and worker bees of *L. bottegoi* (E) and nest of *L. bottegoi* (G).

## Discussion

The diversity of stingless bees varies among different habitats and usually species diversity is highest in natural forests (Nkoba et al., 2012; Suriawanto et al., 2017). In this result, only two species of stingless bees found in different habitats occupying two distinct nesting substrate types. The stingless bee species (*L. bottegoi* and *M. baccarii*), were found in overlapping areas in the midland, whereas only *M. baccarii* was found in the highland areas of Oromia. In general, highland ecosystem has less diverse stingless bee species (Salim et al., 2012; Suriawanto et al., 2017). Although there are five stingless bee species existing in the county (Pauly and Zewdu, 2013), only two species were found in the current study. *M. baccarii* nest is the widely distributed, in all sampled districts covering all habitat types with three various soil types. The two nests of *L. bottegoi* was found in natural coffee forest nesting in the cavities of *Albizia shimperiana* trunk and detected only in one district. The occurrence of two distinct stingless bee species in natural forest suggests that it provides different nesting substrates that serve as a shelter for the bees (Siqueira et al., 2012). Natural forest present different physiognomies that accounts for the stingless bee species diversity through providing suitable nesting sites, which are considered as limiting factors for

diversity of stingless bee (Starr and Sakagami, 1987). The occurrence of *M. baccarii* nests in soils with different physical properties suggests that selection of nesting cavity is not affected by soil type like in other subterranean stingless bee species, such as *Geotrigona subterranean* and *Geotrigona nombuca* (Barbosa et al., 2013).

Stingless bees have a wide range of nesting and feeding behaviours which make them to adapt to various types of habitats for nesting (Roubik, 2006). Habitat type and its characteristics are important factor regulating nest population and nesting pattern as stingless bees, like other animals, are highly depending on the quality of their habitats (Nkoba et al., 2012). To this end, the variation in the number of nests among the six habitat with the highest number of *M. baccarii* nests in cultivated lands found within one kilometer distance from forest areas. The highest number of nests in croplands near the forests suggests that the environment with low and open vegetation give chance to get more nests easily. Nevertheless, in various kinds of forest habitats with closed canopy finding stingless bee nests is more difficult (Siqueira et al., 2012). Moreover, habitat heterogeneity adjoining forest areas (Siqueira et al., 2012) and edges of cultivated lands (Wondmeneh et al., 2020) account for the abundance of stingless bee nests. Furthermore, it has been suggested that stingless bees prefer to nest in open areas than dense vegetation as open areas allow high light incidence, which directly affect the external activities of bees (Barbosa et al., 2013). These characteristics could elucidate the abundance of nests in croplands near the forests, which provide a nesting sites and food resources.

The characteristics of stingless bees' nest entrances found to be different according to the genus (Kajobe, 2007; Kelly et al., 2014). Nest entrance properties of stingless bees related to many factors, such as age of the nest, microclimate, foraging activities and defense (Roubik, 2006). For instance, the eastward facing of the nest entrance of *L. bottegoi* is probably to positioned the nest to the early morning sun light, which brings about warm temperature initiating foraging flight (Kajobe, 2007). However, it is not possible to deduce that all *L. bottegoi* colonies prefer the eastward facing, since the information from two colonies is not enough. The deposition of sticky resin around the entrance of this bee species may serve to guide foraging bees to the nest and helps to protect the colony from natural enemies as for other stingless bee species (Barbosa et al., 2013).

The entrance is an important parts of the nest in which bees are communicating with their surroundings (resource and natural enemies). Thus, the characteristics of external nest entrance and its size play a fundamental role for the survival of the colony in the nest (Couvillon et al., 2008). In this study, the upward directed, hardly visible, rounded and soft in rigidity of external nest entrance of *M. baccarii* with the varying tube size may serve the purpose of survival of the colony in the nest. The raised structure of the entrance serves as guiding and landing purpose for the forages, as well as for colony protection (Biesmeijer et al., 2005; Viana et al., 2015). The current finding in the variation of nest entrance diameter among the colonies supports the previous report indicating a variation in nest entrance diameter of *M. baccarii* colonies (Wondmeneh et al., 2020), which may be related to nest defense and foraging activities of the species (Viana et al., 2015). Lesser entrances are easier to defend, however, larger entrances allow easy passage of heavier foraging traffic (Couvillon et al., 2008).

The nests of eusocial insects are protected by guard bees. This behaviour helps to defend colony resources inside the nest (Grüter et al., 2011). In this context, several guard bees (0 – 13 guard bees) positioned at the inner part of the external entrance suggest that the significance of guard bees to defend their colonies.



The range of the number of guard bees keeping the entrance is almost similar with the previous reports for *M. baccarii* (Alemayehu and Zewdu, 2021; Wondmeneh et al., 2020). The variation in the number of guard bees may be related to several factors, including the time of the day, size of the colony, weather condition, size of the entrance, level of foragers trafficking and presence or absence of natural enemies (Alemayehu and Zewdu, 2021; Couvillon et al., 2008; Grüter et al., 2011; Wondmeneh et al., 2020).

The ground nesting stingless bees apparently nest at a great enough depth to attain a stable nest environment (Moritz and Crewe, 1988). The nest cavities of subterranean stingless bees were connected to the outside through entrance tunnel. The length of the access tunnel depends on the nest depth and shape. This is reflected in the nest cavities of *M. baccarii* constructed at great depth in the ground (34.00 – 74.00 cm) and connected with long entrance tunnel above the nest cavity to the ground level (14.00 – 43.00 cm). Such long entrance tunnel was not observed in the *L. bottegoi*, suggesting that different stingless bee species construct their nests with varying lengths of internal tunnel. However, it is within the ranges of entrance tunnel length of *M. baccarii* reported from Northern Ethiopia (23.4 to 35.4 cm) and Cameroon (27.5 to 34.5 cm) (Wondmeneh et al., 2020). The shape of the entrance tunnel is almost straight like that of *Geotrigona* subterranean (Barbosa et al., 2013). The entire nest chamber is bounded in layers of black and brittle batumen with short pillars projected to wall of the cavity as well as between layers of batumen (Wondmeneh et al., 2020). The current nest chamber width and length of *M. baccarii* is relatively smaller than the previous report from Northern Ethiopia (Wondmeneh et al., 2020), whereas it is larger than the nest size reported in Cameroon. This is likely the result of restrictions imposed by the nesting cavity limitations, and is often geographically and phylogenetically related (Gonzalez et al., 2018; Roubik, 2006), which suggests further work on factors affecting nest cavity of *M. baccarii*.

The nest of stingless bees, which is enclosed in a batumen, contains food storage and brood zone. In this study, the brood chamber of *M. baccarii* was externally covered by layers of involucrem made of cerumen. The involucrem has a function of separating the brood chamber from storage zone (Barbosa et al., 2013). Moreover, the layers of involucrem play a key role in thermoregulation of the brood chamber, as stingless bees do not thermoregulate their nests as precisely as honeybees do (Alves et al., 2019). In these bees, the involucrem retains thermal energy generated by adult bees and partly by the mass of brood in the brood area (Sung et al., 2008). In general, temperature variation in the soil is thought to be low but the soil temperature at the average depth where subterranean bees like *M. baccarii* built their nests is lower than the brood nest (Barbosa et al., 2013). This may be elucidated the need of several involucrem sheets around the brood area. Unlike in *M. baccarii*, involucrem was absent in *L. bottegoi* colony that built its brood cells in cluster. This result is in agreement with the finding that some species of stingless bees like *Trigona fulviventris* build no involucrem around their brood (Barbosa et al., 2013). This indicates involucrem may be an optional structure in stingless bee nests, and its construction is related to external temperature conditions.

In general, most stingless bees are cavity nesters and some of them arrange their brood cells in combs, others build brood cells in clusters (Michener, 2007). In our result, the former type of cell arrangement was present in *M. baccarii* and the latter type of cell arrangement was found in *L. bottegoi*, suggesting that the two species adapted different types of brood cells arrangement. Cluster cell arrangement is found in several groups of small to minute stingless bees (Melo and Costa, 2009; Michener, 2007) and it seems to be an adaptive feature of small bees nesting in small and irregular cavities (Gonzalez et al., 2018). On the other hands, *M. baccarii* observed to build vertical brood cells arranged in horizontal brood combs.

The number of brood combs per colony of *M. baccarii* varies from 5 – 12, which is similar to what has been previously reported for nests of the same species from Northern Ethiopia (Wondmeneh et al., 2020). However, the diameter of the comb is slightly shorter than the range reported from the North, which may be influenced by geographical factor that can limit size of nest cavities (Alves et al., 2019).

In the present study, varying number of queen cells (0 – 12 cells/colony) were detected at the periphery of horizontal brood combs connected to the combs through short pillar in all colonies except in two colonies, suggesting that most of *M. baccarii* colonies construct queen cells in wild colonies as reported in other stingless bee species (Alves et al., 2019). The significant positive correlation ( $p < 0.05$ ) between number of queen cells and brood combs, suggests that a higher number of brood layer represents an increase in number of queen cells which in turn indicating high tendency of reproduction. This evidence suggests the existence of high probability of swarming in wild colonies of *M. baccarii*, which could create a good opportunity to extend their generation

## **Conclusion**

This study underlined stingless bee species diversity varies among different habitats and usually species diversity is the highest in natural forest. Moreover, habitat type and its characteristics are among important factors regulating nest population and the chance of observing stingless bees' nests in cultivated lands adjoining forest areas. Furthermore, this study successfully described the external and internal nest characteristics, and their bionomics. The nest characteristics and internal nest biology also varied between colonies of stingless bees. These variabilities among different nest characteristics measured are related to many factors, such as microclimate, predators and foraging activities of the species. Understanding the nesting habitat and substrate preferences, as well as nest architecture and nest biology of each species might not only help to characterize the species but also helps for development and implementation of proper conservation strategies, and provides important information for the sustainable colony management. Thus, the information generated on nesting substrates, nest architectures and nest biology are helpful and starting points to design suitable bee hives for keeping and conserving both species. Moreover, further investigation will be required to generate enough data for smaller stingless bees at lower altitudes

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# Investigating the defensive mechanisms of stingless bees, *Melluponela baccarii*, against Ants (*Dorylus fulvus*)

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## Abstracts

*Stingless bees are unable to sting as the stinger is vestigial. Despite lacking a sting, stingless bees are active in defending their enemies possess numerous defensive mechanisms. The study was conducted at Holeta Bee Research Center, stingless bee (Melluponela Baccarii) apiary site to investigate the defensive mechanism of stingless bees (M. baccarii) against ants (D. fulvus). Defensive behavior of M. baccarii against D. fulvus was tested in Holeta Bee Research Center laboratory. To Identify how stingless bee, M. baccarii defend itself from ant, D.fulvus, three different defensive reactions: Dyadic encounter, a (defensive between single worker of D. fulvus and M. baccarii), Group Interaction (Defensive between thirty two workers from each group) and Colony interaction (between a mass of colonies) were tested. The interaction between ants and stingless bees in dyadic and group interaction were overpowered by ants where as in colony interaction, ants run away without tempting any physical fight. This is may explain why stingless bees living in free environment without being attacked by ants though they share the same ecology. Therefore, identifying these ant-deterrent and exploring for the development of ant protection is suggested.*

**Key words:** *Stingless bees, ants, defensive, dyadic, group, colony interactions*

## Introduction

Social insects use a variety of strategies to defend their nests against predators. In extreme cases, some involves the self-sacrifice of defenders (Shorter and Rueppell 2012). Social insect nests are worth defending as they contain not only offspring (brood) but also the reproductive individuals, food stores, and nesting material, while the nest itself is often a valuable resource (Seeley 1985; Roubik 2006). Some social insect workers involve self-suicidal defenses, while others seal their nest entrances from the outside to protect against their enemies and during harsh conditions. In honey bees (*Apis mellifera* L.), sting autotomy is well known that involves the self-amputation of the sting apparatus from their body. This increases venom delivery, releases alarm pheromone and the apparatus can continue to pulsate long after the stinging event (Hermann 1971; Burrell and Smith 1995). Autothysis, the rupturing of the body wall to release defensive chemicals, is known in some ant species (Davidson et al. 2012) and termites (Bordereau et al. 1997). A similar mechanism has been described in aphids, which produce a sticky secretion causing the defending aphid adhere to the predator, thereby immobilizing it (Uematsu et al. 2010). All of these strategies combine a behavioral component with morphological adaptations which inevitably cause mortality in the defending workers.

Stingless bees (Meliponinae), comprises many hundred described species, are found worldwide in the tropics and sub-tropics. They are closely related to honey bees, and live in perennial eusocial colonies (Michener 2000; Roubik 2006). As their name suggests, stingless bees are unable to sting as the stinger is

vestigial (Michener 2000). Even though defense is important for colony survival, non-stinging particularly in stingless bees is an important phenomenon to protect them from self-scarify unlike body rupture in stinging *A. mellifera* workers. But, stingless bees face predation at the nest from many sources ranging from mammals to nest-robbing bees (Wille 1983; Suka and Inoue 1993; Roubik 2006). Despite lacking a sting, stingless bees are active in defending their enemies possess numerous defensive mechanisms including biting, harassment, caustic chemicals, alarm pheromones, and hovering guards (Van Zweden et al. 2011). As a result, stingless bees (Meliponinae) are less likely attacked by their natural enemies like safari ants, wasps, spiders and small hive beetle (personal observations). Rather, they are highly affected by anthropogenic effects like deforestation and habitat fragmentations with poor management.

Driver ants (*Dorylus fulvov*) are the most damaging pests known to attack honeybees in various countries in sub-tropical countries of Africa (Adjare, 1990; Hepburn & Radloff, 1998). It is reported to be the most serious and widespread natural enemies of honeybees in Sudan (El-Niweiri *et al.*, 2004), in Uganda (Kajobe *et al.*, 2009) and in Ethiopia (Desalegn, 2007). Ants cause serious damage to honeybees and their products and as a result many productive bee colonies have been killed due to ants attack. Furthermore, persistent attack by ants has been reported as an important cause of the absconding of honeybee colonies (Hepburn & Radloff, 1998; Rachna & Kaushik, 2004; Desalegn, 2007.) The estimated annual economic losses as a result of loss of honeybee colonies and their products due to ants attack in the West and South West Shewa Zones of Ethiopia is to exceed \$ 250, 000 per year (Desalegn, 2007).

The interaction between driver ants (*D. fulvus*) and stingless bees *M.Baccarii* is not yet investigated despite their living environment is similar (both harboring underground). Even though stingless bees are much vulnerable to ants attack due to their nesting nature, it is not common to observe that stingless bees are attacked by ants as that of the honeybees (Personal observation). This indicates that the stingless bees may involve different and effective defensive mechanism against ants as compared to honeybees. Exploring the behavioral defensive strategies of the stingless bees against their enemies was a paramount important to co-manage them with honeybees and in turn prevent against ants. Therefore, confirmation of our observation and understanding the defensive mechanisms of the stingless bees against ants is a paramount important. Thus this study was initiated to determine whether stingless bees defend themselves from ants attack and identify their defensive mechanisms.

## **Materials and methods**

The study was conducted at Holeta Bee Research Center (HBRC). Twenty stingless bee colonies established in pot hives at HBRC were used as source of stingless bee for the envisaged experiment.. An observation platform with circular dimension was prepared as a media to easily observe behavioral and interaction between the two species during the experiments.

### ***Study on defensive mechanisms***

To describe and quantify the behavioral defensive mechanisms and inter specific encounters between *M. Baccarii* and *D. fulvus*, three different bioassays were used: Dyadic encounters, Interaction between groups of workers and interaction between colonies.

### ***Dyadic encounters***

Dyadic (one-to-one) encounter was used to determine the fighting ability of individuals. An observation platform with a perforated cover that allowed circular dimension (diameter 5 cm, height 7 cm) with a cover that allow air circulation was made as a media. Single worker from each species was randomly picked and then placed simultaneously in an observation platform. A total of 20 fighting reactions were conducted. Fight initiators, type of interaction (physical fighting), the winner and the time required to win were observed and recorded. In addition to physical fighting, the incidence of a strong abdominal gesture flex as an indicator of chemical defense were observed.

### ***Group interaction***

The test using interaction between groups of worker *M. baccarii* and *D. fulvus* aimed to assess the fighting abilities of both species and to determine the level of workers aggressiveness. Thirty two workers from each species were randomly selected and kept in separate observation platforms according to procedure of N. Adgaba et al., (2014). For *D. fulvus*, equal proportions of the major, median and minor morphs were used. Both sets of workers were tipped gently into a single plastic container (30 ×20×8 cm) with a transparent cover. The workers of each species were placed in opposite sides of the box separated by piece of wooden board before the start of observation. The results of fight in 10, 20, 30, 40 and 50 minutes were recorded using fresh groups each time. Each duration were repeated three times. The mean number of ants and stingless bees that died was recorded. In addition to physical combat, gesture flex of both species was recorded.

### ***Colony interaction***

For the test of interaction between colonies, large numbers of individuals of ant species was used in accordance with the procedures described by Human and Gordon (1996, 1999) and Zee and Holway (2006), with some modifications. The luring of driver ants into *M. baccarii* domestic sites by providing them with raw or rotten meat is common in rural areas of Ethiopia to eradicate pests, such as cockroaches, house bugs, spiders and others. In our first set of experiments, ant colonies was attracted by providing them with honeybee comb containing different stages of broods (Adigaba et al., 2014). Initially, the comb was placed close to the ants bivouac until the worker swarm moved on to it. After approximately 30 min, the comb was gradually dragged along the ground, with an additional brood dropped to motivate foraging along the way, until it will very close to the stingless bee nest. In this way, it was easy to induce large numbers of *D. fulvus* workers into the *M. baccarii* territory and bring the two species into contact. In the second set of experiments, a portion of *M. baccarii* nest with brood and adults was removed from its original nest and placed near the ground close to a *D. fulvus* bivouac from where the latter was marching on honeybee brood combs. To assess how territorial defense was affecting the levels of combat, the experiment was repeated four times in *M. baccarii* territory and four times in *D. fulvus* territory.

### *Statistical analysis*

All the data were analyzed using descriptive statistics and presented by table and graph

### **Results and discussions**

#### *Dyadic encounters*

From all 20 Dyadic tests, 70% (14) of the fights was initiated by *D. fulvus* and the rest 30% (6) was by *M. baccarii* (fig.1). *D. fulvus* worker fighters were imperative to start fight immediately while *M. baccarii* worker fighters were slow to join fight. *M. baccarii* were more injured by *D. fulvus* and injuries were observed on the legs and wings of *M. beccarii*. *D. fulvus* physically fought *M. baccarii* using its mouth part (biting). Thus the individual physical fight was led by ants.

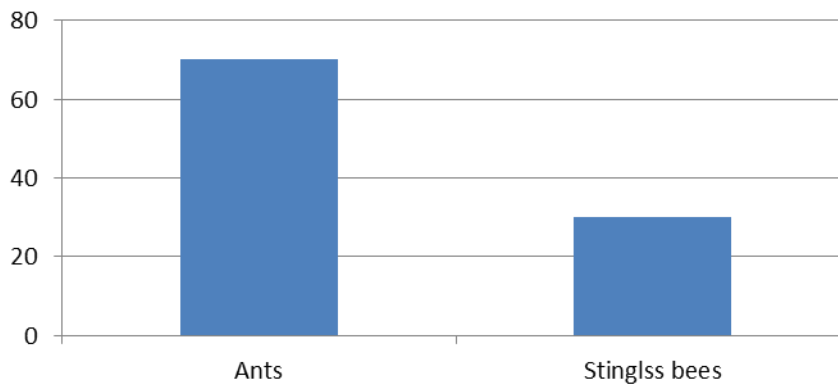


Fig 1. Percent of fight initiators during dyadic interactions

Ten percent (10%) of the stingless bees were died in 20 minutes of the dyadic interaction and the rest 90% of them in 30 min of fighting. On the other hand only 10 of the ants were died until 40 minutes. The physical fighting was controlled by ants mainly by attacking the stingless from back by biting and pulling their legs. This suggests that stingless bees were not defending ants through physical interaction (fighting).



Table .1 Percent of death recorded in different times during dyadic interaction

T Time(minutes)	Stingless bees' death (%)	\Ant death (%)
10	0	0
20	10	0
30	90	0
40	0	10

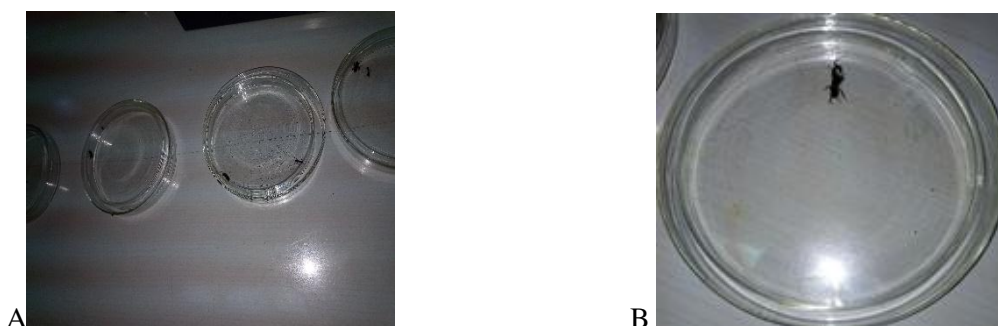


Figure 2. Dyadic interaction: individual stingless bee and Ant introduced to the media (A), fight between stingless bee and ant (B) .

### **Group interaction**

In group interaction, it was observed that ants were fighting in group against stingless bees, while, stingless bees fight in individual. In group interaction, *D. fulvus* showed similar reaction as that of dyadic. *D. fulvus* fought in group as well as individually. During the interaction or fight, *D. fulvus* workers pulled the legs of *M. baccarii* in different directions until they became spread-eagled and motionless. In *M. baccarii*, defending interaction was rare and of short duration. In 30 min fighting, 100% of stingless bees were died. During fight, stingless bees were physically injured and lost their legs and wings. As the duration of fighting increased, the numbers of dead *M. baccarii* workers was increased. No dead ants was observed in group interaction during the study period (Table 2). This test showed that Ants fought aggressively in group as compared to dyadic interaction.

Table 2. Percent of deaths recorded during group interaction between stingless bees and ants in different times

<b>Time)</b> Time(minute)	Stingless bee death (%)	Ant death (%)
10	10.7	0
20	66.7	0
30	22.6	0
40		0
50		0



Figure 4. Group interaction between stingless bees and ants .

### ***Colony interaction***

The interaction between the *D. Fulvus* and *M. baccarii* colonies was observed and recorded. The test was conducted in two situations: first, when a portion of *M. baccarii* nest with brood and adults introduced closer to the *D. fulvus* nest and secondly, when ant colonies collected by honey and brood introduced to the area where stingless bee colony nested.

In both situations no fights was observed between stingless bees and ant colonies. The ants were run away aggressively when introduced to near stingless bees nest and no stingless bees and ants are seen attempting to fight each other. In similar way ants disturbed and run away from its nest when a portion of stingless bee colony containing brood and adult stingless bees introduced closer to ant's nest.

This suggests that the nest of stingless bees may have ant deterrent effect, and defend themselves from ant attack not by physical fight. This result agrees with report of Lehmborg et al. (2007).



Figure 5. Runaway of ants during interaction between ants and stingless bee colonies

### **Conclusions and Recommendation**

Ants were the fight initiators, and defeated stingless bees in Dyadic and group interactions through physical fighting and biting. However, in colony interaction at natural nest sites, stingless bee colony deters ants. This is may explain why stingless bees living in free environment without being attacked by ants though they share the same ecology. Therefore, identifying these ant-deterrent and exploring for the

development of ant protection and for other uses is suggested. Moreover keeping honeybee colonies and stingless bee colonies in the same apiary may serve to protect honeybee colonies from ant attack.

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# Investigating the role of honeybee pollination on fruit and seed yield of coffee (*Coffea arabica*)

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## Abstract

*Coffea arabica* is an autogamous plant, but the use of insect pollinators particularly honeybees would increase the seeds yield of coffee. Therefore, this study was conducted in Jimma zone at Gera Sub-site Research with the objective to identify the role of honeybees 'pollination on fruit set and seed yield of *C. arabica* (74165) variety. It was planted with 2 mx 2 m space. All study plants subjected to similar input and management practices. Fourty eight (48) stands of matured *C. arabica* plants were randomly selected for the study. When the plants reached the age of flowering, each plot was divided into three treatment groups, with 16 observation plants each. T1 plants were caged with a honey bee colony using nylon mesh at 2% of blooming state of coffee flowers; T2 plants were left open for free access to all insect pollinators and T3 plants were caged without any insect pollinators. Coffee plants caged with honeybees had higher fruit set and seed yields compared to self-pollinate and open pollinated; though statistically similar with open treatment. On the other hand, coffee plants caged without insect pollinators had more aborted seeds compared to plots caged with honeybees and open pollination. This study showed 22.61% and 70.98% overall seed yield advantage when using honey bees as pollinators for *C. arabica* compared to open pollination and self-pollination respectively. This shows *Coffea arabica* (74165) variety depends on intensive honey bee pollination. Therefore, integration of honey bee colonies with coffee production is recommended to boost the seed yield of coffee.

**Keywords:** Honeybees, Pollination, *Coffea arabica*, yield, seed

## Introduction

Pollination plays a vital role in maintaining the natural balance of ecosystems. It is the cornerstone of crop production, providing a link between agriculture and pollinators (Shaden *et al.*, 2021). Globally the average economic value of pollination was 153 billion EUR in 2005. The leading categories of insect-pollinated crops are vegetables and fruit, making around EUR 50 billion each, followed by edible petroleum crops, stimulants, nuts and spices (Pashte and Kulkarni, 2015). As a result, the need for insect pollinators is becoming popular among farming communities to increase the productivity of the crops (Hajjar *et al.*, 2008). Globally 75% of important crop species are pollinated by insect (Khalid *et al.*, 2012; Klein *et al.*, 2007).

Honeybees are responsible for 70-80% of insect pollination (Klein *et al.*, 2007). This indicates how much honeybees are the most efficient insect pollinators of cultivated crops and wild flora in agricultural systems. The main reason is that honeybees are abundant and widespread everywhere (Tura and Admassu, 2019). They have well developed mechanism of communication to exploit their environment.

The mutualistic relationship between plants and honey bees depends on the availability of nectar and pollen (Shaden *et al.*, 2021). This means plant species provide food for honeybees while honeybees play an important role in plant reproduction through pollination.

Coffee is a perennial crop that belongs to the Rubiaceae family and originated from Ethiopia (DaMatta, 2004; Belitz *et al.*, 2009). It is the most important cash crop for trading and exports since the 18th century, providing jobs for millions of families worldwide. The flowering period (anthesis) of *C. arabica* is varied from region to region. It flowers from January to April (Ngo *et al.*, 2011) based on the rainfall of the area. *C. arabica* has a short flowering length with the range of 5-7 days (Tura *et al.*, 2021). It gives flowers profusely after rains but some coffee trees are found with flowers at any time of the year (Admassu *et al.*, 2014). *C. arabica* is defined as an autogamous plant with some degree of cross-pollination with insect intervention (José *et al.*, 2002). *C. arabica* is self-pollinated, but the use of honeybees would increase the seeds yield of coffee (José *et al.*, 2002; Shaden *et al.*, 2021). Hence, this study was proposed to identify the effect of honeybee pollination on fruit and seed yield of *Coffea arabica* in Gera district south western Ethiopia.

## **Materials and methods**

### ***Study site***

The study was conducted in Jimma Zone in collaboration with Jimma Agricultural Research Center at Gera Sub-site Research. It is 23 km West of Agaro town. The climate is cool and humid. The area receives an annual rainfall of 1906.3 mm per annum distributed unevenly throughout the year. It has the highest and lowest temperature of 24.4 °C and 10.4 °C, respectively. The soil texture is classified as Sandy clay loam with 103 mm/m of TAW. The study area represents one of the coffee growing areas in the country (Etafa *et al.*, 2021).

### ***Experimental setup***

*Coffea arabica* variety (74165) was planted at Gera Sub-site by Jimma Agricultural Research Center during the planting season in June 2012. The recommended spacing of 2m by 2m between plants for coffee was employed. All study plants were subjected to similar inputs and management practices such as fertilizer application, watering, weeding, and disease control. Forty eight (48) stands of matured *C. arabica* plants were randomly selected for the study. When the plants reached the age of flowering, each plot was divided into three treatment groups, with 16 observation plants in each treatment. T1 plants were caged with a honey bee colony using nylon mesh at 2% of blooming state of coffee flowers; T2 plants were left open for free access to all insect pollinators and T3 plants were caged without any insect pollinators.



Figure 1: Caged coffee plants with and without honeybee

### ***Data collection***

Number of flowers per plant was counted at peak flowering time as well as number of flowers per plant was counted at peak fruit set period. Fruit data was collected when the fruit color become red and matured and then the berry was pulped (Figure 2). Data of seeds those floats over the surface of water were taken and it was considered as aborted seeds. In addition to this, data of seeds those aborted before mature were also taken. Seeds and fruits weight were measured by balance. Thousand seeds weight were also taken.



Figure 2: Coffee berry collection, pulping and pulped seed

### ***Estimation of percentage of fruit setting***

Percentage of fruit set was calculated as total number of fruits set divided by the total number of number of flowers \*100 (Wubs and Ma, 2009)

### ***Yield valuation***

The dependency of the coffee seed yield on honey bee pollination was measured as the follow:

Yield increase from all insects excluding self-pollination was calculated as:

- $(A \%) = (Y_{all} - Y_{self}) / Y_{self} \times 100$ , yield increase from honey bees excluding all other insects
- $(B \%) = (Y_{bee} - Y_{all}) / Y_{all} \times 100$ , yield increase from honey bees excluding self-pollination
- $(C \%) = (Y_{bee} - Y_{self}) / Y_{self} \times 100$ ,

Where, *Y*<sub>all</sub> represents treatment T2 (plants open for all insect visitors), *Y*<sub>self</sub> represents T3 (not accessible to any insects – exclusively self-pollinated), and *Y*<sub>bee</sub> describes coffee plants, accessible only to honey bees (T1) (Tura et al., 2018)

**Data analysis:** Descriptive statistics and One-way ANOVA were used for data analysis

## Results and Discussions

The fruit set and seed yields of coffee plants caged with honeybees and open pollination was statistically similar and both treatments were better than self-pollination (Table 1). This may be due to the presence of other pollinators of coffee around our experimental site. However, coffee plants caged with honeybees had higher fresh fruit and seed yield though statistically similar with open treatment. This confirms that honeybees are an important in augmenting the final fruit set and seed yield of coffee, which is parallel with the reports of Krishnan *et al.* (2012). Rachersberger et al. (2019) also reported that honeybees collect both nectar and pollen from coffee flowers and transfer pollen on stigmas even after one visit which enable coffee plants to be pollinated and set more fruits. In addition to this, Ngo et al. (2011) reported that honeybee pollination resulted in almost a doubling the seed yields of coffee compared to self-pollinated coffee plants.

Table 1: Mean and SE of fresh fruit yield (kg/ha) and mean of seeds yield (kg/ha) of Coffee plants caged with honeybees (T<sub>1</sub>), coffee plants caged without insect pollinators (T<sub>2</sub>) and open coffee plants to the environment (T<sub>3</sub>) under pollination experiments.

Treatment	%of fruit setting ± SE	Fresh fruit yield (kg/ha) ± SE	Seeds yield (kg/ha) ± SE
Caged with honeybees	98.4 ± 0.6a	7785.7 ± 813.1 <sup>a</sup>	6062.5 ± 621.8a
Caged without insect pollinators	93.4 ± 0.8b	5820.5 ± 704.8 <sup>b</sup>	3545.8 ± 421b
Open pollination	96.8 ± 0.3a	6910.7 ± 712.8ab	4944.6 ± 523.6ab

Note: Treatments with the same letter in column are not significantly different.

Coffee plants caged without insects' pollinators had higher aborted seeds than the caged with honeybees and open to the environments for open pollination by all insects (Figure 3). Honeybee pollination improves the seed yields of coffee which is in line with the reports of Alexandra (2003). Similarly the study conducted by Samnegård et al. (2014) confirmed as the honeybees are the main day-time pollinator of *C. arabica*. Coffee plants caged without insects had lower fruit setting than those caged with honeybees and open pollinations. This means male and female gamete did not match and no fertilization happened (Figure 4). As a result lower fruit set was recorded for coffee plants caged without insect pollinators.



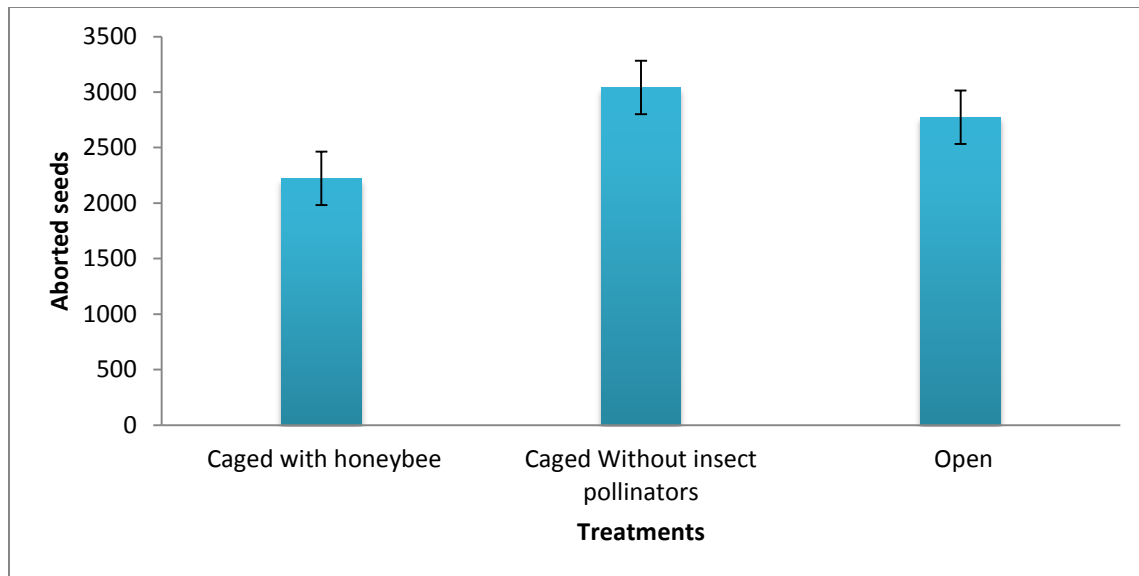


Figure 3: Mean aborted (fly crop) seeds (kg/ha) for Coffee plants caged with honeybees (T<sub>1</sub>), coffee plants caged without insect pollinators (T<sub>2</sub>) and open coffee plant (T<sub>3</sub>) with  $\pm$  SE



Figure 4: Aborted seeds

### ***Seed yield increment***

The results of the study showed a 22.61% overall fruit yield advantage when using honeybees as pollinators for *Coffea arabica* (T<sub>1</sub>) compared to open trees for free access to all insect pollinators (T<sub>2</sub>) (Table 2). This yield increment might be due to the effect of the cage, as it may create stress environments for honey bees and shortage of food, which forces them to visit coffee flowers more frequently. (Pashte and Kulkarni (2015) also reported that honeybees' pollination has direct effects on the profitability and productivity of a substantial amount of global crop varieties, including most vegetables, seeds, and nuts, and some high-value agricultural products, such as coffee.

Table 2: Coffee yield improvement due to honeybees and insects' pollination

Quality seeds yield increment over different treatment	% Of yield increment
A% (Open plots over caged plots without insect pollinators)	39.45
B% (Caged plots with honeybees over open plots)	22.61
C% (Caged plots with honeybees over caged plots without insect pollinators)	70.98

### ***Thousand seed weight***

Thousand seeds weight of coffee plants pollinated by honeybees were higher than plants caged without insect pollinators and open pollinated (Figure 5). This indicates how much honey bees pollination improves the quality of coffee seeds.

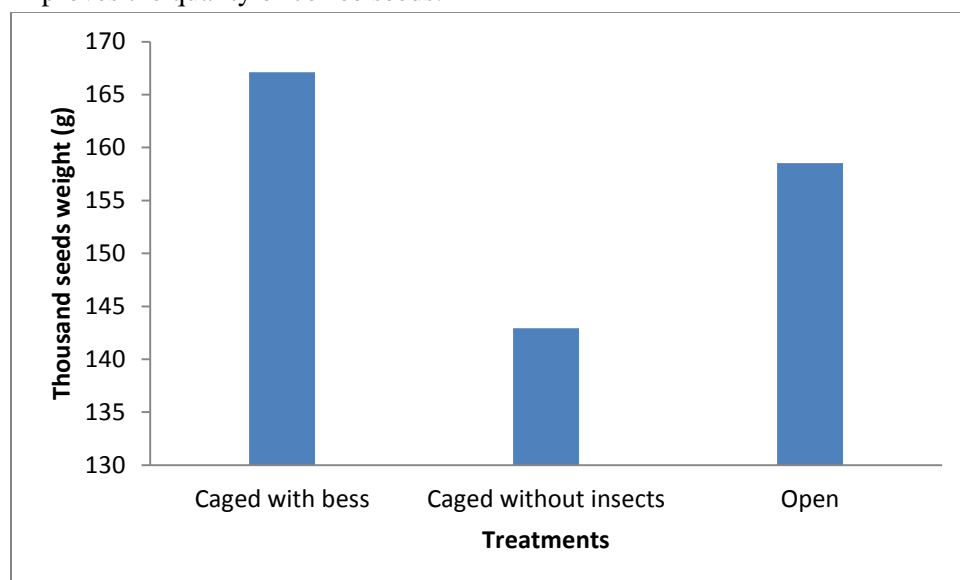


Figure 5: Thousand seeds weight of coffee plants pollinated by honey bees, caged without insect pollinators and open pollinated

### **Conclusions and recommendations**

Our study revealed that *Coffea arabica* (74165) variety depends on intensive honey bee pollination. This study showed 22.61% overall seed yield advantage when using honey bees as pollinators for *C. arabica* compared to open pollination. The use of insect pollinators with free access to the coffee trees also increase seed yield by 39.45% compared to self-pollinated coffee plants. Our findings underline the recommendation to move honey bee colonies to the coffee farm during the blooming season for effective pollination and better seeds yield.

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# Induction of propolis production by *Apis mellifera scutellata* in traditional basket and movable frame hives in Borana Zone, Southern Oromia

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## Abstract

*The study was carried out in Elwoye district of Borana zone with the objective of testing simple ways of inducing propolis production in traditional/basket and moveable-frame hives and to determine its effect on honey production. Four colonies in each type of hive were induced for more propolis production by exposing to cold weather through creating more openings and compared with controls without induction. The harvested Propolis was weighed and the total average propolis production and honey production analyzed. Statistical analysis showed significant differences in propolis collection between the two different hive types ( $P < 0.05$ ). The overall average Propolis collected ( $51.93 \pm 2.30$  g) from movable frame hive (MFH) is higher than the amount collected from basket hive. Moreover, this study showed the overall mean propolis production from induced hives ( $54.16 \pm 2.07$ g) is significantly higher than and the amount of propolis ( $24.19 \pm 2.36$  g) collected from non-induced hive ( $P < 0.05$ ). Honey production was significantly influenced by hive types, the MFH produced an average of 12.08 kg honey/colony and basket type produced on average of 3.25 kg honey/colony. However, in relation to honey harvesting and propolis collecting methods, there was no significant difference ( $P > 0.05$ ) in honey production between induced and control groups Therefore propolis production appears to be a worthwhile practice in the study area for the traditional beekeeper as the aggregate hive productivity will substantially increase without affecting honey production. Awareness regarding, the value of bee products other than honey including propolis and the technique to produce will help to substantially increase hive productivity.*

**Key words:** propolis, *Apis mellifera scutellata*, traditional basket hive, moveable frame hive

## Introduction

Ethiopia has a huge natural resource base for honey production and other hive products and beekeeping is traditionally a well-established household activity in almost all parts of the country. Though honeybees produce several products: honey, beeswax, pollen, propolis, royal jelly and venom, honey and bees wax are the main products used in the country. In most cases propolis production is not practiced in the country at large (Admasu & Fitchel, 1994).

Because of demand in the pharmaceutical trade (Crane, 1990), propolis has become an important honeybee product and is an additional source of income particularly for traditional beekeepers in developing countries. However, due to lack of awareness and absence of suitable means for harvesting propolis, it is not yet exploited in tropical Africa. Like other African countries Ethiopian beekeepers and people at large are unaware about the other bee products. They believe that honey and beeswax are the only products obtained from beekeeping. Moreover, no means have been developed to enhance the propolis harvest from traditional basket beehives, which are a major potential source of propolis. Likewise, in Ethiopia only one scientific experiment has been previously conducted on propolis collection

technique from *Apis mellifera bandasii* honeybee race found at central part of the country. As the result, literature on propolis production is extremely limited in Ethiopia in general and Borena zone in particular. Production of propolis is not a complicated matter and it can be harvested from bee hives without using new technologies by scratching. However, such propolis may be mixed with beeswax, dead bees, hive debris and the quality of the yield would be relatively low (Michael Clarke, 2019). Moreover, because there is no routine inspection in traditional beekeeping harvesting propolis is not as such practiced. Honey bee breed/races, geography, presence of propolis sources in nature, climate condition, beehive types and the strength of the bee colony have been documented as factors affecting propolis production by honey bee colony (Michael Clarke, 2019). Besides, the production methods (Nuray S and Aziz G, 2005) and the optimal time for propolis harvest (Kiziltas and Erkan, 2020) also affect the amounts of propolis that could be harvested per colony. In the Borana zone no studies have been done on the collection of propolis. Moreover, the impact of propolis production on honey yield has not been assessed. Thus this study was initiated to enhance production of propolis through creating openings in MFH and basket hives and determine the effects of this propolis production technique on honey production.

## Materials and Methods

### *Description of the Study Area*

Borana zone is found between 30.36'- 60.38'N and 36.043'- 41. 040'E geographical grids and has altitude ranging from 1000m in the south to 1500 m.a.s.l in the northwest. The climate is generally semi-arid and rainfall pattern is bimodal with the 'ganna' or rainy season from mid-February to mid-May and the 'Hagaya', short rain season from September to the end of November (Yeneayehu F. and Tihunie F., 2020). The type of honeybee race that found in the district is *Apis mellifera Scutellata*. The traditional beekeeping is common and widely practiced method in this zone. There are two honey flow seasons in the region, at the end of Ganna (long rainy season), and at the end of Hagaya (the short rainy season).

### *Experimental setup*

The study was conducted at Elwoye district of Borana zone, Southern Ethiopia for three years (from 2018 to 2020). Sixteen honeybee colonies of *Apis mellifera scutelata*, 8 of which in moveable frame hive (MFH) and 8 in traditional/ basket hives were used. Four colonies in each hive type (MFH and basket hives) were induced for more propolis production by exposing to cold weather through creating more openings and compared with propolis production from eight colonies (four colonies in MFH and other four in basket hives) without induction or treatment, served as the control group. Colonies in the treatment groups, MFH were induced to collect more propolis by exposing them to cold air by inserting a 2cm thick wooden bar between the super and base and the lids and super to create openings and the entrances also fully opened. on other hand, in basket hives, cardboard was carefully cut to properly fit in one end of the basket hive and then about 1/4 of the cardboard was cut away and fitted to the hives entrance to be filled with propolis. The control hives (MFH and basket) left untreated. The propolis collected by bees in treatment groups and entrance and frame in control groups was harvested and the yield was recorded at the interval of three months over three years. Moreover the honey yield was also recorded throughout the study period. All recorded data was analysed using ANOVA. The model used to analyse the data was:

$$y_{ijkl} = \mu + HT_i + TRT_j + (HT * TRT)_k + \epsilon_{ijkl}$$

Where:  $y = ijkl^{\text{th}}$  observation,  $\mu$ =overall mean, HT =the effect  $i^{\text{th}}$  hive type on honey and propolis yield, TRT= the effect  $j^{\text{th}}$  treatment on honey and propolis yield, HT\*TRT= the effect  $k^{\text{th}}$  interaction on honey and propolis yield and  $\epsilon$ = random error

## Results and Discussion

### *Propolis production*

Data on propolis production is presented in table 1. The result showed that there was statistical significant difference between hive types and propolis production techniques (treatments) compared controls ( $P < 0.05$ ). Higher propolis yield was obtained from MFH ( $51.93 \pm 2.30$  g) as compared to that of basket hives ( $26.42 \pm 2.14$ ). Propolis ( $51.93 \pm 2.30$  g) collected from MFH over passed by more than one fold when compared to the one collected from basket hive. On the other hand, the finding obtained in this study showed that the mean propolis production figures 73.11g and 30.74 g for induced and non-induced respectively for moveable frame hive, while 35.20 g and 17.66 g for induced and non-induced respectively for traditional basket hive. Moreover, this study showed the overall mean propolis production from induced hives ( $54.16 \pm 2.07$ g) is significantly higher than and the amount of propolis ( $24.19 \pm 2.36$  g) collected from non-induced hives ( $P < 0.05$ ). Similarly, Nuru et al., (2002) discussed the propolis production in framed hives and local basket hives and reported that the mean production by *A. m. bandasii* bees in framed hives was  $24.2 \pm 22.5$  g and the same figure was  $12.7 \pm 8.6$  g in basket hives for 19 months. Moreover the production of propolis is highly affected by harvesting intervals, climatic condition, races, the strength of the bee colony, production methods and flora (Nuray S and Aziz G, 2005; Michael Clarke, 2019; Kiziltas and Erkan, 2020).

Table 1. Means with standard error of Propolis Yield  $g^{-1}$  and Honey Yield  $kg^{-1}$  (n=40)

Variables	Categories	Propolis yield (g $\pm$ SE)	Honey yield (Kg $\pm$ SE)
<b>Hives</b>	Basket	$26.42 \pm 2.14^b$	$3.25 \pm 0.53^b$
	Box	$51.93 \pm 2.30^a$	$12.08 \pm 0.57^a$
<b>Treatment</b>	Control	$24.19 \pm 2.36^b$	$7.61 \pm 0.51^a$
	Induced	$54.16 \pm 2.07^a$	$7.72 \pm 0.58^a$
<b>Treatment*Hive</b>	Control*Basket	$17.66 \pm 3.23^c$	$3.49 \pm 0.80^b$
	Induced*Basket	$35.20 \pm 2.80^b$	$3.00 \pm 0.70^b$
	Control*Box	$30.74 \pm 3.43^b$	$12.21 \pm 0.76^a$
	Induced*Box	$73.11 \pm 3.07^a$	$11.49 \pm 0.85^a$

### *Honey Production*

Honey production was significantly influenced by hive types, with the box hive producing on average 12.08 kg and 3.25kg honey basket type. However, in association to honey harvesting and propolis collecting methods, there was no significant difference ( $P < 0.05$ ) in honey production. This study is in agreement with Nuru, 2002 who reported that propolis production has no influence on honey production. Osipitan, 2012 found that the more space between top bar gives the higher propolis yield but lower ripe honey combs. The current result is found within the range of research findings by Jager (2001) and

contradicts with Mc Addam (2000) and Mann Lake Int (2000) cited in Jager (2001) who reported positive correlation between honey production and propolis productions.

### **Conclusions and Recommendations**

Generally, the current finding signified that the application of a propolis collecting technique into the hive increased the amount of propolis produced. Moreover, the amount of honey produced suggested as Propolis production did not adversely affect the quantity of honey produced over the study period. Therefore propolis production appears to be a worthwhile practice in the study area for the traditional beekeeper as the aggregate hive productivity will substantially increase without affecting honey production which the solo beekeeping product produced in the study area. Further study on propolis source identification, composition and quality of propolis is recommended. Awareness regarding, the value of bee products other than honey including propolis and the technique how to produce will help to substantially increase hive productivity.

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# Comparative Evaluation of Horizontal and Vertical Frame Hives at Adami Tulu Agricultural Research Center of Oromia, Ethiopia

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## Abstract

The sole purpose of a hive is to encourage the bees to build their nests in such a way that it is easy to manage and maintain them. The study was conducted at Adami Tulu Agricultural Research Center to evaluate the performances of honeybee colonies, honey yield and cost incurred in both horizontal beehives as compare to movable frame beehive. A total of 12 honeybee colonies (*Apis mellifera bandasii*) were established and assigned into four treatment groups. All the established honeybee colonies were managed in uniform manner until they are established properly and acquire uniform strength. Data on bee population, brood area, pollen and nectar stored areas were recorded using Liebefeld method (frame unit area, 10 x10 cm<sup>2</sup>) at every 21 days. In addition, data on average honey yield per harvest/colony, production costs and profit were recorded for each treatment during the study period. All the recorded data were organized by Microsoft excel and analyzed using descriptive statistical analysis of variance ANOVA of SAS software version 20. Results revealed that there was statistically significant difference ( $p < 0.05$ ) among Tensheratach beehive, Modern beehive and Bacho beehive with regarding to honey yield. The highest mean honey yield per hive ( $24.81 \pm 3.24$  kg/hive) was recorded from Tensheratach beehive followed by Modern beehive ( $21.51 \pm 2.36$  kg/hive) and Bacho beehive ( $17.3 \pm 1.43$  kg/hive). Significantly greatest adult bee population, brood area, pollen and nectar stored areas were also recorded from Tensharatech beehive compare to Modern beehive and Bacho beehive. The total costs of production and economic returns of Tensheratach hive were higher than Modern and Bacho beehives. Thus, it is possible to recommend that Tensheratach hive can be used as an alternative hive technology for honey production with full packages in addition to modern hive.

**Keywords:** Honeybee, hive type, brood area, production cost, honey yield

## Introduction

Beekeeping is one of the major incomes generating agricultural activity for rural households and other smallholder communities in Ethiopia. It provides substantial benefits to address household's food security and poverty alleviation through income diversification for beekeepers in potential areas (FAO, 2009). The existence of various agro-climatic zones resulted from the various topographic variations make the country suitable for many bee floras and huge number of bee colonies (Adgaba, 2007). For this reason Ethiopia is one of the top 10 producers of honey in the world (china, Turkey, United states, Ukraine, Argentina, Mexico, Russian Federation, Iran, Ethiopia, and Brazil) and it is the largest one in Africa (USAID, 2012). Despite of having the highest bee density and being the leading honey producer in Africa, the share of the sub-sector in the GDP has never been matched with the huge numbers of honey bee colonies and the country's potentiality for beekeeping. This is due to the traditional method of beekeeping practices which have great impact on the quantity and quality of honey.

Beekeeping has got many advantages for smallholder farmers of our country. It has great role in improving the well being of beekeepers. From the total honey produced in the country beekeepers earn about 360-480 million Birr per year (Nuru, 2002). The beekeeping farming is also important in creating job options in both rural and urban areas through organizing unemployed urban and landless rural youth and women to involve in them in beekeeping activities (Gemechis, 2015).

Ethiopian honey production is characterized by the widespread use of traditional technology, resulting in relatively low honey supply and poor quality of honey harvested when compared to the potential honey yields and quality gains associated with modern beehives. In Ethiopia, honey production remains traditional as 94 to 97% of bees are still kept in traditional hives (Kerealem *et al.*, 2009). Based on the stage of technical development, three different types of beehives have been used for honey production in Ethiopia. These are traditional beehives, transitional beehive and movable frame beehives. A total of about 4,601,806 hives exist in the country of which about 95.5% are traditional, 4.3 % transitional and 0.20% modern hives (Beyene and David, 2007). Based on the national estimate, the average yield of pure honey from movable frame hive is 15-20 kg/year and the amount of beeswax produced is 1-2% of the honey yield (Gezahegne, T, 2001). However, in potential areas, up to 50-60 kg of harvest has been reported (HBRC, 1997).

To improve the livelihoods of rural people in Ethiopia, large numbers of improved hive technologies are in the progress of distribution to beekeepers with rich experience in beekeeping. Movable frame hive was introduced to Ethiopia and being used for more than 50 years. Innovative beekeepers realized that this hive has a limitation like, while inspection, bees boiled out to the air as a result chance of being stung and consequently bee mortality are higher. Hussein Adam and Mohammed Fisaha beekeeper of Shashemene and Bacho district of Ilubabora zone respectively were modified the frame hive to minimize the problem prevailed in frame hive and they are using modified hives at present. The hives that these innovative beekeepers, Hussein Adam and Mohammed Fisaha have come up with are named Tensheratach and Bacho respectively. These hives are made of timber and have two compartments with two open doors, queen excluder and 25 rectangular frames. To improve productivity and production of honeybee products, selection and adoption of different improved beehives is highly essential. Though these modified beehives are being used by the owners, there was no study undertaken to evaluate the performances of these horizontal hives. Therefore this study was carried out to evaluate the performances of honeybee colonies and honey yield and cost incurred in both horizontal hives as compare to movable frame hive under Adami Tulu condition.

## **Materials and Methods**

### ***Description of the Study Area***

The study was conducted at Adami Tulu Agricultural Research Center (ATARC), located at 167 km South of Addis Ababa at altitude 1650 above sea meter, latitude 17°9' N and longitude 38°7'E. The average annual rain fall is 760.9 with an average minimum and maximum temperature of 12.6°C and 27°C respectively (ATARC, 1998). The soil type is fine, sandy loam with sand, silt and clay in the ratio of 34: 38: 18. The average pH of the soil in the area is 7.88.

### ***Experimental treatments***

Three beehive types: namely Modern beehive, Tansheratach beehive and Bacho beehive were used as treatments for evaluation purposes (Figure 1). Modern beehives (Zander model) were manufactured at Jimma Agricultural engineering Research Center. Tansheratach and Bacho hives were purchased from Shashemene and Ilu-Ababor respectively. A total of 12 honeybee colonies (*Apis mellifera bandasii*) were established and assigned into four treatment groups. All the transferred colonies were managed in uniform manner until they are established properly and acquire uniform strength. Each beehive type was replicated four times.

### ***Data collected***

Data such as adult bee population, brood area, pollen and nectar areas were recorded using Liebefeld method (frame unit area, 10 x10 cm<sup>2</sup>) at every 21 days (Keller, *et al*, 2005). Also, honey yield, production costs and economic return were taken. Before establishing of bee colonies, the size of bee space and hive entrance of each hive type was checked (Table1).

Table 1: The bee space and hive entrance dimension of three hive type

Type of hive	Bee-space (cm)	Hive entrance (cm)
Tensheratach	1	1x1
Modern	1	1x15
Bacho	1	1 x 12.3

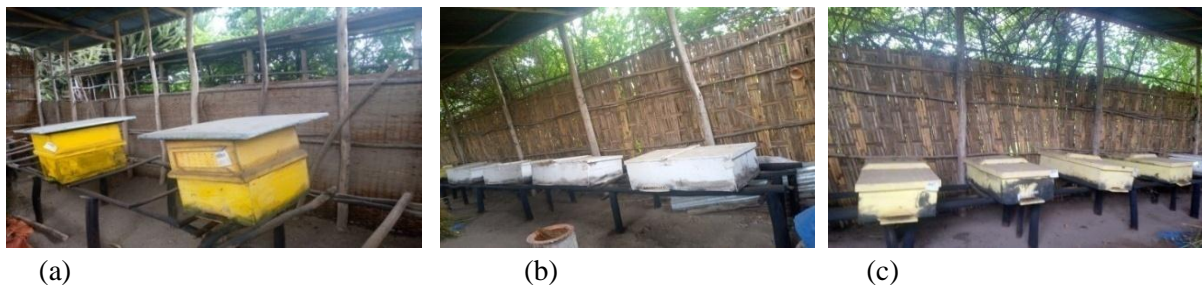


Fig. 1: Vertical and horizontal frame beehives used for comparison: a) Modern hive), b) Tensheratach hive, c) Bacho hive

### ***Rating the characteristics view of each hive type***

Characteristic view after three years of keeping different type of hives were scored as excellent, very good, good, fair and poor.

### ***Statistical Analysis***

All the collected data were organized by Microsoft excel and analyzed using descriptive statistical analysis of variance ANOVA of SAS software version 20. Means were separated by using Tukey's honest significant difference (HSD) at 5% level of significance whenever significant results encountered

between beehive types.

### ***Cost benefit analysis of hive types***

The most important issue of the present study was to determine the type of beehive with better profit for better life of small scale beekeepers. It is involved the calculation of variable costs and benefits. For the calculation of the variable costs, the expenditures incurred on various production cost of beehive, inputs for honey production, honeybee colony purchase and protective clothes were taken into consideration. Finally, the selling price for a kg of honey in local market was assessed in the study areas. Net incomes (NI) were calculated as the amount of money left when total variable costs (TVC) subtracted from the total returns (TR).

$$\text{NI} = \text{TR} - \text{TVC}$$

## **Results and Discussion**

### ***Honey yield performance of different hives***

The results of the study indicated that there was statistically significant difference ( $p < 0.05$ ) between Tensheratach hive, Modern hive and Bacho hive with regarding to the average of honey yield per colony. The highest mean honey yield ( $24.81 \pm 3.24$  kg/hive) was recorded from Tensheratach hive followed by Modern hive ( $21.51 \pm 2.36$  kg/hive) and Bacho hive ( $17.3 \pm 1.43$  kg/hive). This agrees with the study of Nuru, (2007); Workneh *et al.*, (2008) who reported that the average annual honey yield of improved frame hives at national level was 20-25 kg/hive but, lower than Awraris Getachew *et al.*, (2015) who reported that the average annual honey yield performance of improved frame hive was 30.09 kg/hive. The productivity of Tensheratach hive in this study is higher than that of Modern and Bacho hives. This could be due to the high bee population found in Tensheratach beehive than that of Modern and Bacho hives as the mean honey yield per colony is mostly proportional to the population size of colonies. Strong bee colonies rear more brood and produce more honey than weak colonies because of strong colonies make longer flights and bring back to the hive significantly bigger loads of nectar compared to forager bees from weak colonies. This agrees with the study of Neupane *et al.*, (2012) who reported that honey yield increases in line with the increasing number of bees in the colony relative to the amount of open brood. Szabo and Lefkovitch (1989) also reported that the production of honey is significantly positively correlated with the number of brood cells only in the first half of the season.

### **Colony evaluation parameters**

#### ***Worker brood area***

Sealed worker brood area noted after 21 days intervals is presented in table 2. The results of the study indicated that the brood rearing activity was statistically significant difference ( $p < 0.05$ ) between Tensheratach, Modern and Bacho hives (table 2). The highest mean brood area ( $150.21 \pm 0.6$ /colony) was recorded from Tensheratach hive followed by modern hive ( $128.57 \pm 2.4$ /colony) and Bacho hive ( $86.33 \pm 2.1$ /colony). Significantly higher mean of brood area produced in Tensheratach hive could be due to high bee population found in Tensheratach hive than that of Modern and Bacho hives. Strong honeybee colonies make longer flights and bring back to the hive significantly bigger loads of pollen and produce

more brood than weak colonies. This agrees with the study of Bhusal *et al.*, (2006) who reported that strength of bee colonies is significantly positively correlated with the amount of brood reared colonies. This result implies that a hive that encourages population growth will produce more brood and reduce honey consumption per bee during the dearth period (Szabo and Lefkovitch, 1989).

### ***Pollen area***

The result of the study indicated that there is statistically significant difference ( $p < 0.05$ ) between Tensheratach beehive, Modern beehive and Bacho hive. Significantly higher mean of pollen areas ( $112.35 \pm 6.1/\text{colony}$ ) were recorded from Tensheratach hive as compared to Modern hive ( $75.22 \pm 3.6/\text{colony}$ ) and Bacho hive ( $63.4 \pm 0.08$ ). Significantly higher mean area of capped brood in Tensheratach hive could be due to the large bee population found in the Tensheratach hive than that of Modern and Bacho hives. Strong honeybee colonies make longer flights and bring back to the hive significantly bigger loads of pollen and more. This agrees with the study of Shower *et al.*, (1986) who reported that pollen collected by strong colonies was higher.

### ***Nectar area***

Nectar is an aqueous solution secreted from floras of plants profoundly containing sugars mainly glucose, fructose and sucrose with traces of minerals and proteins. The result of the study indicated that there was no statistically significant difference ( $P > 0.05$ ) between Tensheratach hive and Modern hive in nectar area. But there was statistically significant difference ( $P < 0.05$ ) between Modern hive and Bacho hive. Strong and healthy bee colonies stored more nectar as compared to weak colonies.

### ***Adult bee population***

The highest mean of adult bee populations ( $9.54 \pm 2.4$ ) was found in the Tensheratach hive as compared to Modern hive ( $6.82 \pm 0.7$ ) and Bacho hive ( $4.27 \pm 4.3$ ). The reason for higher number of worker bees in Tensheratach hive could be explained by the relatively high mean sealed brood area because the sealed brood represents the next population of workers. This agrees with the study of Harbo *et al.*, (1993) that showed that positive correlation was found between colony populations and sealed brood area.

Table 2: Mean of honey yield, adult bee population, brood area, pollen and nectar stored areas of different hive types

Hive type	Mean $\pm$ SD				
	Honey yield (kg/hive)	Brood area (cm <sup>2</sup> )	Pollen area (cm <sup>2</sup> )	Nectar area (cm <sup>2</sup> )	No of comb covered by bees
Tensheratach	24.82 $\pm$ 3.24 <sup>a</sup>	150.21 $\pm$ 0.6 <sup>a</sup>	112.31 $\pm$ 6.1 <sup>a</sup>	125.23 $\pm$ 4.6 <sup>a</sup>	9.54 $\pm$ 2.4 <sup>a</sup>
Modern	21.51 $\pm$ 2.36 <sup>b</sup>	128.57 $\pm$ 2.4 <sup>b</sup>	75.22 $\pm$ 3.6 <sup>c</sup>	113.15 $\pm$ 1.3 <sup>a</sup>	6.82 $\pm$ 0.7 <sup>b</sup>
Bacho	17.3 $\pm$ 1.43 <sup>c</sup>	86.33 $\pm$ 2.1 <sup>c</sup>	63.4 $\pm$ 0.08 <sup>b</sup>	87.32 $\pm$ 0.5 <sup>c</sup>	4.27 $\pm$ 4.3 <sup>c</sup>

Means in a column having different superscript are statistically different at  $p < 0.05$

Table 3. Characteristics observed after three years of keeping the hive types

Parameters	Hive type			Observed characteristics
	Tensheratach	Modern	Bacho	
Simplicity of hive management practices	5	5	5	<ul style="list-style-type: none"> <li>• Suitable for colony inspection and honey harvest</li> <li>• Easy to control swarming</li> <li>• Possible to manage the volume of the hives according to the strength of colonies</li> <li>• Bee breeding and queen rearing is possible</li> <li>• Can be transported with bees from one place to another for migratory beekeeping practices.</li> </ul>
Minimization of honeybee mortality rate	5	3	5	<ul style="list-style-type: none"> <li>• Tensheratach and Bacho hives are made of timber and have two compartments with two open doors</li> <li>• While inspection and honey harvesting, honeybee colony moved from first compartment to second compartment and return back to the first compartment</li> <li>• Honeybee get enough space without moving outside</li> <li>• Bees could not be boiled out to the air, as a result chance of being stung and bee mortality are lower than modern hive</li> </ul>
Improving of ventilation	5	4	3	<ul style="list-style-type: none"> <li>• Tensheratach and Modern hives are better to emit heat and moisture to rise up and out of the hive</li> </ul>
Minimization of the infestation rate of pests and predators	4	2	2	<ul style="list-style-type: none"> <li>• Tensheratach hive has small hive entrance than Bacho and Modern hives</li> <li>• This reduces colony invasion by pests and predators which adversely affects all aspects of beekeeping</li> </ul>
Simplicity of honey harvesting	5	5	5	<ul style="list-style-type: none"> <li>• Easy to harvest honey</li> <li>• Honey extractor can be used without damaging the frame combs</li> <li>• Pure and standard honey can be harvested</li> </ul>
Hive preference	5	4	3	<ul style="list-style-type: none"> <li>• Colonies show better preference for Tensheratach hives and this could be due to the insulation nature of the hive</li> </ul>

**Score given:** Excellent=5, Very good=4, Good=3, Fair=2, Poor=1

### *Costs and return of hive type*

The production costs and gross return of Tensheratach hive stands first when compared to other hive types (Tables 4 & 5). The net income per beekeeper own 4 hives of Tensheratach hive, Modern hive and Bacho hive was 7840 ETB, 5930 ETB and 1920 ETB respectively.

Table 4: Presentation of production costs of each hive type

Major items	Hive type		
	Tensheratach	Modern	Bacho
Beeswax (Kg)	4500	4500	4500
Overall	1000	1000	1000
Smoker	200	200	200
Hand glove	180	180	180
Bee veil	300	300	300
Battery	100	100	100
Bee colony	2400	2400	2400
Feeding	300	300	300
Hive	8000	5900	6400
Total production cost	16980	15580	15380

Table 5: Total production costs and economic return from the different hives

Hive type	Total production cost (ETB)	Gross return (ETB)	Net income per beekeepers (ETB)*	Net income per hive (ETB)
Tensheratach	16980	24820	7840	1960
Modern	15580	21510	5930	1483
Bacho	15380	17300	1920	480

\*= The price of pure honey per kg was estimated at 250.00 Birr

### **Conclusion and Recommendations**

The present results indicated that Tensheratach hive had better performance in terms of honey yield per hive, workers sealed brood area, pollen and nectar stored area, adult bee population and gross return as compared to Modern hive and Bacho hive. Therefore, it is recommended to use Tensheratach beehive as an alternative technology with full packages in addition to Modern beehive. Bachohive has some drawbacks from the scientific perspective; it is therefore recommended that, before promoting the Bacho hive for wider uses, some modifications should be made to remove its shortcomings and, then, it should be tested with more replications in different areas so as to allow a better understanding and evaluation of the hive. Hive types have a great impact on colony performance and honey yield. Thus, future research should look in to evaluate any new beehive type using several parameters and different bee races.

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# Diagnostic Survey of Honey Bee Diseases and Pests in Guji zone of southern Oromia, Ethiopia

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## Abstract

*Apiculture is among the vibrant agricultural enterprise practiced throughout the Ethiopia. It has multi faceted advantages and plays important role in increasing the productivity of food and cash crop and conservation of natural resources through pollination. Furthermore it is a means of income for landless and people living at harsh areas and it also contributes in poverty reduction and foreign currency earnings. Diseases and pests of honeybees cause the death of honeybees and subsequent reduction of honeybee products and more over have negative impact on domestic and international marketing of honeybee products (van Engelsdorp and Meixner, 2010). Apiculture or beekeeping is well known and well practiced traditionally in Guji zone of southern Oromia of Ethiopia. However, the research activity concerning apiculture is on its infant stage and no research is done on honeybee diseases and pests in the area. Diagnostic survey was conducted in Guji zone of southern Oromia in 2021 to generate baseline data on the status of economically important honeybee diseases and pests in Guji zone of southern Oromia. Three districts were selected purposively, based on their honey production potential and representativeness of agro-ecology. From each districts three representative PAs were randomly selected from which three beekeepers again randomly selected. From the selected beekeepers apiary 3 to 5 honeybee colonies were randomly selected for diagnostic assessment. Twenty- eight (28) samples of brood and adult bees were collected from different apiary site randomly; field observation and laboratory analysis was undertaken to identify the prevalence of diseases and pests. 45 beekeepers were interviewed on pre-structured questionnaire developed to generate appropriate information related to honeybee diseases and pests. Furthermore, we purposively selected five (5) experienced beekeepers from each Kebele/PA and held discussion with them on the prevalence and economical importance of honeybee diseases and pests., 86.67% of respondents keep honeybee colonies in traditional hive, while 4.44% and 8.89% use traditional, both transitional, and modern hive respectively. Honeybee eater birds, Honey Badger, Wax moth, Ants, Small hive beetle, large hive beetle, Bee lice, Monkey, Wasps, and Spiders were prevalent in the area. Furthermore, about 53.57%, 64.29% and 17.86% colonies were infected with Nosema, Amoeba and Chalk brood at a time respectively. The prevalence of varroa mites in the area was 57.14% (16/28). Effective honeybee colony health management is suggested to minimize or protect the effect of these honeybee diseases and pests.*

Keywords: *Bee diseases, Bee pests, Prevalence, Diagnostic survey*

## Introduction

Beekeeping is among the vibrant agricultural enterprise practiced throughout the Ethiopia. It has multifaceted advantages and plays important role in increasing the productivity of food and cash crops and conservation of natural resources through pollination. Furthermore it is a means of income for

landless and people living at harsh areas. Beekeeping also contributes in poverty reduction and foreign currency earnings through marketing of honeybee products.

Diseases and pests of honeybees are major cause for the death of honeybees and subsequent reduction of honeybee products and negatively affect domestic and international marketing of honeybee products (van Engelsdorp and Meixner, 2010). This is because, honeybees have more than frequent contact among themselves and thus, it almost inevitable that any contagious disorder will spread within the hives or other colonies unless it is detected and appropriate treatment is taken as early as possible (Morse, 1990;FAO, 2006 ). Honeybee diseases and pests are major driving force for colony losses in most parts of the world. Serious decline of honeybee populations in Europe and the USA commonly referred the colony collapse disorder (CCD), is mostly associated to honeybee diseases and pests. This disorder also seriously affects the production, quality, safety and marketing of the honeybee produces. In 2010, the USDA reported that overall honeybee losses for the year was 34%, which is statistically similar to losses reported in 2007, 2008, and 2009 (USDA, 2010).

This threat is an alarm for governments, conservationists and the private sector engaged in the subsector in different parts of the world. As honeybee, diseases and pests do not respect borders, similar decline in Africa in general and Ethiopia in particular may happen and would seriously harm the livelihoods of millions of rural resource-poor farmers, processors and exporters. Therefore, early detection and monitoring of honeybee diseases and pests are important to minimize the effects.

The major honeybee diseases and pests associated to honeybees in Ethiopia include Nosema and Amoeba, Chalk brood, Parasitic mite (varroa), Small and Large hive beetles, Wax moth, Ants, Wasps, Death Head Hawk moth, Bee-eater bird, Honey badger, Frog and Toads (Amssalu et al, 2012; Dessalegn, 2015). Although, beekeeping is one of agricultural practice Guji zone of southern Oromia, data on bee health is scars. Therefore, this study initiated to generate baseline data on the status of economically important honeybee diseases and pests in Guji zone of southern Oromia.

## **Materials and Methods**

### ***Study Area and sampling techniques***

The study was conducted in three districts; Anna Sorra, Adola and Gorodola of Guji zone. Guji zone is located at the distance of 385 km south of Finfine, the capital city of Ethiopia. The study districts were selected purposively based on their honey production potential and representativeness of agro-ecology (lowland, midland and highland). From each districts, three representative PAs also selected purposively. On other hand, three bee colonies randomly selected from each PA for internal and external diagnosis to determine the occurrence, infection/ infestation of honeybee diseases and pests. Twenty-eight samples of brood and adult bees collected from selected honeybee colonies and tested for different honeybee diseases and pests. Forty-five beekeepers interviewed on pre-structured questionnaire to generate information related to honeybee diseases and pests. Focused group discussion held with five experienced beekeepers at each PA to support quantitative data with qualitative data.

The Honeybee colony samples required for the diagnosis was determined using Thrusfield method (2005).

$$N = \frac{1.96^2 \times P_{exp} \times (1-P_{exp})}{d^2}$$

Where, n = required sample size Pexp= Expected prevalence (50 %) d= Desired absolute precision (5 %).

### *Diagnosis for major honeybee diseases*

Field observation and laboratory diagnosis were conducted to identify major honeybee diseases including Nosema, Amoeba, Chalk brood, American foul brood, European foul brood (where there is suspected clinical symptoms) following standard methods of respective honeybee diseases

### **Result**

#### *Educational level, Marital Status, Sex and Age of Respondent beekeepers*

Beekeeping seems the work of men in the study area and almost no women participation in beekeeping activity. The age of beekeepers fall between 22 to 55 years. Regarding marital status, 97.78% beekeepers were married and 2.22% beekeepers were single (youth).The educational level of respondents (beekeepers) is summarized in the (table 1). The marital status, age and sex of respondent beekeepers also explained in the (table 2).

**Table 1: Educational level of respondent Beekeepers, Sample size = 45**

<b>Educational level</b>	<b>Frequency</b>	<b>Percentage</b>
Illiterate	9	20.00
Read and Write	8	17.78
Elementary (1-4)	22	48.89
Elementary (5-8)	5	11.11
Secondary school	0	0.00
Preparatory school	1	2.22
College and above	0	0.00

**Table 2: Marital Status, Age and Sex of respondents, Sample size =45**

<b>Marital status</b>	<b>frequency</b>	<b>percentage</b>
Married	44	97.78
Unmarried	1	2.22
<b>Age</b>	<b>frequency</b>	<b>Percentage</b>
22-32	1	2.22
33-43	16	35.56
44-55	28	62.22
<b>Sex</b>	<b>frequency</b>	<b>Percentage</b>
Male	45	100.00
Female	0	0.00

### ***Beekeeping Practice in Guji Zone***

Beekeeping is the inherited agricultural activity from father to offspring or from generation to generation in East Guji zone. Beekeeping is not new trend and rather it is strongly linked to the life of the community in the area. However, up to date Beekeeping technology is on its infant stage and the majority of the community practice beekeeping in traditional way. As the result obtained from respondents and observation, 86.67% traditional hive, 4.44% all traditional, transitional, and modern hive and 8.89% beekeepers were using both traditional and modern hive (table 3).

**Table 3: Types of Hive beekeepers use in the area**

<b>Sample size = 45</b>		
<b>Types of Hives</b>	<b>Frequency</b>	<b>Percentage</b>
Traditional hive (only)	39	86.67
All traditional, transitional and Modern hives	2	4.44
Both transitional and modern hives	4	8.89



**Figure 1:- Traditional hive**

### ***The Major Factors to Decrease Bee colony and Honey Yield***

All (100%) respondents reported that honeybee colony number and honey production yield were on decreasing rate in the area by different causes. Total death of bee colony was occurred widely at the highland area of the zone in the winter (Gana) season in August 2021. The cause of total death of bee colony was reported as extreme cold with fog stayed for a long time and toxic plant. There are many factors to decrease the number of honeybee colony and honey yield. The major factors found in the area are summarized in the (table 4).

**Table 4: Response of beekeepers on the number of bee colony and Honey yield of the area, Sample size =45**

<b>Number of bee colonies and honey yield</b>	<b>frequency</b>	<b>percentage</b>
Decreasing	45	100.00
Increasing	0	0.00

**Table 5: Causes for the decline of honeybee colonies and honey yield**

<b>Causes</b>	<b>Sample size</b>	<b>Frequency</b>	<b>Percentage</b>	<b>Rank</b>
Deforestation/expansion of farm land	45	40	88.89	1 <sup>st</sup>
Pests and predators	45	34	75.56	3 <sup>rd</sup>
Lack of improved beekeeping skill	45	25	55.56	4 <sup>th</sup>
Drought at lowland area of the area	45	15	33.33	7 <sup>th</sup>
Pesticides and herbicides application	45	35	77.78	2 <sup>nd</sup>
Extreme cold of highland area of the area	45	16	35.56	6 <sup>th</sup>
Death of colony	45	7	15.56	8 <sup>th</sup>
Lack of bee forage	45	5	11.11	9 <sup>th</sup>
Absconding	45	17	37.78	5 <sup>th</sup>

### ***Diagnosis for Honey Bee Pests and Predators***

Pests and predators of honeybees are cause devastating damage on honeybee colonies and at most time cause swarming, abscond or colony collapse so that a challenge for beekeeping in tropical and sub-tropical countries.

The occurrence and economic importance of major honeybee pests (including: *Wax moth, small hive beetle, ants, spiders, bee-eater birds, honey badger, bee lice, lizards, Dead hawks moth ..Etc*) in the all the study areas will be determined through beekeepers interview using semi-structured questionnaires and internal and external hive inspections. Moreover, clinical symptoms and infested combs, adult and larvae of small hive beetles and wax moth and other decayed materials were observed in the hive through inspection of the beehives described by Neumann *et al.* (2013).

The presence of small hive beetle infestation (*Aethinatumida*) was identified through its adult, larvae or pupae and colony examination methods as Larvae of SHB have pairs of prominent brownish dorsal spines on each segments with 3 pairs of anterior prolegs only. Based on Ellis *et al.* (2013),the larvae of wax moth has no spines, but number of setae (hairs) on each segments with 8 pairs of prolegs (3 pairs,4 pairs and 1 pairs on anterior, abdominal and last segments respectively).Unlike Small hive beetles, it produces silken galleries.

Diseases and pests of honeybees are known to cause the death of honeybees and subsequent reduction of honeybee products and more over have negative impact on domestic and international marketing of honeybee products (van Engelsdorp and Meixner, 2010). The community believes that there are not honeybee diseases as reported from respondents in east Guji zone. However, the respondents reported that the existenceof different honeybee pests and predators. From these pests and predators, Ants are reported as the major economic important pest. The presence of Honeybee eater birds, Honey Badger, Wax moth, Ants, Small hive beetle, Hive beetle, Bee lice, Monkey, Wasps, and Spiders were identified by observation and Beekeeper reaction.Honeybee pests and predators in the area are ranked based on their economic importancein the(table 4). There are different control mechanisms of pests and predators as reacted by beekeepers. The control mechanisms were summarized in the (table 5). However, some pests such as Bee lice, Hive beetle and small hive beetle reported as that they have no control method.

Table 6: Beekeeper response regarding prevalence of Honeybee pests and predators and their Economic importance, Sample size=45

<b>Pests/predators</b>	<b>prevalence</b>	
	<b>Frequency</b>	<b>percentage</b>
Honeybee eater birds	28	62.22
Wax moth	5	11.11
Ants	45	100.00
Honey Badger	19	42.22
Hive Beetle	32	71.11
Small hive beetle	5	11.11
Spiders	37	82.22
Wasps	21	46.67
Monkey	45	100
.Bee lice/Mites	2	4.44

	<b>Economic Importance of pests/predators</b>		
	<b>Frequency</b>	<b>Percentage</b>	<b>Rank</b>
Honeybee eater birds	33	73.33	2 <sup>nd</sup>
Wax moth	3	6.67	9 <sup>th</sup>
Ants	45	100.00	1 <sup>st</sup>
Honey Badger	10	22.22	4 <sup>th</sup>
Hive Beetle	4	8.89	7 <sup>th</sup>
Small hive beetle	5	11.11	6 <sup>th</sup>
Spiders	32	71.11	3 <sup>rd</sup>
Wasps	1	2.22	10 <sup>th</sup>
Monkey	7	15.56	5 <sup>th</sup>
Bee lice	2	4.44	8 <sup>th</sup>

Table 7:- Pests and predators traditional control method in the area

<b>Pests/Predators</b>	<b>Traditional Control methods</b>
Honeybee eater birds	-Keep and chase away from the apiary
Wax moth	-cleaning the hive and apiary site
Ants	-Cleaning the apiary, burning the ants nest, cleaning the ants by bad odor plants
Honey Badger	-Attaching smooth metal and thorny plants up on the tree which hive hanged over, hanging metal/tin, which can make sound in the apiary site to chase the honey badger
Hive Beetle	-----
Small hive Beetle	-----
Spider	-Cleaning the apiary and hive, removing its nest
Wasps	-Removing the nest, cleaning the apiary site and hive
Monkey	-keeping and chasing away
Bee lice	-----

During diagnosis of twenty-eight colonies randomly selected, bee lice were infested two colonies with high infestation. The number was estimated to be more than 450 and 300 in the colonies. The colonies in

which bee lice found were very poor and its production feat was very poor. The prevalence of bee lice, SHB and LHB was 2/28 (7.14%), 7/28 (25.00%) and 8/28 (28.57%) respectively. Wax moth was found only in the honeybee absconded hive.

Table 8: prevalence of bee lice, SHB and LHB in the study area in 2021, Sample size=28

<b>Districts</b>	<b>Pest</b>	<b>Infested colony(+ve)</b>	<b>NP/colony</b>	<b>Free colony(-ve)</b>	<b>Prevalence</b>
Gorodola	Bee lice	0	0(0.00)	10	0.00
Adola		1	450(50.00) (est.)	8	11.11
Anna Sorra		1	300(33.33) (est.)	8	11.11
<b>Total</b>		<b>2</b>	<b>800(28.57) (est.)</b>	<b>26</b>	<b>7.14</b>
Gorodola	LHB	0	0(0.00)	10	0.00
Adola		3	9(1.00)	6	33.33
Anna Sorra		4	15(1.67)	5	44.44
<b>Total</b>		<b>8</b>	<b>24(0.86)</b>	<b>20</b>	<b>28.57</b>
Gorodola	SHB	0	0(0.00)	10	0.00
Adola		2	17(1.89)	7	22.22
Anna Sorra		5	38(4.22)	4	55.56
<b>Total</b>		<b>7</b>	<b>55(1.96)</b>	<b>21</b>	<b>25.00</b>

Where, NP=Number of pest, LHB=Large hive beetle, SHB=small hive beetle, est. =estimation



Figure 2:- Wax moth found in beehive



Figure 3: Image of Hive beetle taken during diagnosis



## Diagnosis of Ecto-parasites

### 1, Varroa Mites

Varroa mite (*Varroa destructor*) is known to be one of the most serious ecto-parasites of *Apis mellifera*. Varroa mite feeds on the haemolymph of brood and adult honeybees causing colony disorder, weakness, decrease in brood and deformation of bees (Cornman et al., 2012). The mite also considered as a vector for some honey bee viruses such as deformed wing virus (DWV) (Boecking and Genersch, 2008). Brood examinations were done by cutting off 5 X 5 cm brood comb areas from drone and/or worker pupae. About 100 pupae were removed from their cells using forceps and checked for the presence of varroa mites. The numbers of varroa mites found on the body of pupae and in the brood cell were collected and counted. To examine the presence and infestation level of varroa mite in honeybees, the standard *icing sugar-rolling* method was followed according to Dietemann et al. (2013). Samples of 250-300 adult bees were collected from randomly selected colonies and brushed off directly into a wide mouth jar with the #8-mesh lid. To remove varroa mites from the body of adult honeybees, different methods such as: sugar powder, alcohol and hot water were used. From the samples examined or analyzed the prevalence of varroa mite was 57.14%. The range of varroa mites extracted from the adult honeybees was 0 to 75 per 250 and for pupae (brood) was 0 to 37 per 100 cells. The result shown infestation of varroa mites on adult honeybees was 4.41% and 9.96% on pupae or brood across the study area. Varroa mite infestation was high at lowland area of the study area and even no free colony from varroa mites. Data collected had already analyzed (summarized) as shown in the following (table 9).

Table 9: Infestation and prevalence of Varroa mites in the study area in 2021

Districts	sample	NVM/100 pupae (cell)	NVM/250 adult bees	Prevalence
Gorodola (lowland)	1	12(0.12)	57(0.228)	+
	2	10(0.10)	33(0.132)	+
	3	29(0.29)	30(0.120)	+
	4	15(0.15)	5(0.020)	+
	5	10(0.10)	11(0.044)	+
	6	9(0.09)	6(0.024)	+
	7	7(0.07)	23(0.092)	+
	8	13(0.13)	1(0.004)	+
	9	22(0.22)	8(0.032)	+
	10	21(0.21)	45(0.180)	+
<b>Sub Total (%)</b>	<b>10</b>	<b>148(14.80)</b>	<b>219(8.76)</b>	<b>100</b>
Adola(midland)	11	32(0.32)	2(0.008)	+
	12	12(0.12)	1(0.004)	+
	13	0.00	0.00	-
	14	0.00	0.00	-
	15	0.00	0.00	-
	16	0.00	0.00	-
	17	37(0.37)	3(0.012)	+
	18	0.00	0.00	-
	19	0.00	0.00	-

<b>Sub Total (%)</b>	<b>9</b>	<b>81(9.00)</b>	<b>6(0.27)</b>	<b>33.33</b>
Anna Sorra	20	19(0.19)	75(0.30)	+
(Highland)	21	0.00	0.00	-
	22	17(0.17)	5(0.02)	+
	23	0.00	0.00	-
	24	0.00	0.00	-
	25	0.00	0.00	-
	26	0.00	0.00	-
	27	0.00	0.00	-
	28	14(0.14)	31(0.124)	+
<b>Sub Total (%)</b>	<b>9</b>	<b>50(5.56)</b>	<b>111(4.93)</b>	<b>33.33</b>
<b>Overall Total (%)</b>	<b>28</b>	<b>279(9.96)</b>	<b>336(4.41)</b>	<b>57.14</b>

Where, NVM = Number of Varroa Mites, += infested colonies, - = varroa mite free colonies, 28=sample size, Infested colonies=16, Sample size=28

$$\text{Prevalence} = \frac{\text{Infested colonies}}{\text{Total sample size}} \times 100 = \frac{16}{28} \times 100 = 57.14$$



**Figure 4:- Image of Varroa mites taken during diagnosis**

## **2, Protozoan Diseases (Nosema and Amoeba)**

The same method was used to identify both nosema and amoeba infection. Thirty (30) adult bees were collected and abdominal part of the bees was removed from the other body part. Then the removed abdominal part were crushed or grinded in the mortar by pestle. A wet mount will be prepared from the resulting suspension on microscopic slide with cover slips and examined under the light microscope for the presence of the spores with magnification power of 40X. The presence of slippery and rod shaped spores indicates the detection of Nosema and the presence of round cysts and spore balls indicates the detection of Amoeba diseases in the honeybees. In this area, 53.57% and 64.29% colonies were infected with Nosema and Amoeba respectively.

Table 10: Prevalence of protozoan diseases in the study area in 2021

<b>Districts</b>	<b>Number of sample</b>	<b>Nosema (-Ve)</b>	<b>Nosema (+ve)</b>	<b>Amoeba (-ve)</b>	<b>Amoeba (+ve)</b>
Gorodola	10	9(90.00%)	1(10.00%)	4(40.00%)	6(60.00%)
Adola	9	6(66.67%)	3(33.33%)	5(55.56%)	4(44.44%)
Anna Sorra	9	2(22.22%)	7(77.78%)	1(11.11%)	8(88.89%)
<b>Total (%)</b>	<b>28</b>	<b>13(46.428%)</b>	<b>15(53.57%)</b>	<b>10(35.71%)</b>	<b>18(64.29%)</b>

### 3, *Acarine Disease*

For the examination of tracheal mites, samples of 30 adult honeybees were collected randomly and examined according to Sammataro *et al.*, (2013). The sample bees were preserved by adding 70% alcohol during sample collection. The head and first pair of legs of the bees were removed using forceps and scissor. Transverse-section thoracic disks were sliced and placed directly in a small dish containing 10-percent potassium hydroxide (KOH). The sliced thoracic disks in KOH were stayed for 10 minutes until the soft internal tissues dissolved to expose trachea rings. The trachea ring sections was retrieved through filtration and washed with tap water. Then the disk- trachea suspension was examined for infested part and *Acarapis wood* under a dissecting microscope at 10x power. From 28 samples examined, the result shown that acarine diseases infected 7.14% (2/28) colonies.

Table 11: Prevalence of Acarine Disease (Acarapis Wood)

<b>Districts</b>	<b>Disease</b>	<b>Number of sample</b>	<b>Percentage</b>
Gorodola	Acarapis Positive	0	0(0.00%)
	Acarapis Negative	10	10(100.00%)
Adola	Acarapis positive	0	0(0.00%)
	Acarapis Negative	9	9(100.00%)
Anna Sorra	Acarapis positive	2	2(22.22%)
	Acarapis Negative	7	7(77.78%)
<b>Total (%)</b>	<b>Acarapis positive</b>	<b>2</b>	<b>2(7.14%)</b>
	<b>Acarapis Negative</b>	<b>26</b>	<b>26(92.86%)</b>

### 4, *Fungal diseases (Chalk Brood) and Bacterial Diseases (AFB and EFB)*

Both external and internal inspection was conducted for the presence of chalk brood clinical symptoms. Dry scales with white to dark color moulds and chalk brood mummies were carefully observed in the comb cells and on the bottom boards of the hives. From selected apiaries about three and above colonies were randomly inspected internally for major clinical symptoms of bacterial diseases with emphasis to AFB and EFB. Typical clinical symptoms such as irregular brood arrangement, sunken and dark capping with puncture holes, dead and decayed larvae with dark “scales” and slight to pronounce odor was examined for the occurrence of AFB in the colonies. Similarly, twisted larvae with creamy-white guts visible through the body wall, melted and yellow white larvae with unpleasant sour odour and loosely attached brown scales were directly observed for the occurrence of EFB in the colonies. The result of observation and inspection was indicated that chalk brood infected 17.86% colonies and no colony infected by bacterial diseases (AFB and EFB) at all districts.

Table 12: Prevalence of Fungal and Bacterial Diseases

Districts	Diseases (+ve or -ve)	Number of Sample	Percentage
All	AFB Positive	-	0.00
	AFB Negative	28	100.00
All	EFB positive	-	0.00
	EFB Negative	28	100.00
Gorodola	Chalk Brood Positive	3	30.00
	Chalk Brood Negative	7	70.00
Adola	Chalk Brood Positive	0	0.00
	Chalk Brood Negative	9	100.00
Anna Sorra	Chalk Brood Positive	2	22.22
	Chalk Brood Negative	7	77.78
<b>Total (%)</b>	<b>Chalk Brood Positive</b>	<b>5</b>	<b>17.86</b>
	<b>Chalk Brood Negative</b>	<b>23</b>	<b>82.14</b>



Figure 5:- Fungal diseases image taken during diagnosis

### Conclusion and Recommendation

Apiculture or beekeeping is well known and well practiced traditionally in Guji zone of southern Oromia. Beekeeping is the inherited agricultural sector from father to offspring or from generation to generation-in East Guji zone. Beekeeping is not new trend and the community life of the area is related to the activity strongly. However, up to date Beekeeping technology is on its infant stage. Majority of the respondents were using traditional hive for honey production. As the result obtained from respondents and observation, 86.67% traditional hive, 4.44% all traditional, transitional, and modern hive and 8.89% beekeepers were using both traditional and modern hive. All (100%) respondents reported that honeybee colony number and honey production yield were on decreasing trend in the area by different causes. These factors or causes were deforestation, use of pesticide and herbicide, extreme cold at highland, drought at lowland, lack of skill, absconding, diseases (Nosema and amoeba, chalk brood), honeybee pests and predators such as Ants, honeybee eater birds, bee lice/mites, honey badger, hive beetle, small hive beetle, wax moth and monkey. Total death of bee colony was occurred in the highland area of the zone in the August. The cause of mass death of bee colony was reported as extreme cold with fog stayed for long time and this made honeybees starve, lack of feed (dearth period) and toxic plants.

Therefore, in order to increase the production and productivity of Apiculture sector honeybee forage or forest should be preserved. Beekeepers should be able to use beekeeping up to date technology and practice effective management. It is also better to develop protection mechanism for different diseases, pests and predators of honeybees. In addition, during dearth period, supplementary feed should be given to honeybee colonies, pesticides and herbicides should be used wisely at the right time, and apiary site should be far away from farmland.

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# **Fishery Research Results**

# Assessment of Fish Production Potential in Four Bishoftu Crater Lakes, Bishoftu, E.Shoa Zone of Oromia

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## Abstract

*Bishoftu Crater Lakes are economically important lakes in the country. However, the Physico-chemical parameters of the lake seem to be threatened by anthropogenic, which in turn affect biotic factors as reflected in fish catch. The physico-chemical characteristics, fish species composition and production, and the constraints that affect the fisheries production were studied in Bishoftu crater lakes from July 2018 - June 2021. The temperature of the lakes ranged from  $20.06 \pm 0.3$  °C (Hora-Kilole) to  $21.3 \pm 1.5$  °C (Hora-Arsedi) whereas pH ranged from  $8.4 \pm 0.8$  (L. Koriftu) to  $9.1 \pm 0.1$  (Hora-Kilole). Dissolved oxygen varied from  $6.4.31 \pm 0.8$  to  $7.9 \pm 0.9$  mg/l. Low dissolved oxygen was measured at Lake Koriftu categorized as heavily impacted sites. Conductivity varied from  $345.1 \pm 2.4$  in L. Koriftu to  $967 \pm 36$   $\mu$ S/cm (L. Babogaya). Soluble Reactive Phosphate varied from  $39.36 \pm 0.3$  (H. kilole) to  $51.13 \pm 5.2$   $\mu$ g/l (Koriftu) and nitrite ranged from  $31.2 \pm 5.4$  (Babogaya) to  $44.05 \pm 0.6$   $\mu$ g/l (Koriftu). A total of five species of fish, belonging to three families were identified. The species were *Cyprinus carpio*, and *Labeobarbus intermedius* from the Family Cyprinidae, *Clarias gariepinus* from the Family Clariidae, and *Oreochromis niloticus* and *Coptodon zillii* from the Family Cichlidae. *O. niloticus* and *C. zillii* were more abundant in all lakes as compared to other fish species. The estimated mean annual catch in tons were 2.3, 2.8, 1.1, and 1.6 for Lake H. kilole, H. Arsedi, Koriftu, and Babogaya, respectively. Open access to the resource and pollution were the major problems in the lakes. Thus, management tools like mesh size regulations, gear restrictions and limits on the number of fishers for sustainable exploitation of the stocks and developing aquaculture technologies like cage culture in the lakes are recommended.*

**Keywords:** Bishoftu Crater Lakes, Fish catch, Fishery constraints

## Introduction

Ethiopia is a land-locked country that depends on the inland waters for the supply of fish as a cheap source of animal protein. The country's water bodies have an estimated surface area of 7,334 km<sup>2</sup> of major lakes and reservoirs, and 275 km<sup>2</sup> of small water bodies, with 7,185 km of rivers within the country (FAO, 2015). According to different sources of information, the bulk of the fish catch (approximately 75 percent of the total) originates from the six main water bodies: Tana, Ziway, Langano, Awasa, Abaya and Chamo. The remaining production (approximately 25 percent) originates from minor lakes (Hora, Beseka, Ligo, Hyke, Hashengie, and Small Abaya), reservoirs and dams (Koka, Fincha-Amerti, Denbi, Melka-Wakena, Alwero, Tekezé, Gigel Gibe I) and rivers include Abay, Wabi-shebelle, Awash, Genale, Dawa, Omo, Tekezé, Gibe, Mereb, Baro, Akobo, Angereb and their tributaries (Moreau and Scullion in ACP Fish II, 2013). Riverine fishing activities are mostly performed on the Baro River near Gambela in the western part of the country and the Omo River in the southern area near the border with Kenya (Breuil, 1995).

Hence, Ethiopia in general and Oromia region in particular has high production potential and composition of fish fauna, accurate fishery investigation has been carried out only in a few of the numerous freshwater bodies, and relatively a large number of small and medium water bodies have not been well studied and explored. This is the case with the fish fauna of Bishoftu crater Lakes in particular. Thus, the lakes are among such freshwaters that have received no adequate attention on the fishery in general. Therefore, the importance of fish exploration of Ethiopian freshwater systems on one hand and the absence of adequate study on fish composition, production potential on the other hand, justifies this study on Bishoftu Crater Lakes to contribute for food security of the country. Therefore, the objective of this paper was to assess the current physico-chemical characteristics, to assess the composition and production potential of the fishes and to assess the constraints that affects the production of the fishes in the lakes.

## Materials and Methods

### *Study areas*

Lake Hora-Kilole: is one of the Bishoftu crater lakes that a small and shallow water body (Fig.1). It was once grouped among the unique saline lakes of East Africa, along with lakes Abijata, Arenguade, Chitu and Shalla in Ethiopia (Talling *et al.*, 1973; Wood *et al.*, 1984; Wood and Talling, 1988; Talling and Lemoalle, 1998). The volume of the lake is not much affected by irrigation, as there was not much such activity. Most of the agricultural practices were focused on food crops that mainly depended on seasonal rains. However, this anthropogenic effect has turned L. Hora-Kilole from a highly saline-alkaline hypertrophic lake into a highly diluted typical oligotrophic tropical freshwater system. The phytoplankton community, which was almost exclusively dominated by *Arthrospira fusiformis*, the zooplankton community, which was dominated by *Paradiaptomus africanus* and the flamingoes that thrived on these species of plankton completely disappeared as the essential preconditions of saline-alkaline nature of the lake was altered by the inflow from River Modjo (Brook Lemma, 1994). The lake supports a commercial fishery, based largely on *Oreochromis niloticus* (Rediat Habteselase, 2012).

Lake Hora-Arsedi: It is one of the Bishoftu crater lakes (Fig.1), was believed to be created around 7000 years ago by volcanic collapse above zone of fractured rocks. The lake is a double crater with a maximum depth of 38 m (North crater) and 31m (South crater) and a mean depth of 17.5 m, located 47 km away from Addis Ababa in the south eastern direction at 8°50' and 39°E at an altitude of 1850 m (Prosser, *et al.*, 1968; Wood, *et al.*, 1984).The vertical distance from the crater rim to the lake surface is about 80 m. Lake Hora-Arsedi receives 43% of its total inflow from groundwater, but almost all water lose (97%) is by evaporation. The lake has a surface area of about 1.03 km<sup>2</sup> (Table 1). The region around the lake characterized by moderate rainfall, varying around about 850 mm per annum (Rippey and Wood, 1985), high incident solar radiation and low relative humidity. The region has two rainy periods, the minor one extending roughly from February to April and the major one beginning in June and ending in September. The temperature of its surface water was frequently found to be about 22°C with a maximum of 24.5°C and minimum of 19.2°C, while the bottom temperature was almost constant (19.2°C-19.4°C) (Wood *et al.*, 1976). Its seasonal cycle of stratification and mixing is probably similar to that of the nearby Lake Babogaya, which mostly resembles hydrochemically (Lamb, 2001).

Lake Koriftu: Lake Kuriftu is another lake found in the town of Bishoftu (Fig.1). It is found at an altitude of 1860 m, some 47 Km southeast of Addis Ababa. The lake is located at 8° 47' N and 39° 00'E. It is a



shallow around 6 m maximum depth (Brook Lemma *et al.*, 2001). The region around the lake is characterized by moderate rainfall, varying around about 850 mm per annum (Rippey and Wood, 1985), high incident solar radiation and low relative humidity. The region has two rainy periods, the minor one extending roughly from February to April and the major one beginning in June and ending in September (Rippey and Wood, 1985.).

With the establishment of Kale Hiwot Children's and Integrated Development Center in the proximity of the lake, plantation of trees, construction of utilities and establishment of livestock and agricultural farms were made around the southern shore of the lake. The trees found around the lake include *Accacia abyssinica*, *Jacaranda mimosifolia* and species of Eucalyptus and Juniperus. Macrophytes including Passifloraceae is observed. The zooplankton of Lake Kuriftu is composed primarily of rotifers, some cladocerans and a few copepods (Brook Lemma *et al.*, 2001; Girum Tamire, 2006). The piscifauna of the lake constituted by *Oreochromis niloticus* and *Cyprinus carpio*. Some birds (pelicans and ducks) are often seen on the lake (personal observation).

Table 1. Some physical characteristics of the sampling sites

Lake	Location	Altitude (masl)	Surface area (Km <sup>2</sup> )	Maximum depth (m)
Hora-Kilole	8 <sup>0</sup> 48' N, 39 <sup>0</sup> 5'E	1920	1.18	7.8
Lake Hora-Arsedi	8 <sup>0</sup> 50'N and 39 <sup>0</sup> E	1850	1.03	38
Lake Koriftu	8 <sup>0</sup> 47' N and 39 <sup>0</sup> E	1860	0.4	6
Lake Babogaya	8 <sup>0</sup> 51N and 39 <sup>0</sup> E	1870	0.58	71

Lake Babogaya: Lake Babogaya is one of the volcanic crater lakes found in Bishoftu town at about 45 Km East of Addis Ababa (Fig.1). The lake is small, roughly circular and deep, and is found at an altitude of 1870 m and at about 9<sup>0</sup>N latitude and 39<sup>0</sup>E longitude (Prosser *et al.*, 1968; Wood, *et al.*, 1984). Like the other volcanic crater lakes of the area, it is a closed system surrounded by very steep and rocky hills. The vertical distance from the lake's surface to the crater rim is 20 m, and this affords moderate protection from wind (Baxter, 2002). The lake fed primarily by precipitation falling directly on its surface and run-off from its small catchment area (Prosser *et al.*, 1968), which was formed from volcanic rocks of basalt, rhyolite and tuff (Mohr, 1961). The phytoplankton community is dominated by blue-green algae, particularly *Microcystis aeruginosa* (Wood and Talling, 1988), while the zooplankton is composed of copepods (*Afrocyclus gibsoni*, *Lovenula africana*), rotifers (*Asplancha sieboldi*, *Brachionus calyciflorus* and *Hexarthra jenkinsae*) and cladocera (Yeshimebet Major, 2006). The fish community found in Lake Babogaya is composed African catfish, *Clarias gariepinus*, and two tilapiine species (*Oreochromis niloticus* and *Tilapia zilli*), which were introduced in to the Lake by MOA (Ministry of Agriculture) with the aim of fishery development.

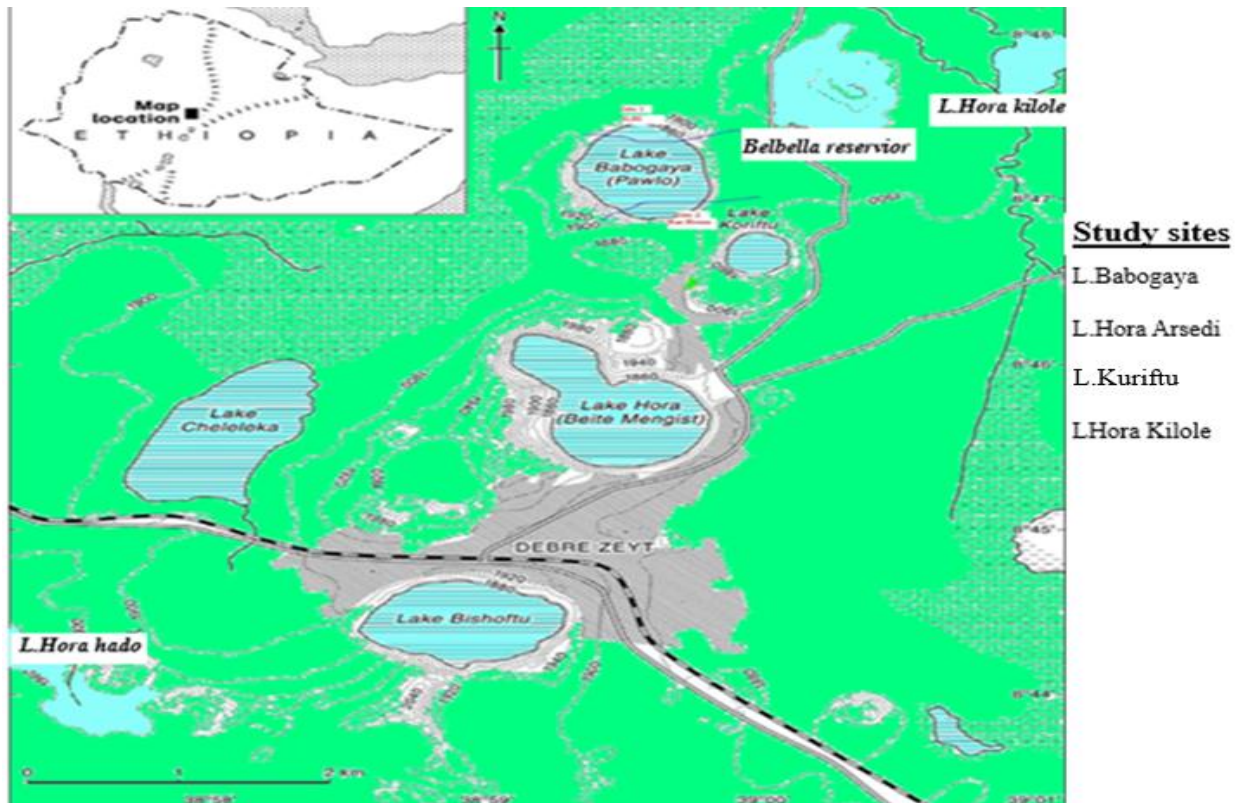


Figure 1. Location of Bishoftu Crater Lakes

### ***Site selection and sampling procedure***

Physico-chemical parameters: To assess the physico-chemical factors of the lakes, six sampling sites were selected based on geographical proximity and/or habitat similarity, their distance from anthropogenic effects. Samples were collected monthly with a Van Dorn bottle sampler from the pre-selected sampling station during the period spanning from July 2018 - June 2021. Samples were collected from selected depths distributed within the euphotic zone and mixed in equal proportions to produce composite samples. The composite samples were used for the analysis of inorganic nutrients, identification of phytoplankton taxa, and estimation of phytoplankton biomass as Chlorophyll a (Chl-*a*) production. For the identification and enumeration of zooplankton, separate samples were collected by towing upward using a tow net (55  $\mu$ m).

Fish parameters: Parallel to the physico-chemical sampling, every sampling periods the fishes were collected at all sites using variety of fishing gears. The gears include gill nets of various mesh sizes (6, 8, 10 and 12 cm stretched mesh size), monofilament nets with various stretched mesh sizes (5 mm to 55 mm stretched mesh size) and multiple long-lines with hooks of different size (9, 10, 11 and 12). The gears were set in the afternoon (4:00 pm) and were collected in the following day (7:00 am). Immediately after capture, some morphometric measurements were measured and the fish samples were put in plastic jars containing 10% formalin and labeled with all necessary information. Then the preserved specimens were soaked in tap water for many days to wash the formalin away. Then, the samples were transferred to 75% ethanol before species identification was conducted. The specimens were identified to species level using taxonomic keys of Shiberu Tedla, 1973; Golubtsov *et al.*, 1995; Witte and Wim, 1995; Stiassny and

Abebe Getahun, 2007; Rediet Habteselassie, 2012 and figures from Fish base. Catch and effort: Estimation of catch composition of fishes in the lakes were made by taking the contribution in number and weight of each species in the total catch in each sampling effort. Yield of the fish were analyzed for better management of the fish in the lakes. In addition, secondary data were collected from published and unpublished sources to identify the major constraints that affects the production of the fishes in the lakes

### *Data Analysis*

Fish species composition were presented as a numerical contribution by each species. This was determined by calculating the percentage of each species represented in the total catch for each station. Descriptive statistics were also used to analyze the remaining collected data using SPSS software (SPSS V.19.0).

### **Results and discussions**

#### *Physico-chemical factors that influences the fish community structure of the lakes*

The nutrients displayed relatively more variation among the lakes than did the physical parameters except secchi depth (Table 2). The lower level in Secchi depth reading in Lake Koriftu shows that turbidity in the lake is increasing, which can be attributed to catchment degradation, siltation and perhaps introduced bottom-stirring fish such as carp becoming more and more abundant in the lake (Fig.2).

Table 2. Mean and standard error values of physico-chemical variables of the lakes

Parameters	Lakes			
	Hora-Kilole	Hora-Arsedi	Koriftu	Babogaya
Temperature ( $^{\circ}$ C)	20.06 $\pm$ 0.3	21.3 $\pm$ 1.5	21.15 $\pm$ 0.2	21.01 $\pm$ 0.6
Secchi depth (cm)	38.58 $\pm$ 0.4	110.54 $\pm$ 0.1	20.2 $\pm$ 0.4	98.7.0 $\pm$ 0.5
Nitrate ( $\mu$ g/L)	39 $\pm$ 9.3	41.11 $\pm$ 0.5	44.05 $\pm$ 0.6	31.2 $\pm$ 5.4
SRP ( $\mu$ g/L)	39.36 $\pm$ 0.3	49.14 $\pm$ 0.9	51.13 $\pm$ 5.2	44.15 $\pm$ 4.2
Dissolved Oxygen(mg/l)	7.9 $\pm$ 0.9	7.1 $\pm$ 0.2	6.4.31 $\pm$ 0.8	7.8 $\pm$ 1.7
Chl- a ( $\mu$ g/L)	35 $\pm$ 0.9	37.2 $\pm$ 6.1	47 $\pm$ 0.6	32.2 $\pm$ 1.3
pH	9.1 $\pm$ 0.1	9 $\pm$ 0.3	8.4 $\pm$ 0.8	8.5 $\pm$ 0.6
Conductivity ( $\mu$ S/cm)	510.1 $\pm$ 2.3	350 $\pm$ 1.2	345.1 $\pm$ 2.4	967 $\pm$ 36

Temperature ranged from 20.06  $\pm$  0.3  $^{\circ}$  C (L. Hora-Kilole) to 21.3  $\pm$  1.5  $^{\circ}$  C (L. Hora-Arsedi) whereas pH ranged from 8.4  $\pm$  0.8 (L. Koriftu) to 9.1  $\pm$  0.1(Hora-Kilole). Hence, the pH values of the lake water in all lake is suitable for normal biological activity set as 6.5 - 8.5 by the European Economic Community (EEC, 1980). Dissolved oxygen varied from 6.4.31  $\pm$  0.8 to 7.9  $\pm$  0.9 mg/l. Low dissolved oxygen was measured at Lake Koriftu categorized as heavily impacted sites.

Conductivity varied from 345.1 $\pm$ 2.4 in L.Koriftu to 967 $\pm$ 36  $\mu$ S/cm (L.Babogaya). Soluble Reactive Phosphate varied from 39.36 $\pm$ 0.3 (H.kilole) to 51.13 $\pm$ 5.2  $\mu$ g/l (Koriftu) and nitrite ranged from 31.2 $\pm$ 5.4 (Babogaya) to 44.05 $\pm$ 0.6  $\mu$ g/l (Koriftu). Relatively an extremely high value of nitrate was recorded in

Lake Koriftu. Water quality testing is an important part of environmental monitoring. When water quality is poor, it affects not only aquatic life but also the surrounding ecosystem.

***Composition of the fishes in the lakes***

A total of five species of fishes in the Families Cyprinidae, Clariidae and Cichilidae were identified from the lakes (Table 3). The species were *Cyprinus carpio*, *Labeobarbus intermedius* from the Family Cyprinidae, *Clarias gariepinus* from the Family Clariidae and *Oreochromis niloticus* and *Tilapia zillii* from the Family Cichilidae. The status (presence/absence) of the species from the sampling sites provided in Table 3.

Table 3. Fish species identified from the Lakes (Present (+), Absent (-))

Family	Fish species	Lakes			
		Hora-Kilole	Hora-Arsedi	Koriftu	Babogaya
Cyprinidae	<i>Cyprinus carpio</i>	+	-	+	-
	<i>Labeobarbus intermedius</i>	+	-	+	-
Clariidae	<i>Clarias gariepinus</i>	+	+	+	+
Cichilidae	<i>Oreochromis niloticus</i>	+	+	+	+
	<i>Tilapia zillii</i>	+	+	+	+

A total of 11,254 fish specimens were recorded from the three families during the study period and the composition of the fish described in Figure 2. *Oreochromis niloticus* and *T.zillii* were more abundant in all lakes as compare to other fish species (Fig.2). The most abundant fishes (*O. niloticus* and *T.zillii*) were also the most widely distributed in different water bodies, as they are highly tolerant to a wide range of environmental conditions (Greenwood, 1966; Demeke Admassu *et al.*, 2015). In most rift valley lakes, *O. niloticus* is one of the fish species that are abundantly found in the littoral zones during juvenile stage (Eyuaem Abebe, 1984; Elias Dadebo, 1988; Eyuaem Abebe and Getachew Tefera, 1992; Lemma Abera, 2012) and similar phenomenon has been observed in Lake Naivasha (Mavuti, 1983). Currently, the littoral zone is being affected due to different anthropogenic factors, and the composition of the fishes will be changed as a result, progressive increase in the proportion of other fish species like *C. carpio*. Currently, *Cyprinus carpio* was more capture in Lake H.kilole (10%) next to Koriftu (18%) (Fig2). *Labeobarbus intermedius* was found only in the two lakes with least percentage composition (Fig.2).

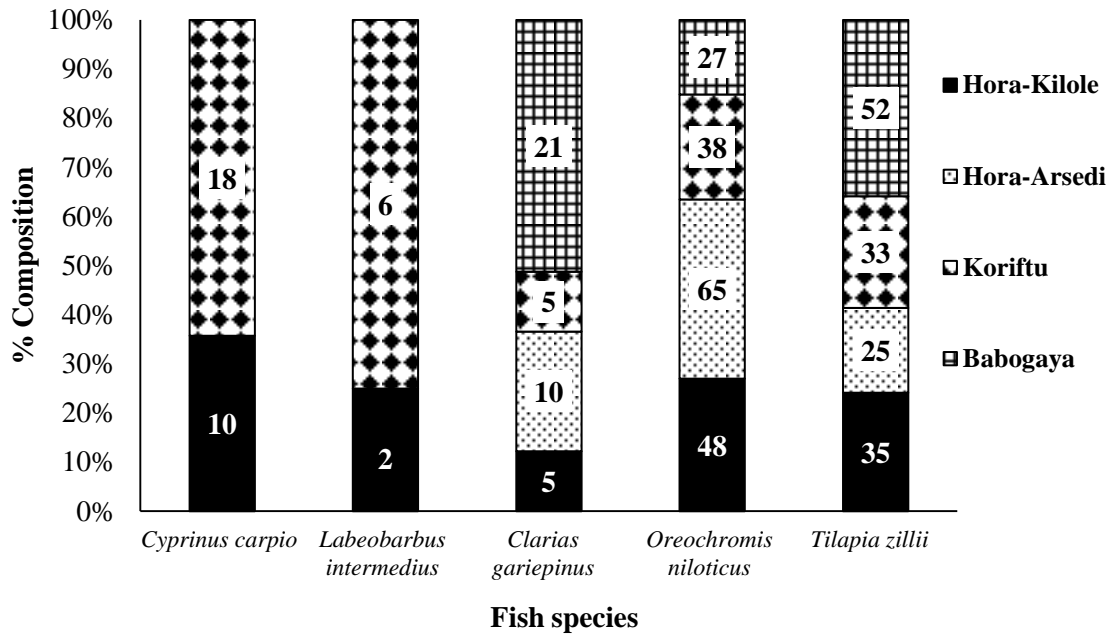


Figure 2. Fish species composition by number (%)

#### ***Fish catch of the lakes***

The estimated mean annual catch in tons per gear type were summarized in Tables 4 -7. The total CpUE were 2.3, 2.8, 1.1 and 1.6 for Lake H.kilole, H.Arsedi, Koriftu and Babogaya, respectively (Table 4 - 7). Gill net the most common gear that operated in all lakes as compare th the two gears (circular net and long line).

Table 4. Catch percentage and yield of fishes by species and gears for Lake Hora-kilole

Gear	Total catch in tons					Total
	<i>O. niloticus</i>	<i>C. gariepinus</i>	<i>C. carpio</i>	<i>L.intermedius</i>	<i>T.zillii</i>	
Gill net	1.02	0.115	0.23	0.046	0.655	2.066
Circular net	0.084	0	0	0	0.15	0.234
Long line	0	0	0	0	0	0
Total	1.104	0.115	0.23	0.046	0.805	2.3
Catch percentage	48	5	10	2	35	

Table 5. Catch percentage and yield of fishes by species and gears for Lake Hora-Arsedi

Gear	Total catch in tons					
	<i>O. niloticus</i>	<i>C. gariepinus</i>	<i>C. carpio</i>	<i>L.intermedius</i>	<i>T.zillii</i>	Total
Gill net	1.729	0.28	0	0	0.57	2.579
Circular net	0.091	0	0	0	0.13	0.221
Long line	0	0	0	0	0	0
Total	1.82	0.28	0		0.7	2.8
Catch percentage	65	10	0	0	25	

Table 6. Catch percentage and yield of fishes by species and gears for Lake Koriftu

Gear	Total catch in tons					
	<i>O. niloticus</i>	<i>C. gariepinus</i>	<i>C. carpio</i>	<i>L.intermedius</i>	<i>T.zillii</i>	Total
Gill net	0.418	0.055	0.198	0.066	0.363	1.1
Circular net	0	0	0	0	0	0
Long line	0	0	0	0	0	0
Total	0.418	0.055	0.198	0.066	0.363	1.1
Catch percentage	38	5	18	6	33	

Table 7. Catch percentage and yield of fishes by species and gears for Lake Babogaya

Gear	Total catch in tons					
	<i>O. niloticus</i>	<i>C. gariepinus</i>	<i>C. carpio</i>	<i>L.intermedius</i>	<i>T.zillii</i>	Total
Gill net	0.382	0.336	0	0	0.832	1.55
Circular net	0.05	0	0	0	0	0.05
Long line	0	0	0	0	0	0
Total	0.432	0.336	0	0	0.832	1.6
Catch percentage	27	21	0	0	52	

Total annual fish catch from the lakes during the study period was described in figure 3. Relatively, high production potential of fish in Lake Babogaya next to H.Arsedi (Fig. 3). The problem of Bishoftu Crater Lakes, fishery was that fishing is open to everyone who wishes to do so as source of income and food. Hence, the current annual fish production of the lakes declining due to overexploitation. There was a continuous decline in the fish species composition of some fish species, like *C.gariepinus* and *O.niloticus* from the lake Babogaya was the dominant in 2007, but in this study, there was an increase in *T.zillii* population which was also observed from the fishermen catches to some extent.

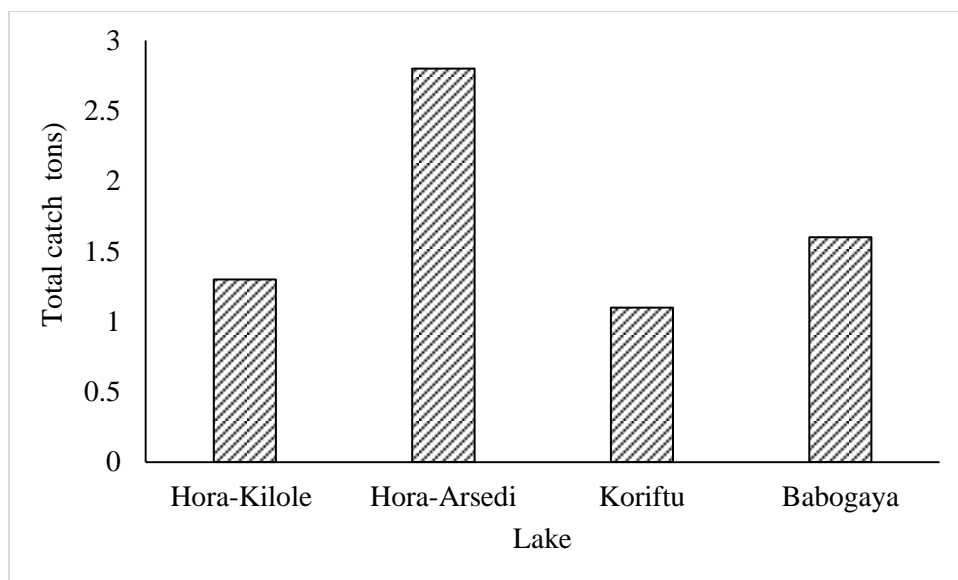


Figure 3. Total annual fish catch from the lakes during the study period

#### ***Major constraints that affects the production of the fishes in the lakes***

Fishing gears: Different fishing methods catch different types and sizes of fishes in the same area. Gill nets (monofilament), circular net and long-line were the most common fishing gears that were being used in the lakes (Table 4 - 7). The recommended minimum mesh size of the lake for *O. niloticus* was 10 cm (LFDP, 1993) and currently that not practiced in the study areas. The fishermen utilize wooden boats for casting gillnets and long-lines and.

Change of water qualities: The physico-chemical and biological features of the lake were documented by various papers (Table 8). Currently the mean value of nutrients in all of the water bodies were increased (Table 8). However; values of physical parameters were decreased. An increase in the conductivity in the present study in all water bodies may be explained by the concentration of ions accumulated from the surface runoff from the town and agricultural areas around the lake.

Table 8. Trends in some physico-chemical factors of the lakes

Lake	Parameters	Units	Previous study	Present study	Trends
Hora Kilole	Secchi depth	cm	180 (Brook Lemma,1994)	38	Decreasing
	pH	-	9.6 (Talling <i>et.al.</i> , 1973)	9.1	Decreasing
	D. Oxygen	mg/L	9.7 (Brook Lemma, 2006)	7.9	Decreasing
	Chl-a	µg/L	535 (Wood and Talling,1988)	35	Decreasing
	Nitrate	µg/L	39±9.3 (Lemma Abera, 2007)	42	Increasing
	SRP	µg/L	27 (Zinabu G/Mariam,,1994)	39	Increasing
	Conductivity	µS/cm	100 (Zinabu G/Mariam,,1994)	110	Increasing
Hora Arsedi	Secchi depth	cm	168 (Tigest <i>et al.</i> , 2008)	110.5	Decreasing
	pH	-	9.2 (Prosser, <i>et al.</i> ,1968)	9	Decreasing
	D. Oxygen	mg/L	9.7 (Tigest <i>et al.</i> , 2008)	7.1	Decreasing
	Chl-a	µg/L	106 (Tigest <i>et al.</i> , 2008)	37	Decreasing
	Nitrate	µg/L	37.92(Tigest <i>et al.</i> , 2008)	41	Increasing

	SRP	µg/L	58.84 (Tigest <i>et al.</i> , 2008)	60	Increasing
	Conductivity	µS/cm	319 (Zinabu G/Mariam <i>et al.</i> 2002)	350	Increasing
Koriftu	Secchi depth	cm	20 (Brook Lemma <i>et al.</i> , 2001)	18	Decreasing
	pH	-	8.4 (Brook Lemma <i>et al.</i> ,2001)	8.5	Decreasing
	D. Oxygen	mg/L	7.6 (Zelalem Dessalegn, 2007)	6.3	Decreasing
	Chl-a	µg/L	55.6 (Zelalem Dessalegn, 2007)	47	Decreasing
	Nitrate	µg/L	33.3(Zelalem Dessalegn, 2007)	38	Increasing
	SRP	µg/L	41.5 (Zelalem Dessalegn, 2007)	51	Increasing
	Conductivity	µS/cm	319 (Brook Lemma <i>et al.</i> ,2001)	345	Increasing
Babogaya	Secchi depth	cm	139 (Yeshemebet Major,2006)	98.7	Decreasing
	pH	-	9.5 (Zinabu G/Mariam.,1994)	8.5	Decreasing
	D. Oxygen	mg/L	8.3 (Lemma Abera,2007)	7.8	Decreasing
	Chl-a	µg/L	37 (Zinabu G/Mariam and Tavlör,1997)	32.2	Decreasing
	Nitrate	µg/L	20 (Prosser <i>et al.</i> ,1968)	31.2	Increasing
	SRP	µg/L	5 ( Prosser <i>et al.</i> ,1968)	42	Increasing
	Conductivity	µS/cm	900 ( Prosser <i>et al.</i> ,1968)	967	Increasing

As described in Table 8, some of the physico-chemical trends of the lakes water were changed. Some of them may affect the aquatic organisms, such as fish kills occurring at the shore of the lake (personal observation in Lake H.Arsedi and Babogaya). Also evidence from other lakes in Ethiopia show that the longer term impact of human induced changes like deforestation increases the risk of flooding such as Hawassa Lake and Lake Ziway (Lemma Abera, 2018) or even complete degradation of the lake like in the case of Haramaya (Brook Lemma, 2011).

According to Zinabu Gebremariam (1998), population pressures and urbanization significantly affect cities near lakes and the lakes themselves were among the greatest potential causes of change in water quality and quantity (e.g. L. Ziway). Therefore, the current population pressure around the lakes, cause pollution and water scarcity, and in turn impairing fish population and future development of the resources.

### Conclusions and Recommendations

The nutrients displayed relatively more variation among the lakes than did the physical parameters. The fish fauna of Bishoftu Crater Lakes dominated by *O. niloticus* and *T.zillii* in all lakes as compare to other fish species. The estimated mean annual catch in tons per gear type were 2.3, 2.8, 1.1 and 1.6 for Lake H.kilole, H.Arsedi, Koriftu and Babogaya, respectively. The problem of Bishoftu Crater Lakes, fishery was that fishing is open to everyone who wishes to do so as source of income and food. Hence, the current annual fish production of the lakes declining due to overexploitation. Hence, the current fisheries management system in Ethiopia mainly consists of a fishing licensing system aimed at regulating access to the fishery and some technical conservation measures including mesh-size limitations for gillnets. The waste-disposal mechanisms of the adjacent businesses and/or residential areas should be investigated to establish the exact source and nature of the pollutants. Those people responsible for the pollution should be encouraged to implement appropriate mitigating measures as soon as possible. The development of



aquaculture and other related alternative fisheries (cage culture) to reduce the pressure on the natural system of capture fishery should be encouraged to utilize the resources appropriately.

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# Limnological Assessment of Wodecha Reservoir and Harkiso Lake, Ethiopia

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## Abstract

The physico-chemical water quality and plankton composition of Wodecha Reservoir and Harkiso Lake were studied from July 2020 to June 2021. Both the reservoir and lake were characterized by low water transparencies, with secchi disc depths of 19 cm and 21.5 cm for Wodecha reservoir and Lake Harkiso respectively. The mean water temperature (22.51 °C) of L. Harkiso was greater than Wodecha reservoir (16 °C). Dissolved oxygen content has higher values in the reservoir, due high circulation of water in the river as compared to the lake. Conductivity was recorded 230.96  $\mu\text{S cm}^{-1}$  and pH was 8.23 for Harkiso and 393  $\mu\text{S cm}^{-1}$  and pH was 8.01 for Wodecha Reservoir. The mean SRP was 43.31 and 78.3  $\mu\text{g L}^{-1}$  for the lake and reservoir, respectively. Five phytoplankton groups were identified in both waterbodies, includes *Chlorophyceae*, *Bacillariophyceae*, *Cyanophyceae*, *Dinophyta* and *Euglenophyta*. Zooplankton species encountered include 11 taxa from Wodecha Reservoir and 16 taxa in Lake Harkiso. Hence, as a conclusion, based on the current limnological parameter results the two water bodies are suitable for the production of the fishes and need to be enhanced *Cyprinus carpio* in Wodecha Reservoir and *Oreochromis niloticus* and *Clarias gariepinus* for Lake Harkiso. Key words: Lake Harkiso, Physico-chemical parameters, Planktons, Wodecha Reservoir.

**Keywords:** Lake Harkiso, Physico-chemical parameters, Planktons, Wodecha Reservoir

## Introduction

Tropical lakes and reservoirs are often highly productive and like any other water body they provide water for power generation, irrigation, recreation, fishery and other uses. Although they are limited and sensitive resources, they are probably among the most abused natural resources (Zinabu Gebremariam, 2002). Human activity is contributing much for accelerating the eutrophication process in the lakes throughout the world (WHO, 2003).

Reservoir ecosystems are intermediate between riverine and lacustrine environment in relation to morphology, hydrology, nutrient loadings and cycling and sources of organic matter (Wetzel, 2001). They are essential component of most irrigation systems besides for power generation, flood control and surface water harvesting which retain a large volume of water worldwide. In addition to their primary role in providing water for agriculture, industrial and domestic purposes and their role in power generation, most of these reservoirs have the potential to play an important role in fish production contributing significantly to the livelihoods of riparian communities in Ethiopia.

In Ethiopia, there are many manmade reservoirs and lakes that suitable for fish production. Wodecha reservoir is one such reservoir built across the Wodecha River to provide for irrigation and Harkiso Lake also virgin water body till now not used for any fishery activities. Scientific information on the biotic and physical features of these water bodies are crucial to use for fish production along with other purposes. In Ethiopia, apparently, more information exists on the physico-chemical features and biotic composition of

natural water bodies, however, no information available on the limnology of Lake Harkiso and Wodecha reservoir for fisher production and this study proposed to assess the physico-chemical features and biotic composition (phytoplankton and zooplankton) of the two water bodies.

## **Materials and Methods**

### ***Description of the study site***

Wodecha Reservoir established in Gimbichu district in East Showa Zone of the Oromia region by damming Wodecha River. It is located 31 km east of Bishoftu town and about 74 km southeast of Addis Ababa along the road to Chafe Donsa town. The reservoir situated on a highland with an average elevation of 2,600 m.a.s.l. Lake Harkiso is found adjacent to Lake Langeno in Rift valley area with an average elevation of 1,585 m.a.s.l.

### ***Field sampling***

The study was made to assess physico-chemical features and phytoplankton as well as zooplankton composition during July 2020 to June 2021 in the water bodies. Conductivity, Dissolved oxygen (DO), pH, and water temperature were measured in-situ using a multi-probe (Model HQ40d, HACH Instruments) from a central sampling station, about 3 m deep for Wodecha reservoir and 2m deep for Lake Harkiso. Surface water samples were collected and transported to laboratory for nutrient analysis. Analysis were done by following the Standard methods (APHA, 1995). Water transparency was estimated using a standard Secchi disc of 20 cm diameter. Phytoplankton and zooplankton samples were collected with plankton net of 30 and 60  $\mu\text{m}$  mesh size from the open water respectively. Identification of phytoplankton was made using different identification keys of Talling and Lemoalle (1998) and Willen (1991), while zooplankton species was identified by following the descriptions of Fernando *et al.* (2002). Phytoplankton biomass was estimated as chlorophyll *a* (Chl-*a*) concentration (Wetzel & Likens, 2000).

## **Results and Discussions**

### ***Physico-chemical parameters in the study area***

Both the reservoir and the lake were characterized by low water transparencies, with secchi disc depths of 19 cm and 21.5 cm for Wodecha reservoir and Lake Harkiso respectively. The water mean temperatures was 22.51  $^{\circ}\text{C}$  and the mean dissolved oxygen concentration was 5.67  $\text{mg L}^{-1}$  for Harkiso and 16.43 $^{\circ}\text{C}$  and 6.81 $\text{mg L}^{-1}$  for the reservoir (Table 1). Dissolved oxygen content has higher values in the reservoir, due high circulation of water in the river as compared to the lake, the possible reason for such high oxygen values. Offem *et al.*, (2009) reported similar findings in, Nigeria and Lemma Abera (2018), in Lake Ziway around inlet of Katar River. Conductivity was recorded 230.96  $\mu\text{S cm}^{-1}$  and pH was 8.23 for Harkiso and 393  $\mu\text{S cm}^{-1}$  and pH was 8.01 for Wodecha Reservoir. The mean SRP was 43.31 and 78.3  $\mu\text{g L}^{-1}$  for the lake and reservoir, respectively and the results of nitrate, nitrate and Ammonium-quality parameters were presented in Table 1.

Table 1: Physico-chemical analysis of the two water bodies

Parameters	Values	
	Lake Harkiso	Wodecha reservoir
SRP ( $\mu\text{g L}^{-1}$ )	43.31 $\pm$ 2.5	78.3 $\pm$ 1.8
Secchi depth (cm)	21.5 $\pm$ 0.4	19 $\pm$ 0.9
Temperature ( $^{\circ}\text{C}$ )	22.51 $\pm$ 0.6	16.43 $\pm$ 1.5
Nitrite ( $\mu\text{g L}^{-1}$ )	30.81 $\pm$ 0.3	22.16 $\pm$ 0.7
Ammonium ( $\mu\text{g L}^{-1}$ )	57.13 $\pm$ 1.2	44.26 $\pm$ 4.6
Nitrate ( $\mu\text{g L}^{-1}$ )	35.26 $\pm$ 0.7	29.48 $\pm$ 1.9
Chl-a ( $\mu\text{g L}^{-1}$ )	54.16 $\pm$ 7.6	48.11 $\pm$ 0.8
pH	8.23 $\pm$ 0.4	8.01 $\pm$ 1.3
Dissolved oxygen ( $\text{mg L}^{-1}$ )	5.67 $\pm$ 0.5	6.81 $\pm$ 0.4
Conductivity ( $\mu\text{S/cm}^{-1}$ )	230.96 $\pm$ 5.2	393 $\pm$ 4.9

### ***Biotic communities***

Five phytoplankton groups were identified in both waterbodies, includes *Chlorophyceae*, *Bacillariophyceae*, *Cyanophyceae*, *Dinophyta* and *Euglenophyta* (Table 2). The highest taxa number was observed in the *Chlorophyceae* followed by *Bacillariophyta* and *Cyanophyceae* while *Dinophyta* and *Euglenophyta* were only represented by two and one species in the reservoir. In case of Lake Harkiso the predominant phytoplankton algae were *Chlorophyceae* and *Bacillariophyceae* next to *Cyanophyceae* with five species (Fig.1).

Table 2. List of phytoplankton identified from Wodecha Reservoir and Lake Harkiso

Phytoplankton group	Species	
	Wodecha Reservoir	Lake Harkiso
<i>Chlorophyceae (Green algae)</i>	<i>Staurastrum sp.</i> <i>Chlamydomonas sp.</i> <i>Chlorella sp.</i> <i>Scenedesmus sp.</i> <i>Volvox sp.</i> <i>Pediastrum sp</i> <i>Melosira spirogyra</i>	<i>Pediastrum sp.</i> <i>Scenedesmus sp</i> <i>Chlamydomonas sp.</i> <i>Phacotus sp.</i>
<i>Bacillariophyceae (Diatoms)</i>	<i>Cyclotella sp.</i> <i>Navicula sp.</i> <i>Nitzschia sp.</i> <i>Synedra sp.</i>	<i>Rhopahodia sp.</i> <i>Thalassiosira sp.</i> <i>Navicula sp.</i> <i>Nitzschia sp.</i>
<i>Cyanophyceae (Blue-green algae)</i>	<i>Anabaena sp</i> <i>Anabaenopsis sp</i> <i>Microcystis sp.</i>	<i>Raphidiopsis sp.</i> <i>Anabaena sp.</i> <i>Microcystis sp.</i> <i>Planktolyngbya sp.</i> <i>Cylindrospermopsis sp.</i>
<i>Dinophyta (Dinoflagellates)</i>	<i>Ceratium sp.</i> <i>Peridinium sp.</i>	<i>Peridinium sp</i>
<i>Euglenoids (Euglenophyceae)</i>	<i>Euglena sp</i>	<i>Lepocincilis sp.</i> <i>Euglena sp</i>

The dominance of *Chlorophyceae* and *Cyanophyceae* in the two water bodies were favored by several environmental factors such as low light intensity and high pH. Some members of this group have the ability to withstand grazing (Hargreaves & Heusel, 2000) and some are buoyant, which is especially important in turbid waters as of these water bodies. The result of this study also in agreement with Mwaura *et al.* (2002), who documented the dominance of these algae in eight Kenyan highland reservoirs due to high nutrient loading after long rain periods, to high temperature, to their ability to fix nitrogen during dry periods and due to their ability to regulate their vertical position in the water column.

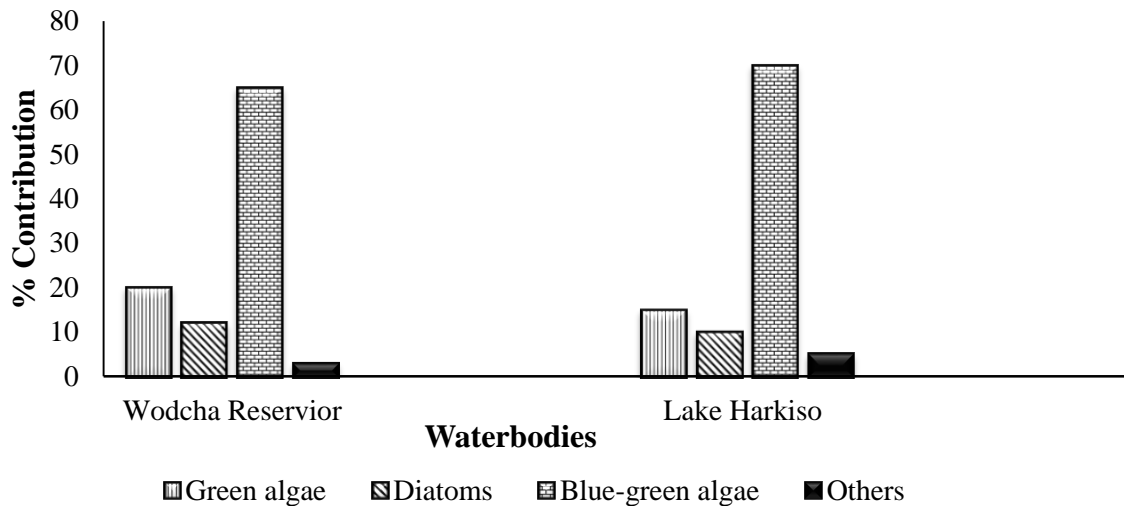


Fig.1. Percent composition of different taxonomic groups to the abundance of total phytoplankton in Wodecha reservoir and Lake Harkiso

Zooplankton species encountered include a total of 11 taxa from Wodecha Reservoir and 16 taxa in Lake Harkiso (Table 3). Rotifers were the richest group with eight and six genera in the reservoir and, lake respectively. Cladocerans and copepods were each represented by four genera in the lake.

Table 3. List of Zooplankton identified from Wodecha Reservoir and Lake Harkiso during the study period

Zooplankton group	Genus	
	Wodecha Reservoir	Lake Harkiso
Cladocera	<i>Diapohanosma</i> <i>Daphnia barbata</i> <i>Moina</i>	<i>Ceriodaphnia sp</i> <i>Daphnia pulex</i> <i>Daphnia cuculata</i> <i>Bosmina sp</i>
Copepoda	<i>Eucyclops</i> <i>Thermocyclops</i>	<i>Callanoids</i> <i>Copepoda sp</i> <i>Mesocyclops</i> <i>Thermocyclop</i>

Rotifera	<i>Trichocerca</i> <i>Barchionus</i> <i>Filinia</i> <i>Asplanchna</i> <i>Hexarthra</i> <i>Polyarthra</i>	<i>Keratella</i> <i>Brachionus</i> <i>Asplanchna</i> <i>Keratella</i> <i>Filinia</i> <i>Polyarthra</i> <i>Trichocerca</i> <i>Hexarthra</i>
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The percent composition of different taxonomic groups were described in figure 2. Numerically, rotifer taxa formed the major component of the zooplankton in both water bodies, accounts 54% and 46 % in the reservoir and lake, respectively (Fig.2). Rotifers were known to be richer than cladocerans and copepods in tropical water bodies (Rocha *et al.*, 1995). The dominance of rotifers in terms of species richness and abundance, like in this study, seems to be a common pattern in tropical water bodies. Rotifer dominance in reservoirs and shallow water bodies, can also be related to their opportunist organic matters and thus a large proportion of SRP is assimilated and converted to algal biomass; a common phenomenon in rift valley lakes.

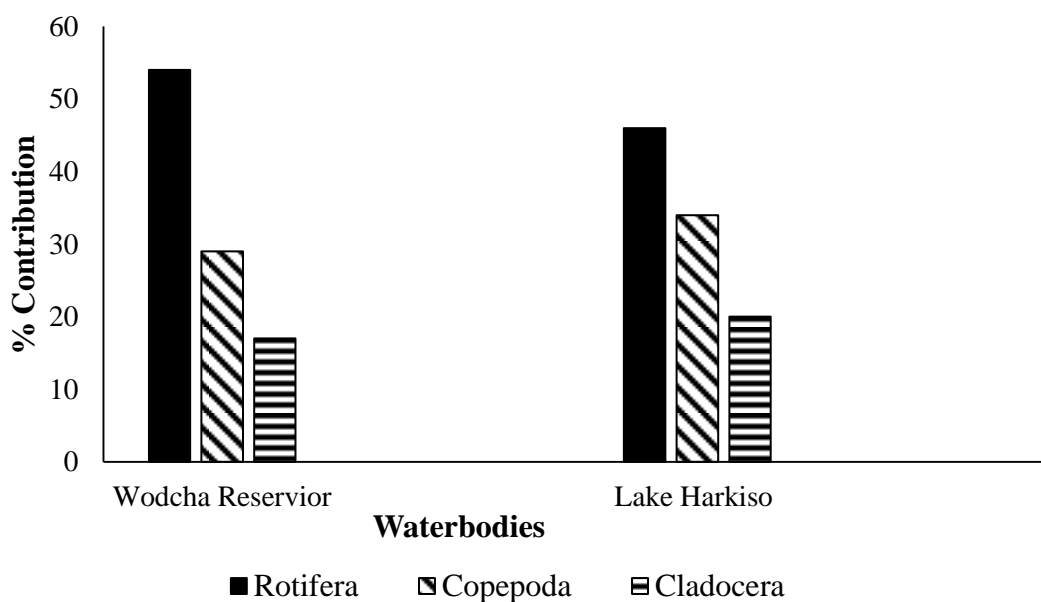


Fig.2. Percent composition of different taxonomic groups to the abundance of total Zooplankton in Wodecha reservoir and Lake Harkiso

The highest density of zooplankton was associated with an increase in nutrient concentration in the reservoir and Lake Harkiso, which probably stimulated algal growth. Cladoceran and copepod density were the same trend in both waterbodies. Arcifa *et al.* (1992) presented an evidence that predation by invertebrates also be an important factor influencing the structure of the whole zooplankton community.

## Conclusions and Recommendations

Physico-chemical and plankton conditions of Lake Harkiso and Wodecha Reservoir varied due to the nature of the water bodies (Lake and Reservoir). A large amount of abiogenic material was washed in the reservoir as compare to the lake due to the River Wodecha. This, together with the predominantly low concentrations of ammonia and soluble reactive phosphorus, has most probably been limiting the growth of phytoplankton. These changes water transparency and nutrient concentration that negatively affect the water quality in the future. Hence, as a conclusion, the current limnological parameter of the two water bodies indicated that both are suitable for the production of the fishes (Wodecha Reservoir for *Cyprinus carpio* and Harkiso for *Oreochromis niloticus* and *Clarias gariepinus*). As a recommendation before undesirable phenomena similar to that of Koka reservoir take place wetland management should be carried out. As far as the reservoir used, as the primary source of drinking water by local inhabitants and their cattle, continuous monitoring of the reservoir water quality standards by the concerned authorities is recommended.

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# Assessment of Limnological Parameters, Fish Species Composition and Gear selectivity in Belbella Reservoir

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## Abstract

*Limnological parameters, fish species composition and some biology of Oreochromis niloticus in Belbella Reservoir was studied during the period between July 2018 to June 2021 using different limnological kits and fishing gears. The result obtained for conductivity and pH as the mean  $\pm$  SE values were  $377 \pm 5.7 \mu S cm^{-1}$  and  $8.4 \pm 0.7$ , respectively. Among the phytoplankton taxa, Blue green algae with 11 genus, followed by green algae and diatoms with six and four genus respectively. Rotifers were the most species-rich zooplankton group with seven genus. The second most important zooplankton group was the cladocerans. The copepods were, poorly represented, with only two genus, in the zooplankton community of Belbella reservoir. Oreochromis niloticus was the dominant fish species in the reservoir and Length-weight relationship of the fish was curvilinear, show isometric growth and statistically highly significant ( $P < 0.05$ ). Sex ratio was not significantly different from 1:1 for all sampling months. The 50% maturity length ( $L_{50}$ ) of *O. niloticus* was estimated to be 18 cm total length for females and 17.5 cm total length for males. The current gill net mesh size (10 cm) used in the reservoir was captured the fish with the range of length class between 16-20 cm that coincide with at their  $L_{50}$  of both sexes of the fish. Hence, care should be taken based on the length at first maturity that immature fish will not be affected by in the water body.*

**Key words:** Belbella reservoir, fishes, fishing gear, physico-chemical parameters, planktons.

## Introduction

Ethiopia has a number of lakes and rivers with substantial quantity of fish stocks. Many artificial water bodies including reservoir have also been stocked with fish for fishery. Reservoirs are water bodies that are formed by humans for purposes of drinking and municipal water supply, industrial cooling water supply, power generation, irrigation, river regulation, flood control, fisheries, recreational uses and waste disposal (Chapman, 1996). Reservoirs show many of the same basic hydrodynamic, chemical and biological characteristics as the natural lakes. Like natural lakes, reservoirs harbor phytoplankton and zooplankton, which form integral parts of the aquatic food web and influence other aspects of the lake including color and clarity of the water and fish production.

Belbella reservoir is one of the dam that constructed in 1980 by a Cuban Civil Mission in collaboration with Ethiopian Water Resources Authority. The protection works, canals, and on-farm structures for the dam were later constructed by the Ethiopian Water Works Construction Authority, with an objective of irrigating land area to be used by State Farms for fruit production. The other storage dam (Wadecha dam) through hydrological catchments transfers recharges the reservoir. The reservoir also serves as the primary source of drinking water for both local inhabitants and livestock.

Although some studies were conducted on the reservoir, the changes on the liminological parameters of the water body is fast and hence the information on the reservoir gets outdated quickly. Therefore, regular updating, monitoring and control are essential and this study becomes important and relevant for better management and sustainable use of the resources for fishery development. Hence, this paper attempts to assess some physico-chemical factors of the reservoir; to assess fish species composition and the biology of the dominant fish; and finally to identify appropriate fishing gear to enhance the fish production of the water body.

## Materials and Methods

### *Description of the study area*

Belbella Reservoir (Fig. 1), established in Ada'a district in East Showa zone of the Oromia region by damming Wodecha River. It is located 17 km east of Bishoftu town and about 64 km southeast of Addis Ababa along the road to Chafe Donsa town. It found between  $38^{\circ} 01'$  -  $40^{\circ} 04'$  E longitude and  $08^{\circ} 47'$  -  $09^{\circ} 00'$  N latitude. The reservoir situated on a highland with an average elevation of 2,300 m.a.s.l.

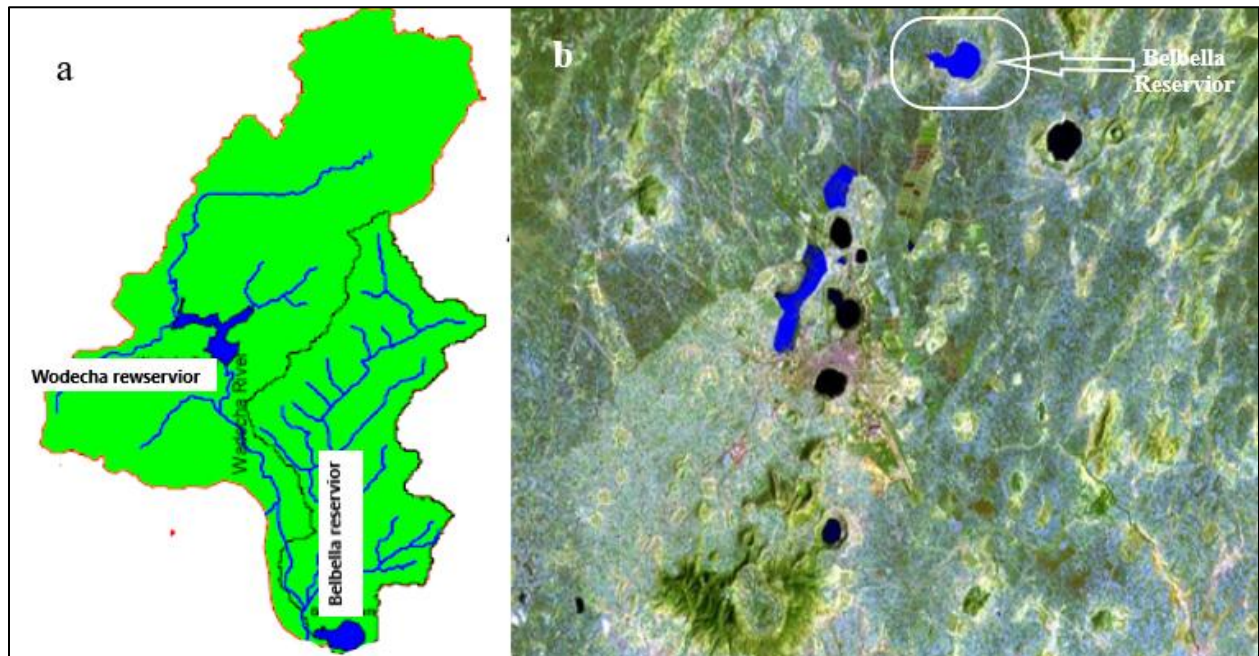


Fig 1. Map showing the drainage system (a) and google map of Belbella Reservoir (b)

Table 1. Physical characteristics of Belbella Reservoir (Wakena Totoba, 2006).

Description	values
Catchment area	85 Km <sup>2</sup>
Outlet-Pipe Diameter	0.76m
Maximum discharge	3.87m <sup>3</sup> /sec
Mean discharge	2.4m <sup>3</sup> /sec
Maximum depth	6m

Some of the physical characteristics described in table 1 and, agriculture is the major source of employment, revenue, export earnings and a means for ensuring food security in the reservoir. As a result, there has been an ever-increasing expansion of irrigation-agricultural development practices in the area although workable management and exploitation strategies are not yet in place.

The climatic condition of the reservoir area is wet to sub-humid. The wet season (March-September) with mean monthly rainfall varying from 65 to 239 mm. The rainfall of the study region is unimodal with the highest amount of rainfall occurring between June and August and accounting for about 76% of the mean annual precipitation in the catchment areas of the reservoir.

#### *Site selection and sampling procedure*

Physico-chemical parameters: To assess the physico-chemical factors that influences the fish community structure of the reservoir, four sampling sites were selected based on geographical proximity and/or habitat similarity, their distance from anthropogenic effects.

Samples were collected monthly with a Van Dorn bottle sampler from the pre-selected sampling station during the period spanning from July 2018 - June 2021. Samples were collected from selected depths distributed within the euphotic zone and mixed in equal proportions to produce composite samples. The composite samples were used for the analysis of inorganic nutrients, identification of phytoplankton taxa, and estimation of phytoplankton biomass as Chlorophyll a (Chl-*a*) production. For the identification and enumeration of zooplankton, separate samples were collected by towing upward using a tow net of zooplankton net.

Fish parameters: Parallel to the physico-chemical sampling, every month the fishes were collected at all sites using variety of fishing gears, which included gill nets of various mesh sizes (6, 8, 10 and 12 cm stretched mesh size), monofilament nets with various stretched mesh sizes (5 mm to 55 mm stretched mesh size) and multiple long-lines with hooks of different size (9, 10, 11 and 12). The gears were set in the afternoon (4:00 pm) and were collected in the following day (7:00 am). Immediately after capture, some morphometric measurements were measured and the fish samples were put in plastic jars containing 10% formalin and labeled with all necessary information. Then the preserved specimens were soaked in tap water for many days to wash the formalin away. Then, the samples were transferred to 75% ethanol before species identification was conducted. The specimens were identified to species level using taxonomic keys of Golubtsov *et al.* (1995); Witte and Wim (1995); Stiassny and Abebe Getahun (2007); Redeit Habteselassie (2012) and figures from Fish base.

#### *Data Analysis*

Plankton and fish species composition were presented as a numerical contribution by each species. This was determined by calculating the percentage of each species represented in the total catch for each station. Descriptive statistics were also used to analyze the remaining collected data using SPSS software (SPSS V.19.0).

## Results and Discussions

### *Physico- chemical parameters of the reservoir*

Some of the parameters that affect the quality of water in the reservoir, which indicated in Table 2. The result obtained for conductivity and pH as the mean  $\pm$  SE values were  $377\pm 5.7 \mu\text{S cm}^{-1}$  and  $8.4\pm 0.7$ , respectively, were comparable with the values ( $410 \mu\text{S/cm}$  and  $8.5$ , respectively) reported by Elizabeth Kebede *et al.* (1994) and very recently by Girum Tamire and Seyoum Mengistou (2012) with similar water system. Hence, the pH values of the lake water, it is suitable for normal biological activity set as  $6.5 - 8.5$  by the European Economic Community (EEC, 1980).

The mean nitrate value ( $148 \pm 1.5 \mu\text{g}^{-1}$ ) was higher than previously reported values for the same lake (Feyisa Girma, 2011), indicating an eutrophication trend with time. However, nitrate varied more than soluble reactive phosphate suggesting that it may be a limiting factor for phytoplankton growth in addition to siltation in the reservoir. Talling and Talling (1965) highlighted that nitrogen was the limiting nutrient for phytoplankton growth in tropical African lakes. The high concentration of dissolved oxygen was satisfactory for fish production (EEC, 1980). Dissolved oxygen content has higher values in the area and suitable for the production of fish. High circulation of water in the reservoir is the possible reason for such high oxygen values. Offem *et al.* (2009) reported similar findings in reservoir, Nigeria.

Table 2. Pooled mean and standard error values of physico-chemical variables of the reservoir (From July 2018 - June 2021).

Parameters	Values
Secchi depth (m)	$0.23\pm 0.9$
Temperature ( $^{\circ}\text{C}$ )	$21\pm 0.5$
pH	$8.4\pm 0.7$
Dissolved oxygen ( $\text{mg L}^{-1}$ )	$5.4\pm 0.1$
Conductivity ( $\mu\text{S cm}^{-1}$ )	$277\pm 5.7$
Nitrate ( $\mu\text{g L}^{-1}$ )	$148\pm 1.5$
Ammonium ( $\mu\text{g L}^{-1}$ )	$96\pm 9.4$
SRP ( $\mu\text{g L}^{-1}$ )	$82\pm 0.5$
Chl-a ( $\mu\text{g L}^{-1}$ )	$57.8 \pm 0.4$

Although the nitrate concentrations in the reservoir are comparable to those reported for an offshore station in Koka reservoir (Hadgembes Tesfay, 2007), they are much lower than the values recorded for Geffersa ( $10$  to  $300 \mu\text{g}^{-1}$ , Nigatu Ebisa, 2010) and Legedadi ( $240$  to  $1850 \mu\text{g}^{-1}$ , Adane Sirage, 2006). This was probably reflecting the differences among the reservoirs in the extent of application of fertilizers and external loading of the nutrient through runoff.

The concentrations of soluble reactive phosphate (SRP) and Chl *a* were  $82\pm 0.5 \mu\text{g L}^{-1}$  and  $57.8 \pm 0.4 \mu\text{g L}^{-1}$  respectively. Despite the fact that phosphorous is regarded as an extremely important element in controlling the trophic status of some tropical lakes (Kalff, 1983) and chlorophyll *a* was found to be strongly coupled to measured concentrations of phosphorus (Schindler, 1977; Praire *et al.*, 1989), the correlation between SRP and Chl *a* biomass of phytoplankton was poor in Belbella reservoir.

### *Plankton species composition*

The phytoplankton community was constituted by 25 genus (Table 2) belonging to six algal families. These were Cyanophyceae (Blue-green algae), Chlorophyceae (Green algae), Bacillariophyceae (Diatoms), Euglenophyceae (Euglenoids), Cryptophyceae (Cryptomonads) and Dinophyceae (Dinoflagelates) (Table 2).

Table 2. List of phytoplankton taxa identified in Belbella reservoir

<b>Family</b>	<b>Genus</b>
Chlorophyceae (Green algae)	<i>Actinastrum spp.</i>
	<i>Elakatothrix spp.</i>
	<i>Closteridium spp.</i>
	<i>Pediastrum spp.</i>
	<i>Scendesmus spp.</i>
	<i>Tetrastrum spp.</i>
Cyanophyceae (Blue-green algae)	<i>Anabena spp.</i>
	<i>Aphanocapsa spp.</i>
	<i>Aphanothece spp.</i>
	<i>Arthrospira spp.</i>
	<i>Chroococcus spp.</i>
	<i>Coelosphaerium spp.</i>
	<i>Cylindrospermopsis spp.</i>
	<i>Dactylococcopsis spp.</i>
	<i>Microcystis spp.</i>
	<i>Planktothrix spp.</i>
	<i>Pseudoanabaena spp.</i>
Bacillariophyceae (Diatoms)	<i>Nitzschia spp.</i>
	<i>Opephora spp.</i>
	<i>Pleurosigma spp.</i>
	<i>Synedra spp.</i>
Dinophyceae	<i>Peridinium spp.</i>
Euglenophyceae	<i>Euglena spp.</i>
	<i>Trachelomonas spp.</i>
Cryptophyceae	<i>Cryptomonas spp.</i>

Species composition of phytoplankton communities were observed in the reservoir (Fig. 2). Among the phytoplankton taxa, Blue green algae with 11 genus, followed by green algae and diatoms with six and four genus respectively. Other taxonomic groups of phytoplankton were relatively poorly represented in the phytoplankton community of the study reservoir (Fig.2).

The overwhelming dominance of blue-greens algae constituted primarily by *Cylindrospermopsis*. The dominance of algal groups enhanced by one or several environmental conditions such as light, temperature, water column mixing and availability of nutrients in the reservoir depending on the particular algal groups (Reynolds, 2006). High turbidity (Smith, 1986) and temperature (Shapiro, 1990)

and high total phosphate (Watson *et al.*, 1997) were among the physico-chemical factors known to initiate and enhance cyanobacterial dominance.

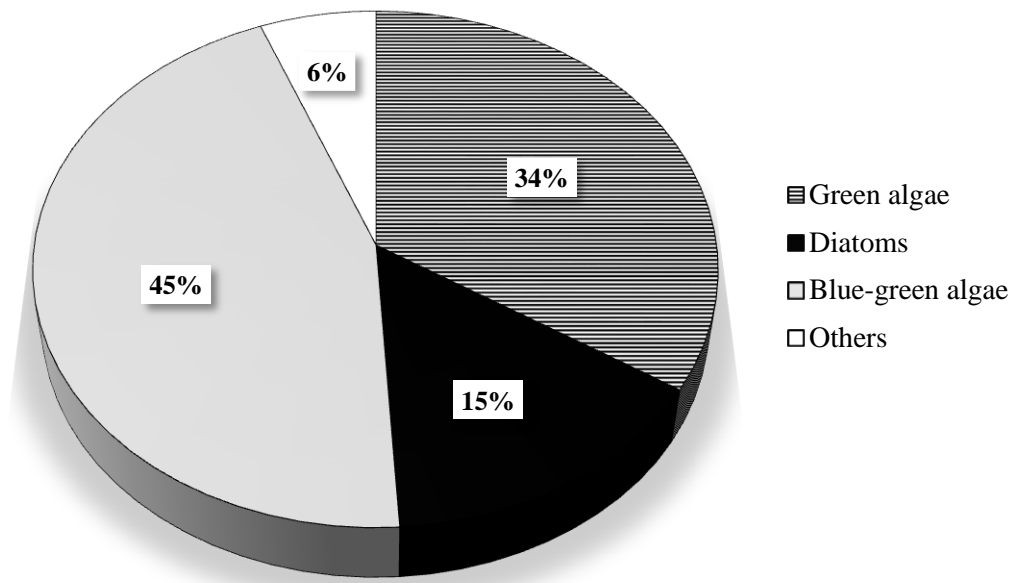


Fig. 2. Composition of phytoplankton in Belbella Reservoir

Blue-green algae can regain their vertical position quickly owing to their effective buoyancy mechanism associated with gas vacuoles (Reynolds, 1987). Consequently, mixing of the water column in this reservoir can affect the blue-green algae only temporarily. Owing to the same adaptive feature of blue-green algae, the poor underwater climate of the reservoir would not be a factor of overriding importance. In fact, positive buoyancy gives blue-green algae a competitive advantage over other algal groups as other algae like diatoms lack an effective mechanism of maintaining their vertical position in the water column.

The major zooplankton species identified in the reservoir were summarized in Table 3.

Table 3. List of zooplankton taxa identified in Belbella reservoir

Family	Genus
Copepoda	<i>Thermocyclops</i>
	<i>Eucyclops</i>
Cladocera	<i>Daphnia barbata</i>
	<i>Diapohanosma</i>
	<i>Moina</i>
	<i>Ceriodaphnia</i>
Rotifera	<i>Barchionus</i>
	<i>Asplanchna</i>
	<i>Lecane</i>
	<i>Polyarthra</i>
	<i>Hexarthra</i>
	<i>Filinia</i>
	<i>Trichocerca</i>

Rotifers were the most species-rich zooplankton group with seven genus. The second most important zooplankton group was the cladocerans. The copepods were, poorly represented, with only two genus, in the zooplankton community of Belbella reservoir. The zooplankton community of the reservoir was reflective of tropical African freshwater systems modified by altitude and salinity-alkalinity series (Green & Seyoum Mengestou, 1991).

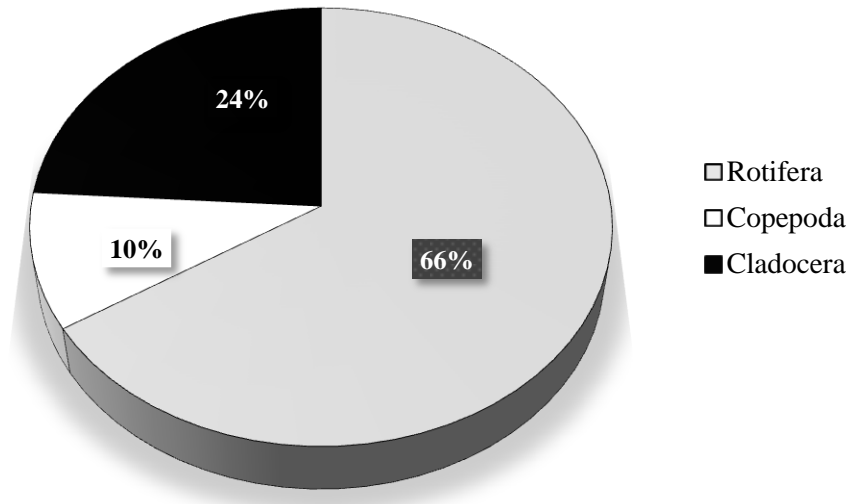


Figure.3. Composition of Zooplankton

The major species of rotifers (Fig. 3), which were responsible for the dominance of the group included *Brachionus* and *Filinia*. Green and Seyoum Mengestou (1991) also asserted that the zooplankton of Ethiopian lakes exhibit high rotifer diversity with *Branchionus* dominance.

#### ***Composition of fish species***

A total of 857 fish specimens were recorded from the three families during the study period (Fig. 4). The species was *Oreochromis niloticus* from the family Cichilidae (86 %) and the remaining was *Cyprinus carpio* and *Labeobarbus intermedius* from the family Cyprinidae 4 % and 10 % respectively (Fig. 4).

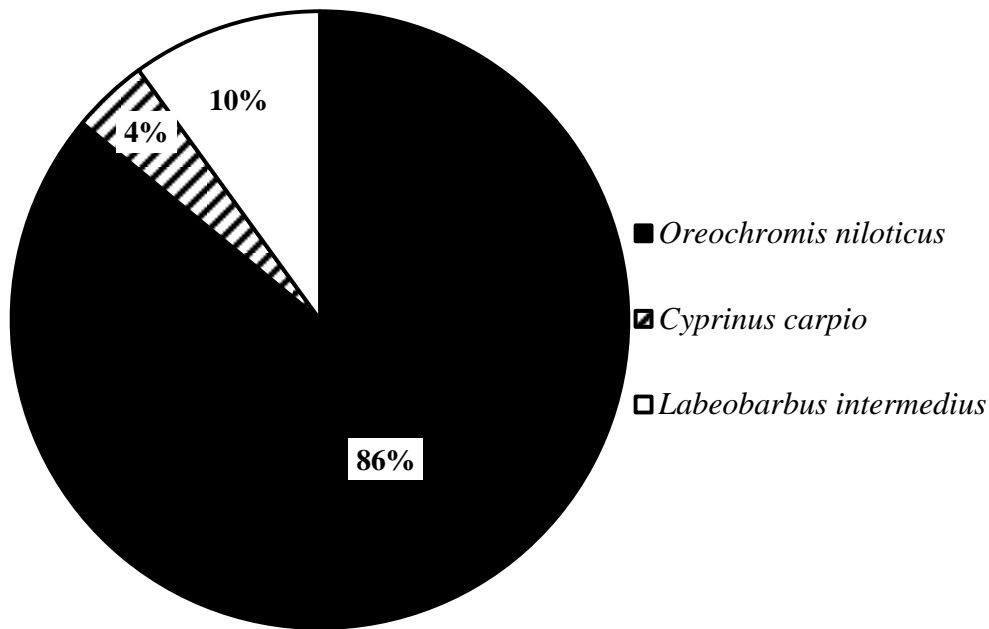


Figure 4. Composition of fish species in the waterbodies

***Some biology of the dominant fish species***

Length-Weight relationship of *Oreochromis niloticus*: The Length-weight relationship of *O. niloticus* in the reservoir was curvilinear, show isometric growth and statistically highly significant ( $P < 0.05$ ) (Figure 5).

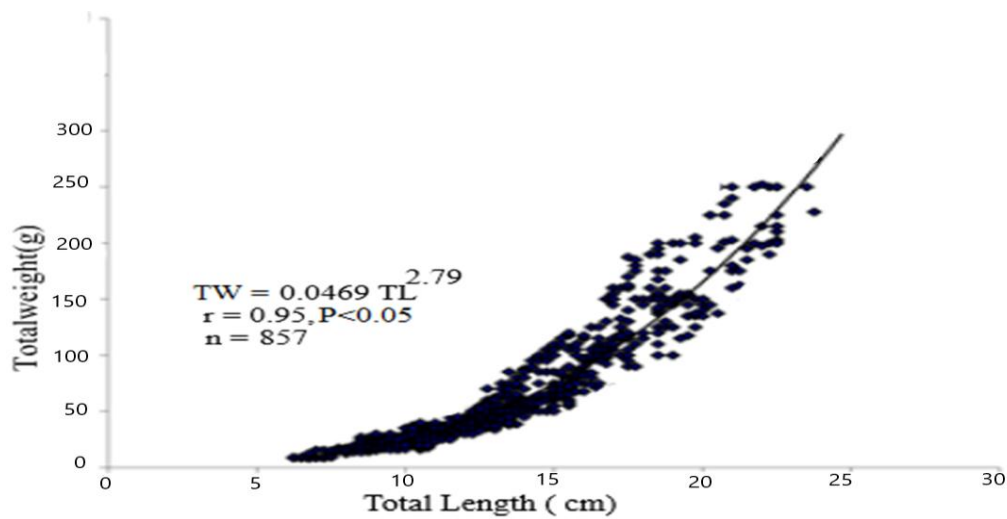


Figure 5. Length-weight relationship of *O. niloticus* in the reservoir



Sex ratio, length at maturity and fishing gears selectivity of *Oreochromis niloticus*: Sex ratio results of *O. niloticus* were presented in Tables 4. The ratio was not significantly different from 1:1 for all sampling months (Table. 4). In addition, the overall sex ratio (1:0.74) was also not significantly different from 1:1 (Table 4).

Table 4. Pooled sex ratio of the *O. niloticus* (From July 2018 - June 2021).

Month	F	M	F:M	X <sup>2</sup>
Sep.	73	57	1:0.78	0.086
Oct	47	31	1:0.65	0.071
Nov.	36	24	1:0.66	0.072
Dec.	16	10	1:0.63	0.069
Jan.	11	7	1:0.64	0.070
Feb.	14	9	1:0.64	0.070
Mar.	17	13	1:0.76	0.084
Apr.	21	18	1:0.86	0.096
May	42	25	1:0.59	0.062
Jun.	48	51	1:1.06	0.59
Jul.	73	59	1:0.8	0.214
Aug.	94	61	1:0.65	0.072
Total	492	365	1:0.74	0.081

The 50% maturity length ( $L_{50}$ ) of *O. niloticus* was estimated to be 18 cm total length for females and 17.5 cm total length for males (Fig.6). On the average, males appeared to attain sexual maturity at a relatively smaller size than females. As reported for both temperate and tropical aquatic ecosystems, males attain maturity at a smaller size than females (Tempero *et al.*, 2006; Britton *et al.*, 2007). Size at first sexual maturity of *O. niloticus* in Rift Valley Lakes (Lake Ziway, Koka and Langano). This might be due to the high fishing pressure and productivity of the water body.

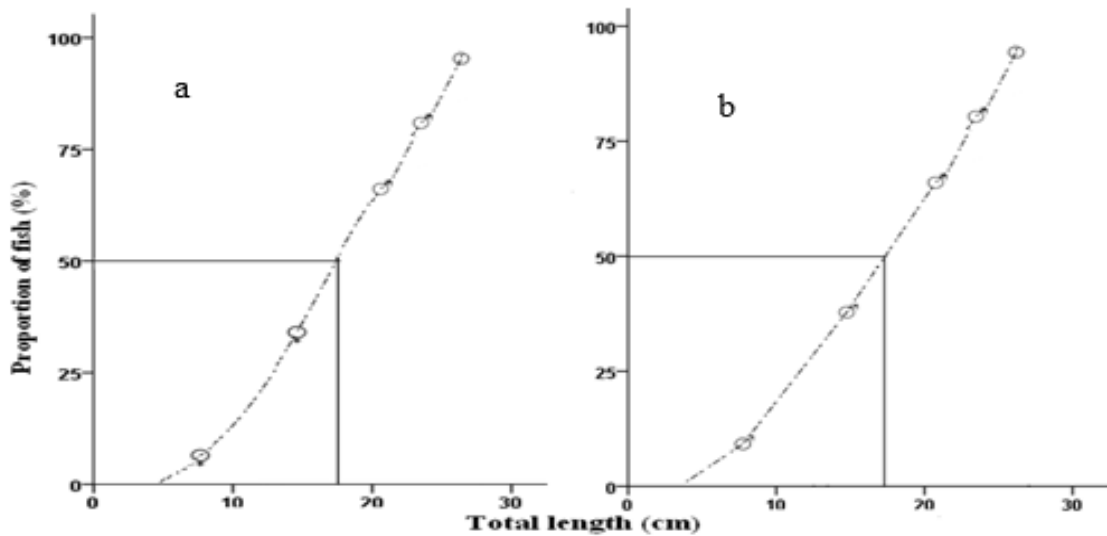


Figure 6. The proportion of fish in different length groups of females (a) and males (b) of

### *O. niloticus* in Belbella Reservoir

The catch size frequency distributions of the fish caught by the gillnet was indicated in figure 7. The current mesh size (10 cm) used in the reservoir was captured the fish with the range of length class between 16 -20 cm that coincide with at their  $L_{50}$  around 18 cm and 17.5 cm respectively for female and male (Figure 6 and 7). Hence, care should be taken not to lower the mesh size any further to the immature fish will be affected by the fishery.

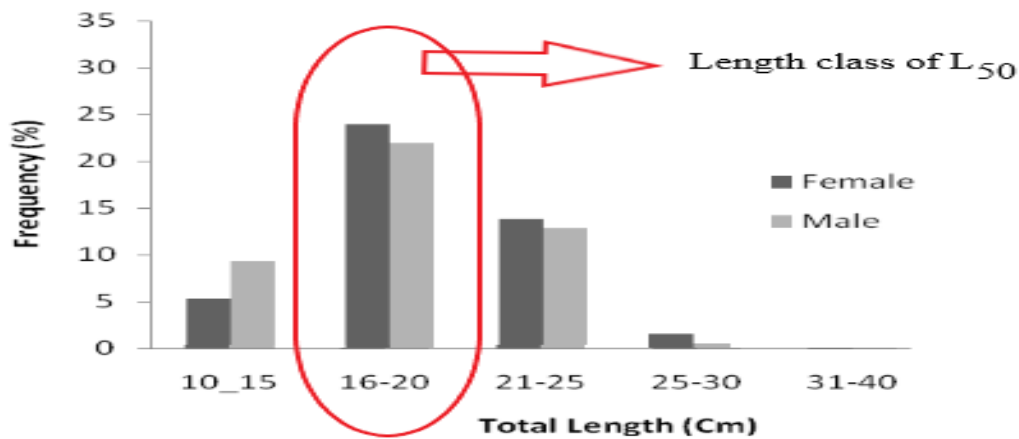


Figure 7. Number of fish catch per length class of *O. niloticus* using 10 cm mesh size gill net

The results of gill selectivity show that the use of recommended gillnets with mesh sizes that select fish below the size at first maturity ( $L_{50}$ ) of the target resources for the fish that reduced the spawning stock (Figure 6 and 7).

### Conclusions and Recommendations

Belbella reservoir is a very low salinity turbid eutrophic body of water that supports phytoplankton community, which is primarily constituted by cyanobacteria, green algae and diatoms. The reservoir water is suitable for human and animal consumption in light of the levels of aggregate chemical parameters and such physical variables as turbidity. The acceptability of the water from Belbella reservoir for human consumption is, however, questionable in light of cyanotoxins and heavy metals as the reservoir harbors potentially toxic cyanobacteria of high levels of abundance and is found in the proximity of floriculture industries that use a variety of chemicals.

The family Cichilidae dominates the fish fauna of Belbella Reservoir. *Oreochromis niloticus* was the most abundant and commercially the most important fish species under the family in the reservoirs. Based on percentage composition, *O. niloticus* was relatively the most dominant fish in the reservoir, which contributed to 86 % of the total catch. Among the Cyprinids, *C. carpio* and *L. intermedius* contributed 4 % and 10 %, respectively, of the total fishes catch. From the length-weight relationship, it can be stated that the only one dominant fish species (*O. niloticus*) show isometric growth and the length-weight relationships were also curvilinear. The smallest sexually mature male *O. niloticus* was 17.5 cm TL and the female was 18 cm TL.

Fishing technology on Belbella Reservoir was not modern in its nature and only makes use of traditional boats of wooden manual boat. Gillnets of not recommended mesh size was practiced on the water body that decreased the fish resources of the reservoir. Hence, as recommendation, with increase in nutrients and more degradation in the catchment, fish species composition may continue changing in the future. Therefore, monitoring of fishes and management of nutrient inputs should be carried out on regular basis and the shore of reservoir should be restored with macrophytes and protected in order to control external nutrient loading.

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# Preliminary study on fishery status during the initial filling of Didesa Reservoir

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## Abstract

*Scientific studies on water quality and productivity can contribute to the assessment of the suitability of water for particular uses such as domestic, irrigation, sanitation, fish production. The primary productivity of different water bodies has been widely investigated to assess the fish production capacity of a water body to formulate fishery management policies. This study aimed at identifying the retained fish species during the initial impoundment of the Didesa reservoir and studying the fish establishment feature of the newly built reservoir for estimating their fisheries production potential. Three fish species of two families were recorded in Didesa Reservoir. In the present study, *Labeobarbus intermedius* (Rüppell, 1835) dominated the catch, followed by *Oreochromis niloticus* (Linnaeus, 1758) and *Labeo cylindricus* (Peters, 1852). Length-weight relationships were highly significant with high coefficients of determination ranging from 0.97 for *Oreochromis niloticus* up to 0.990 for *Labeobarbus intermedius*. The length at first sexual maturity ( $L_{50}$ ) was 32.9 cm TL while it was 25.4 for *Oreochromis niloticus* at Didesa reservoir.*

**Keywords:** limnological parameters, fish biodiversity, reservoir fishery

## Introduction

Reservoir development creates an invaluable option for the production of fisheries, in areas where there is a shortage of fish products. Reservoirs create a habitat for the maintenance of critically important fisheries worldwide (Agostinho *et al.*, 2016), with the potential to increase food security and rural livelihoods in tropical regions (Arantes *et al.*, 2019).

The negative impacts of impoundments have been well documented, predicting changes in fish assemblage structure and consequences for fisheries is challenging because of the high diversity and complexity of tropical rivers (Monaghan *et al.*, 2020). Impacts from hydrologic alteration are unevenly distributed within fish assemblages, with certain species being severely impacted while others thrive in reservoirs created by dams. Shifts in species abundances affect ecological processes and important ecosystem services, such as nutrient cycling and fisheries production (Yurk and Ney, 1989). Reservoirs impact the spatio-temporal patterns of fish community structure and fisheries production by obstructing migration routes, altering sediment transport and water quality, promoting invasions by exotic species and biotic homogenization, and favoring generalist over specialist species (Teame *et al.*, 2016).

Studies on the responses of fish stocks to environmental changes caused by dams are critical for mitigating the socioeconomic and ecological impacts of these projects, particularly in tropical regions which are faced with food insecurity and malnutrition.

## Materials and Methods

### *Study area*

Didessa Reservoir is located along Didessa River, one of the largest tributaries of the Blue Nile River. The major tributaries of to Didessa River among others include Rivers of Wama from the east, Dabana from the west, and Angar from the east. The catchment area at the Dam site is 5,280 km<sup>2</sup>, and it extends to area 8 districts of the two zones (Jima-Arjo and Bedele) of the National Regional State of Oromia. When completed the Dam will be 40.6 m high earth and rock-fill dam

### *Fish sampling*

The fish communities were sampled by multifilament gill net having (60, 80, 100 and 120 mm) stretched mesh size. Immediately after capture Fish Species were identified, and the total (TL) and total weight (TW) were measured and weighted to the nearest 1mm and 0.1g respectively.

### *Data analysis*

Length-weight relationships parameters were estimated by a linear model fitted to log-transformed data using the following formula:

$$\log_{10} W \text{ (g)} = \log_{10} a + b * \log_{10} \text{ TL (cm)}$$

Where, W is the fish weight, TL is the total fish length and a and b are LWR parameters.

Length at which 50% of both sexes reach maturity (L50) was determined from the percentages of mature fish that were grouped in 1 cm length classes and fitted to the logistic equation described by (Echeverria 1987).

$$PL = (\exp(\alpha + \beta L)) / (1 + \exp(\alpha + \beta L))$$

Where, PL is the proportion of mature fish at length (L) and L, is the total length (cm), and  $\alpha$  (the intercept) and  $\beta$  (the slope) of least-squares estimates.

The length-frequency from gillnet fleets was corrected to provide an unbiased estimate of the length structure by determining the gillnet selectivity by using SELECT (Share Each Length's Catch Total) method. The SELECT method applies maximum likelihood, which estimates selectivity parameters from a general log-linear model (Millar 2003).

Catch data were pooled by mesh size into 1 cm length classes, and the midpoint of each size class was used to estimate a selectivity curve for each mesh size. The four gillnet selectivity models (normal location, normal scale, gamma, and log-normal) were fitted to the data by using the “gillnet functions” package in R statistical software (R Core Team and 2020). For each model, the data were fitted under the assumptions of equal effort and proportional effort to the size of the mesh. The goodness of fit statistics in the form of model deviance was used to choose the best model.

### ***Fish production estimation***

Several predictive models, based on a variety of morphological physicochemical and biological parameters, have been developed to provide a general indication of potential fish yields from lakes and reservoirs (MRAG, 1995). From several models developed by MARG, 1995 three models were selected due to their documented positive relationships between Catchment area, reservoir area, and maximum with fish species richness and production.

Model 1: Area (km<sup>2</sup>), Catchment area (km<sup>2</sup>) and Rainfall (mm), (based on 32 Lakes and Reservoirs, r<sup>2</sup> = 0.844)

$$\ln(\text{Catch}) = -10.502 + 0.484 * \ln(\text{Area}) + 0.45 * \ln(\text{Catchment}) + 1.57 * \ln(\text{Rainfall})$$

Model 2: Area (km<sup>2</sup>), Altitude (masl.), (based on 132 Lakes and Reservoirs, r<sup>2</sup> = 0.711)

$$\ln(\text{Catch}) = 3.844 + 0.891 * \ln(\text{Area}) - 0.342 * \ln(\text{Altitude})$$

Model 3: Area (km<sup>2</sup>), Maximum depth (M), (based on 83 reservoirs, r<sup>2</sup> = 0.820)

$$\ln(\text{Catch}) = 2.625 + 0.879 * \ln(\text{Area}) - 0.121 * \ln(\text{Zmax})$$

### **Results and Discussion**

Three fish species of two families were recorded in Didesa Reservoir (Table 6). In the present study, *Labeobarbus intermedius* (Rüppell, 1835) dominated the catch, Followed by *Oreochromis niloticus* (Linnaeus, 1758) and *Labeo cylindricus* (Peters, 1852).

Table 6. Fish species composition of Didesa Reservoir

<b>Family/ Species</b>
<b>Cichlidae:</b> <i>Oreochromis niloticus</i> (Linnaeus, 1758)
<b>Cyprinidae:</b> <i>Labeobarbus intermedius</i> (Rüppell, 1835) <i>Labeo cylindricus</i> (Peters, 1852)

### ***Length-weight relationships***

LWRs were highly significant with high coefficients of determination ranging from 0.97 for *Oreochromis niloticus* up to 0.990 for *Labeobarbus intermedius* (**Error! Reference source not found.**). The exponent is near to 3.00 for both species, with a central value of 2.9 for *Oreochromis niloticus* and 3.14 for *Labeobarbus intermedius* in the Didesa reservoir.

Table 2. Length Weight relationship of fishes in Didesa Reservoir

Species	TL (cm)		Regression parameters				
	Min	Max	a	b	b CI 95 %	p	R <sup>2</sup>
<i>Oreochromis niloticus</i>	19.5	33	0.0256	2.9184	2.69 - 3.15	<0.001	0.976
<i>Labeobarbus intermedius</i>	20.5	54	0.0060	3.1471	2.90 - 3.38	<0.001	0.990

### *Sexual maturity*

The smallest fully ripe female captured during the sampling period was 27.5 cm for *Labeobarbus intermedius*, the length at first sexual maturity ( $L_{50}$ ) was 32.9 cm TL while it was 25.4 for *Oreochromis niloticus* at Didesa reservoir. The  $L_{50}$  obtained for *O. niloticus* was smaller than the findings from Lake Chamo (42 cm TL) (Yirgaw Teferi *et al.* 2000), Lake Victoria (30.81cm and 34.56 cm TL) (Njiru *et al.* 2004). However higher than Lake Hawassa (18.8 cm and 19.8 cm TL) (Demeye 1994), Fincha Reservoir (21.2 cm and 23.4 cm TL) (Fasil Degefu *et al.* 2012), of the same species, respectively. This indicated that *O. niloticus* in Lake Didesa reservoir exhibited a rapid first sexual maturity.

### *Gear selectivity*

The results of the SELECT method fitted for *L. intermedius*, the dominant fish species in Didesa Reservoir are given in (Table ). The Log-normal model provided the best fit as it had the lowest deviance value, indicating that both models can describe the gillnet catches proportionally for this species. The Normal location (fixed spread) model was the worst fit model.

Table 3 Gear Selectivity parameters of Didesa Reservoir

Model	Fishing power relative to mesh size		dof
	Model deviance	Null deviance	
Normal	62.48	62.01	92
Normal_sca	53.61	71.38	92
Gamma	47.61	51.25	92
Lognormal	42.62	47.64	92

### *Fish production estimation*

Based on the morphometric characteristics of the reservoirs, an average fish production of 183 t/year was estimated for the Didesa reservoir (Table ).

Table 4 Estimated annual fish production potential of Didesa Reservoir

Model	Total yield (t/year)
1	123
2	143
3	285
Average	183



This study represents the first published assessment of length-weight relationships for several species and contributes to the knowledge base of these species for which data were limited. Analysis of length and weight data can be used to mathematically describe this relationship so that one can be converted to another, or to measure variation in expected weight, providing a measure of overall well-being (King 1996).

The range of the exponent  $b$  of the LWRs was consistent with the expected range of (Froese 2006), except *Labeo cylindricus* which has shown unexpected  $b$  value, This could be due to the narrow range of fish caught during the study for the species. Further, LWR parameters determined for populations with those published in Fish Base were compared (Froese and Pauly 2000). Parameters  $a$  and  $b$  were within 95% of observed parameter values based on Bayesian LWR estimates calculated in Fish Base (Froese *et al.* 2014) records for *Labeobarbus intermedius*, *Bagrus docmak* and *Mormyrus kannume*.

Length-weight relationships (LWRs) are useful for estimating the biomass of stocks when a number of specimens by length class are obtained (Froese, 2006) and the parameters  $a$  and  $b$  of LWRs are useful in stock assessment (King 2013).

### **Conclusion and Recommendation**

Potential species and yield from reservoirs have been related to the trophic status of the water body. Estimation of potential fish yield has to be estimated for each reservoir to determine the appropriate stocking management regime, which will avoid problems due to over and under-stocking of fish seed. Therefore, this study generated a baseline database on ecology, morpho-edaphic parameters, natural fish diversity patterns, and potential fish yield for sustainable utilization and enhancement of fish production in the reservoirs.

In conclusion, results show a clear adaptation and fish colonization pattern for some riverine fisheries in the reservoir during initial impoundment. This helps in the establishment of successful fisheries in the Reservoir. The results also highlight the importance of fisheries management for of *Oreochromis niloticus* as successional patterns of fish colonization to improve the fisheries in the future.

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# Effect of Stocking Density on Growth Parameters of African Catfish (*Clarias gariepinus*) in Small Scale Aquaponics System

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## Abstract

*Aquaponics is a fish-plant recirculation system where nutrients from the fish culture are absorbed by the plants for growth. Stocking density of fish in the recirculation system affects growth performance of the fish and the integrated plants in Aquaponics system. The aim of the current study was to assess the growth performances and survival rates of African catfish stocked into the system at different stocking densities. Four different stocking densities by weight of African catfish, 1.93 kg/m<sup>3</sup>, 3.87 kg/m<sup>3</sup>, 5.81 kg/m<sup>3</sup> and 7.75 kg/m<sup>3</sup> were evaluated under Aquaponics system with lettuce vegetable grown in nutrient film technique. The fish were grown for 65 days where fish length (cm) and fish weight (g) data were collected every two weeks. The survival percentage and the fish yield were calculated at the end of the experiment. Total fish yield from the treatments were 2.35kg/m<sup>3</sup> in T1, 3.40kg/m<sup>3</sup> in T2, 4.80kg/m<sup>3</sup> T3 and 5.97kg/m<sup>3</sup> in T4. The results show that though the final mean length and weight of the fish in T1 was higher than the others, there was no statistically significant differences in fish growth parameters between all the treatments. However, the fish survival rate decreased significantly from 92% in T1 to 60% in T4 as the stocking density increased. Accordingly total yield of fish increased only in T1 while the yield decreased below the biomass of the fish at stocking in T4, T3 and T2. Hence, stocking densities above 1.93 kg/m<sup>2</sup> of African catfish affects survival rates and yield of the fish in the small Aquaponics system.*

**Key words:** stocking density, African catfish, Aquaponics.

## Introduction

The globally growing and developing human population within the save limits of the planetary boundaries increasingly depends on the level of sustainability of the agricultural systems of the future (Rockstrom *et al.*, 2009). The systems must produce higher yields by using fewer resources, but also entail fewer emissions when compared with those of the present times. Future food production must be intensified to meet the increasing food demands (Rockstrom *et al.*, 2013) and intergradations with other farming systems is required.

Aquaponics is one of the integrated sustainable food production technologies (Lennard & Goddek, 2019) that link hydroponics with a recirculating aquaculture system (RAS) intending to increase fish and vegetable production (Konig *et al.*, 2018). It is a bio-integrated system that usually links RAS with hydroponic vegetable, flower, and/or herb production and now gaining increased attention globally. In Aquaponics system, the waste products of one biological system serve as nutrients for a second biological system. The integration of fish and plants in Aquaponics results in a poly-culture that increases diversity and yields multiple products serves becoming a model of sustainable food production. Water is re-used in

Aquaponics system through biological filtration and recirculation. It enables local food production which provides access to healthy foods and enhances the local economy.

African catfish (*Clarias gariepinus*) is one of the fish species that can be cultured in Aquaponics system whereby the waste from the fish is used as a source of macro- and micro-nutrients required by plants which the total concentration and the ratio influence the productivity and quality of plant produce (Seawright *et al.*, 1998).

So far, only few studies registered all the essential plant nutrients in water of RAS process, such as for Nile tilapia (*Oreochromis niloticus*) (Delaide *et al.*, 2017). Compared with African catfish, Tilapia resulted in better yields in lettuce, cucumber, tomato (Palm *et al.*, 2014). Hence, the aim of this research was to assess the growth performances and survival rates of African catfish stocked at different stocking densities in small scale Aquaponics.

### **Materials and methods**

Four sets of Aquaponics were installed in an aquaculture setting at Batu Fish and other Aquatic Life Research Center. Each set consisted of fish tank, bio-filter tank and NFT for plant growth. Each fish tank was filled with 500 L water. The NFT in each set consisted three rows of hydroponic gullies, each with 23 bio-ball points for single plant growth. Bio-filter tank was filled with substrate on which bacteria converting fish waste into plant nutrient live. Water was pumped from the fish rearing tank to the bio-filter unit with a 15 watt submersible water pump, pass to Hydroponic unit and back to the fish tank via gravity flow and make full recirculation system. A back flow of water from pipe in a spray form was used to aerate the fish tank. Temperature of water in the system was ranged from 19 °C at night to 23 °C after noon.

African catfish (*C. gariepinus*) were stocked in to the fish tanks at stocking densities of 1.93 kg/m<sup>3</sup>, 3.87 kg/m<sup>3</sup>, 5.81 kg/m<sup>3</sup>, 7.75 kg/m<sup>3</sup>. The fish at stocking have mean total length of 22.75±5.00 cm and mean weight of 77.55±48.49 g. The fish were uniformly distributed size wise to each fish tank.

The fish were fed with locally prepared feed at feeding rate of 3% of their total body weight daily; the feed provided twice a day at 3:00 am and 9:00 pm. The fish were reared for 65 days and growth data recorded each two weeks.

Seeds of the lettuce (*Lactuca sativa longifolia*) were sown and sprouted in the sowing trays before being transferred to the hydroponic system. After 21 days the lettuce seedlings were transplanted to the NFT of the Aquaponic system.

### ***Fish growth parameters data***

Fish was sampled for body weight and body length measurement every two weeks to determine how much the fish have grown. Total lengths of each sampled fish were measured individually using measuring board with ruler at 0.1 cm accuracy. Weight of the fish were measured using digital weight balance (Ohaus portable balance) at 0.1 g accuracy. Mortality of the fish in each treatment unit was calculated at the end of the experiment.

**Specific growth rate and survival rate of fish**

Specific Growth Rate (SGR) was calculated from average initial weight ( $W_i$ ), average final weight ( $W_f$ ), and experimental duration (dt), while survival rate was calculated from number of fish stocked at the start of the experiment and final number of fish harvested at the end of the experiment according to the following formulas:

$$\text{SGR (\% day}^{-1}\text{)} = ((\ln W_f(\text{g}) - \ln W_i(\text{g})) / \text{dt}) \times 100$$

Where,  $W_f$  is mean final body weight (g) of fish at the end of the experiment;  $W_i$  is mean initial body weight (g) of fish at stocking; dt is experiment duration in days.

$$\text{Survival rate (\%)} = (\text{Nf}/\text{Ni}) \times 100$$

Where, Nf is number of fish survived to the end of the experiment; Ni is number of fish stocked at the beginning of the experiment.

Data on average length and weight of the fish were analyzed at the end of the experiment. Differences in mean values of the parameters between the treatments were analyzed using One-way analysis of variance (ANOVA) and further mean separation was done by LSD at 0.05 level of significance. The results were presented in tables and graphs.

**Results and Discussion**

Overall production performance as measured by mean growth and survival rate of the fish were presented in table 1.

Table 1. Mean length and weight of African catfish (*Clarias gariepinus*) at stocking and after 65 days of experimental period in four stocking densities.

Treatment	mean length(cm)		mean weight(g)		Fish biomass (kg/m <sup>3</sup> )		SGR (%/day)	Survival rate (%)
	Initial	Final	Initial	Final	Initial	Final		
1	22.75±5.00	26.00 ±4.01	77.55±48.49	110.2±48.09	1.93	2.35	0.54	92
2	22.75±5.00	24.31 ±3.80	77.55±48.49	93.11±49.13	3.87	3.40	0.28	72
3	22.75±5.00	24.35 ±2.90	77.55±48.49	95.57±40.05	5.81	4.80	0.32	67
4	22.75±5.00	24.49±2.74	77.55±48.49	93.98±31.09	7.75	5.97	0.30	60
		p>0.05		p>0.05				

The initial mean length and weight of the fish were similar across the four treatments at the beginning of the experiment. The final mean length and weight of the fish after 65 days, varied slightly across the treatments with fish in lower stocking density of 1.93 kg/m<sup>3</sup> (T1) attained relatively higher final length and weight. than the other treatments, though the differences were not statistically significant (table 1). Similarly, the specific growth rate (SGR) of the fish in T1 was higher than the other treatments. The differences in the parameters were related to the effects of fish stocking densities.

The specific growth rate recorded in the current study was better in T1 (0.54) than the rest of the treatments; this highest value was very poor as compared to the growth rate recorded for the same species

in Aquaponics system from 0.79 to 1.15 by Yuan *et al.* (2019). The poor growth rate in the current experiment could be due to poor aeration system and no temperature regulation in the system, both factors affecting fish growth.

The final fish biomass at the end of the experiment was increased only in the first treatment (T1 with the stocking density of 1.93 kg/m<sup>3</sup>) as compared to the biomass at stocking. The fish biomass in the treatments T2, T3 and T4, with stocking densities greater than 1.93 kg/m<sup>3</sup> were decreased below the biomass at stocking. Fish survival rate decreased with the increasing biomass at stocking where T1 with lowest stocking density attained the highest survival rate of 92% when the survival rate decreased to 72%, 67% and 60% in T2, T3 and T4 respectively. This may probably be due to the crowded condition created as a result of the higher densities and the resulting competition, even though the fish were fed to satiation. Similar results were also reported by Suresh & Lin (1992) and Ashagrie Gibtan *et al.* (2008).

Stocking density is a major factor that determines profitability from production systems, because it directly affects fish survival, growth, behavior, health, water quality and feeding requirements (Gibtan *et al.*, 2008). Positive and negative relationships between stocking density and growth have been reported (Abou *et al.*, 2016). High stocking density is considered a potential source of stress for fish (Abou *et al.*, 2016 & Ellis *et al.*, 2002), with a negative effect on specific growth rate and final weight (Abou *et al.*, 2016 & Ullah *et al.*, 2018) as well as survival and feeding rates (Das *et al.*, 2016 & Rowland *et al.*, 2006). Regarding survival, regression analysis indicated a significant linear decrease in survival with increasing stocking density and although no definitive reason for this was apparent, it is probable that the decreased water quality may be partially responsible for this phenomenon, seeing that decreased water quality and increased stocking density are both factors which could increase fish stress and therefore mortality (Babmann *et al.*, 2017).

Mortality rates of African catfish were reported by Hossain *et al.* (1998) with 10–20% (in larvae); Toko *et al.*, (2007) with 0–4%; Adewolu *et al.* (2008) with 6–19%; and van de Nieuwegiessen *et al.*, (2009) with 0–3%. Palm *et al.* (2018). Increasing stocking density affected survival rate in the current experiment. The mortality in the current experiment were higher than those reports except in T1. The lower survival rate in the current experiment was due to insufficient aerating system in the fish unit and mortality caused when few individuals jumped out of the fish tank around the start of the experiment.

## **Conclusion and Recommendations**

Fish stocking density in terms of biomass affected the growth performance, survival and final yield of fish in small scale Aquaponics system. Stocking density beyond 1.93 kg/m<sup>3</sup> of African catfish critically decreased the fish SGR survival rate and the biomass. Hence the stocking density of the African catfish in similar small scale Aquaponics system should not exceed 1.93 kg/m<sup>3</sup> unless better biofilter and aeration system is installed in the recirculation system.

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# Evaluation of Water Hyacinth (*Eichhornia crassipes*) Nutritional Value as a Diet for Common Carp (*Cyprinus carpio*) Fingerlings

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## Abstract

*The experiment was conducted at Batu Fish and Other Aquatic Life Research Center to evaluate the effect of water hyacinth incorporated diet on growth parameters of common carp in concrete ponds. Fish diets were prepared in four different levels of water hyacinth (WH) compositions as energy source: T1 with 25% WH, T2 with 50% WH, T3 with 75% WH and T4 without WH inclusion as a control. A total of 312 Cyprinus carpio fingerlings having average weight of  $13.41 \pm 2.28$  g, mean total length of  $9.58 \pm 1.07$  cm and mean fork length of  $8.56 \pm 0.85$  cm were stocked into 12 ponds, each with 6 m<sup>2</sup> area, at a stocking density of 26 fish per pond. The experiment lasted for 90 days, during April 2021 to June 2021. Accordingly, the feed was iso nitrogenous and iso calorific with values of  $31.45 \pm 0.1$  crude protein and  $365.61 \pm 7.1$  kcal/100 g for protein and energy, respectively for each treatment. The fish were fed 5% their body weight two times a day at 3:30 and 10:00 local time. At the end of the experiment, no significant difference ( $p > 0.05$ ) was observed between all treatments in all growth parameters. However, there was a variation in final mean value of the parameters between the treatments. Accordingly, a control group attained highest final weight, length, PGR and FCF followed by T1 and T2 respectively while the least value was observed in T3. Hence, based on the mean values obtained in the treatments, replacement of wheat bran by *Eichhornia crassipes* as energy source for *C. carpio* diet in the ratio of 25 % ( T1), 50% (T2) and 75 % ( T3) are recommended.*

**Keywords:** *Cyprinus carpio*, *Eichhornia crassipes*, feed, fingerlings, growth

## Introduction

Aquaculture is the fastest growing global food production sector in the recent years. FAO (2019) reported that world total aquaculture production, including aquatic plants, reached at 111.9 million tonnes in live weight in the year 2017 of which 80.1 million tonnes belongs to aquatic animals. From 20 major cultured fin fish species, common carp is ranked first followed by Nile tilapia (*Oreochromis niloticus*) (FAO, 2019). With the rapid growth in the aquaculture sector in the recent years, the demand for quality fish feed is continuously increasing. The fish feed used in aquaculture is expensive, irregular and short in supply in different world countries especially developing countries due to unavailability of fish feed supplying organizations (Bag *et al.*, 2011). Thus, providing quality fish feed has become a first priority in aquaculture sector (Dorothy *et al.*, 2018).

For this reasons, research in fish nutrition that will utilize locally available and alternative low-cost feed ingredients of plant protein sources and fabricated equipment without reducing the quality of the feed is urgent and crucial to the overall success of aquaculture development, growth and expansion in Africa (Sulieman and Lado, 2011; Zenebe Tadesse *et al.*, 2012). Conducting research on alternative raw materials is used to reduce dependence on imported raw materials from developed countries. For maximum growth

of fish, optimum protein content in the feed is necessary. Water hyacinth is one of the materials that could potentially be used as feed ingredients in fish feed (Zaman *et al.*, 2017).

Water hyacinth is a potential ingredient in farm-mixed feeds for the farming of herbivorous or omnivorous freshwater fish in simple farming systems where it is available at low cost ( Mohapatra, 2015). Water hyacinth meal has a positive nutrient utilization effect on fish growth and that it can assist fish farmers in ensuring sustainable fish production through production of least-cost diets (Sotolu & Sule, 2011). Water hyacinth can partially replace conventional energy sources (wheat bran) in fingerling Nile tilapia feeds. El-Sayed (2003), also evaluated the effects of ensilaged WH for Nile tilapia fingerlings using 5% sugar cane molasses as an additive. The results indicate that the silage gave better performance than fresh WH as a replacement of wheat bran at a substitution level of 10 to 20%. A result reported by Derribew Hailu *et al.* (2020) also indicated that, growth of Nile tilapia at different proportion of water hyacinth was a promising result. Hence, the current study was aimed to evaluate growth performance of common carp grown under by feeding water hyacinth incorporated diet.

## **Materials and Methods**

### ***Experimental fish***

Three hundred and twelve similar size *Cyprinus carpio* fingerlings were collected from Koka reservoir and transferred to experimental ponds of Batu Fish and Other Aquatic Life Research Center. The size of fingerlings used for this experiment was  $13.41 \pm 2.28$  g average weight, average total and fork lengths of  $9.58 \pm 1.07$  cm and  $8.56 \pm 0.85$  cm respectively.

### ***Water hyacinth collection***

Water hyacinth leaf was collected from Lake Denbel while noug cake, molasses, iodized salt, vitamins and wheat bran were purchased from local market. Fish offal was obtained from fish processing shade of Ziway/Batu fishery cooperatives around Hara Denbel (Lake Ziway).

### ***Feed preparation***

Water hyacinth leaf was collected from Hara Denbel/Lake Ziway and allowed to air dried (in shade area) to about 65-70% % moisture content in Batu Fish and Other Aquatic Life Research Center for 72 hrs and chopped by using chopper machine.

After chopping, water hyacinth was mixed with 20% molasses (6.4 kg Molasses to 32 Kg WH) and 2 ml of Acetic acid ( $\text{CH}_3\text{COOH}$ )/kg with continuous mixing. Then it was sealed and put into black plastic dram to ferment an aerobically at a temperature of 25.6 °C but away from direct sun light for 30 days. After 30 days of fermentation, the sealed and fermented water hyacinth was opened and direct sun and air dried for three days (72 hrs). Finally the fermented and sun dried water hyacinth was manually grinded with pistil and mortar and analyzed for its nutrient content at laboratory of Oromia Agricultural Research Institute (OARI) as of AOAC (2005) procedure shown in Plate 1 and mixed with standard diets for final preparation of fish feed after analysis. In the same way, noug cake and wheat bran were purchased from private animal feed trading company and analyzed for their nutrient content also.

*Fishmeal* was prepared by cooking, pressing, drying, and grinding of fish or fish offal into a solid where most of the water and some or all of the oil was removed. The raw material used for production of fishmeal was Nile tilapia offal which was collected from Ziway-Batu fishermen cooperative processing shade and cooked according to the procedure of (Abera Degebassa *et al.*, 2008) as follows.

*Cooking:* The purpose of the heating process is to liberate the oil from the fat depots of the fish, and to condition the material for the subsequent treatment. The fresh fish offal was cooked-until the flesh opaque and separates to about 95 °C -100 °C for about 30 minutes (FAO,1986). In this step, oil from fat deposits and moisture content liberated and un-wanted materials was sterilized.

The cooked offal was left to settle in the barrel over night. That is from 6 pm to 8 am (morning) to separate the oil, water and solid components.

*Pressing:* The purpose of the press was to squeeze out as much liquid as possible from the solid phase. This is important not only to improve the oil yield and the quality of the meal, but also to reduce the moisture content of the press cake as far as possible, thereby reducing the fuel consumption of the dryers and increasing their capacity. The pressed by-products were spread on the laminated tin on rack for drying in the sun.

*Drying:* The purpose of the drying process is to convert the wet and unstable mixture of press cake, decanter sludge and concentration into a dry and stable fish meal. In practice, this means drying to a moisture content below 12%, which generally may be considered low enough to check microbial activity. This drying was done by heating the material to a temperature where the rate of evaporation of the water is considered satisfactory.

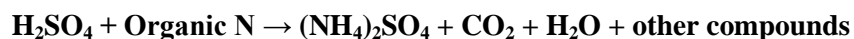
*Grinding, packaging and Storing:* The dried meal was then ground using Laboratory miller to appropriate size. The ground powder was packaged using polyethylene bags stored in dessicator until analyzed.

### ***Determination of proximate composition***

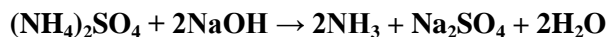
Moisture percentage was determined by weighing every petri dish and the wet sample. Consequently sample was dried in an oven at 105°C for 3 hours. Then petridish along with dried sample was placed in desiccators for 30 minutes. Eventually, the weight of Petri dish and dried sample was weight (AOAC, 2005). The moisture percentage was calculated using the following formula:

$$\text{Moisture \%} = 100 * ((\text{wet sample weight} - \text{dry sample weight})/\text{wet sample weight})$$

Crude Protein percentage was determined through the Kjeldahl method according to (AOAC, 2005). VELP DK Series Digestions Unit and a VELP UDK 159 Automatic Digestion were used subsequently. Every Kjeldahl flask was prepared to add 2 catalysts for digestion. This included 1 g of the dried sample, 0.2 g CuSO<sub>4</sub> and 7 g of K<sub>2</sub>SO<sub>4</sub>. For the actual digestion, also 12 ml of H<sub>2</sub>SO<sub>4</sub> was added. Digestion was completed at 420 °C for 1 hour. Eventually the sample was cooled down to 50 °C. The sulfuric acid converts all nitrogen in organic form into inorganic form that is stable and suitable for analysis:



For the distillation, distilled water was added. The nitrogen was separated from the digested mixture by steam distilling with the UDK 159 in order to extract ammonia from the alkaline solution. Sodium hydroxide (40%) was added to raise the pH and convert solid  $\text{NH}_4^+$  into gaseous  $\text{NH}_3$ . The distilled nitrogen was then trapped by adding boric acid.



For the final colorimetric titration, hydrochloric acid was added in order to react with the ammonia and the indicators. The volume of titrant that was needed to reach the end point, allows to automatically calculating the amount of nitrogen, expressed as % N.

Crude fat was determined using Soxhlet fat extractor, consisting of a rounded-bottom flask, an extraction chamber and a condenser. For 200 ml extraction chambers, 50 ml of diethyl ether was used as extraction solvent. Approximately 2 grams of dried sample in a filter was placed into the extraction chamber to conduct the fat extraction. The rounded-bottom flask was weighed before each sample in case some fat remained from the previous sample. The sample was subjected to 3 hours of extraction. Afterwards the rounded-bottom flasks containing the fat was placed in the oven to eliminate any remaining petroleum diethyl ether and was weighed again (AOAC, 2005). The fat percentage will be calculated using the following formula:

$$\text{Fat \%} = 100 * ((\text{weight rounded-bottom flask \& fat} - \text{weight rounded-bottom flask}) / \text{dry sample weight}).$$

To determine Crude ash, the sample was ashed in order to be able to analyze the mineral content afterwards. Two grams of the dried samples was placed in crucible cups that were weighed beforehand. Prior to placing the samples in the oven, they will be charred on hot plates for 30 minutes to prevent excess smoke production. The ashing itself will be done by placing the crucible cups in a 550°C oven for 8 hours (AOAC, 2005). The following formula was used to calculate the ash percentage:

$$\text{Ash \%} = 100 * (\text{weight ash} / \text{weight dry sample})$$

Proximate composition of water hyacinth, noug cake and wheat bran were analyzed in the same procedure of fish offal except the difference in ml of  $\text{H}_2\text{SO}_4$  used during protein determination (15 ml sulphuric acid used during fish offal protein determination while only 12 ml of sulphuric acid used for all plant source protein determination).

The percentage of total carbohydrate was calculated using the formula: 100-(percentage of ash + percentage of moisture + percentage of fat + percentage of protein) (Indrayan, A. K. *et al.*, 2005).

Nutritive value */Energy/* of the sample was determined by multiplying the values obtained for protein, fat and carbohydrate by 4.00, 9.00 and 4.00, respectively and adding up the values (Indrayan, A. K. *et al.*, 2005).

### ***Final formulated experimental feed***

After proximate analysis, the experimental feed was formulated for all treatments according to the following Table 1. A total of 15 kg feed was formulated for each treatment (5 kg/replicas).

Table 1: Experimental diets used as feed for *C. carpio* in different treatments.

No	Feed ingredients	T1	T2	T3	T4(control)
1	Fish meal (%)	20.30	20.67	20.97	20.0
2	NC (%)	47.4	48.20	48.93	48.4
3	WH (%)	7.46	14.33	20.71	0
4	WB (%)	22.4	14.33	7	28.0
5	Salt (%)	1.5	1.5	1.5	1.5
6	Minerals (%)	1	1	1	1
7	Total (%)	100	100	100	100
8	Total protein (%)	31.36	31.49	31.59	31.38
9	Gross Energy (kcal/100g)	369.54	362.84	357.03	373.02
10	Average GE	365.61±7.1			

### ***Experimental design***

Similar size *C. carpio* fingerlings were collected from Koka reservoir and transferred to experimental ponds of Batu Fish and Other Aquatic Life Research Center. The initial size of fingerlings used for this experiment was  $13.41 \pm 2.28$  g average weight,  $9.58 \pm 1.07$  cm total length and  $8.56 \pm 0.85$  fork length. A total of three hundred and twelve (312) fingerlings were randomly distributed to twelve (12) experimental ponds having an area of 6 m<sup>2</sup> each making a stocking density of twenty six fingerlings (26) per pond. The experimental fingerlings were fed with the experimental diets according to the treatments. Accordingly, the experimental feeds were iso nitrogenous and iso calorific with values of  $31.45 \pm 0.1\%$  crude protein and  $365.61 \pm 7.1$  kcal/100g for protein and energy, respectively for each treatment. The feeding frequency was two times per day at 5% of their body weight throughout the experimental period. Accordingly, there were four treatments with three replications and the experiment lasted for three months (90 days).

### ***Data collection***

Weight and length data of the fish were collected at stocking time and monthly throughout the experimental period as shown in plate 2. All water quality parameters were in the range of *C. carpio* throughout the experiment.



Plate 2. Fish length and weight data collection during experimental period.

### ***Data analysis***

Differences in weight, length and condition factor of fish in different treatment groups were analyzed using one way ANOVA in SPSS statistical package version 20. Duncan's multiple range test was applied to compare between means of the parameters at 0.05 level of significance. Accordingly, growth performances of fish were determined in terms of final individual weight (g). Survival rate (%), specific growth rate (SGR) and Fulton's condition factor (FCF) were also computed in the experimental study. Growth parameters of the fish were calculated according to standard equation given by Adebayo *et al.* (2004). Specific growth rate (SGR) was calculated as:

$$\text{SGR (\% d-1)} = [(\ln \text{ final body mass in g}) - (\ln \text{ initial body mass in g}) / \text{number of trial days}] \times 100;$$

Weight gain of fish in (g) was calculated as:

$$\text{Weight gain (g)} = \text{Final weight (g)} - \text{Initial weight (g) and}$$

$$\text{Survival rate (\%)} = (\text{Number of fish harvested} / \text{Number of fish stocked}) \times 100$$

Fulton's Condition Factor (FCF) is calculated as

$$\text{FCF} = \text{TW} / \text{TL}^3 \times 100,$$

Where, TW is final total body weight in g, TL final total length in cm.

### **Results and Discussion**

#### ***Proximate composition of fish feed***

The proximate composition of feed items analyzed at laboratory of OARI is indicated in Table 2. The protein content of *Eichhornia crassipes* collected from Lake Denbel was low (13.4% CP) as compared to that of Lake Koka with 19.57 % CP content (Derribew Hailu *et al.*, 2020). This variation may be caused by the difference in nutrient concentration of the two lakes (Denbel and Koka) and the difference in season of collection (January 2019 from Lake Koka and September 2020 from Lake Denbel). In January, plant nutrient may stagnate around specific area supporting plant growth while in early September the water may almost diluted and the nutrients required for plant growth become less. The proximate composition of the plant depends on nutrient availability of the water body on which it grows on (Mako *et al.*, 2011).

Table 2: Proximate composition of fish feed items.

Ingredients	Moisture (%)	Ash (%)	CP (%)	CF (%)	Fibers (%)	Carbohydrate	Gross energy (Kcal/100g)
Water Hyacinth	11.47	18.71	13.4	1.2	8.82	55.22	285.28
Fish Offal	7.41	27.84	56.91	7.74	0.28	0.1	297.7
Noug Cake	3.6	8.1	32.3	14.7	23.9	41.3	426.7
Wheat Bran	5.2	4.3	15.6	4	13.6	70.9	382

*N.b. CP is crude protein, CF is crude fat*

### ***Growth response of C. carpio fingerlings under different treatment diets***

The mean value of fish weight and length after 90 experimental days were given in Table 3. The growth performances of common carp fed by diets consisting different inclusion levels of fermented water hyacinth (25%, 50%, 75% and control levels replacing wheat bran as energy source) were not statistically ( $p>0.05$ ) different.

However, there were slight mean variations in all parameters across the treatment groups. Accordingly, the highest growth was observed in a control group (T4) with final mean values; weight of  $23.38\pm 2.36$  g, total length of  $11.68\pm 1.08$  cm and fork length of  $10.27\pm 0.91$  cm followed by T1 (25% WH) where they attained values as  $22.81\pm 2.43$  g,  $11.42\pm 0.94$  cm and a fork length of  $10.21\pm 1.22$  cm. T2 (50% WH) attained a final mean weight of  $22.37\pm 2.7$  g, a final mean of  $11.47\pm 1.06$  cm and a fork length of  $10.51\pm 0.97$  cm. The least growth performance was observed in T3 (with the highest 75% inclusion rate of WH). The final mean weight of T3 was  $20.94\pm 2.06$  g, final mean total length of  $11.38\pm 1.16$  cm and a final fork length of  $9.93\pm 1.00$  cm. With increasing levels of water hyacinth in fish feed, the growth of fish decreases (Mohaptra, 2015; Zaman *et al.*, 2017; Derribew Hailu *et al.*, 2020). This may be due to increased anti nutritional factor as level of water hyacinth increase across the treatments.

Specific growth rate (SGR) and weight gain (g) was almost similar in all treatments except T3 in which the values recorded below other treatments (0.155 and 7.51 g) respectively (Table 3). In addition, there was no significance difference ( $p>0.05$ ) in survival rate and Fulton's condition factor across the treatments and the control group also.

Table 3: Growth performance and survival of *C. carpio* fed diet with different proportions of WH

Growth parameters	T1	T2	T3	Control	P-value
Initial Total length(cm)	9.75±1.04	9.59±1.25	9.57±0.7	9.56±0.78	-
Initial fork length (cm)	8.58±0.89	8.54±1.03	8.57±0.74	8.58±0.7	-
Initial weight(g)	13.59±2.29	13.15±2.4	13.43±2.5	13.57±2.6	-
Final Total length(cm)	11.42±0.94	11.47±1.06	11.38±1.16 <sup>a</sup>	11.68±1.08	0.821
Final fork length(cm)	10.21±1.22	10.51±0.97	9.93±1.00	10.27±0.91	0.676
Final weight(g)	22.81±±2.43 <sup>a</sup>	22.37±2.7	20.94±2.06	23.38±2.36 <sup>a</sup>	0.603
SGR (% d <sup>-1</sup> ) (g)	0.227	0.245	0.155	0.266	-
Condition Factor (FCF)	1.45± ±0.14	1.42±0.18	1.41±0.16	1.50±0.14	0.215
Weight gain(g)	9.22	9.22	7.51	9.81	-
Survival rate (%)	83.33	80.77	82.05	84.61	-

Note: T1, T2, and T3 stands for treatment number, g=gram, cm=centimeter; SGR= Specific growth rate; FCF=Fulton condition factor.

Growth of fish in different inclusion level of water hyacinth is shown in Figure 1 A and B below.

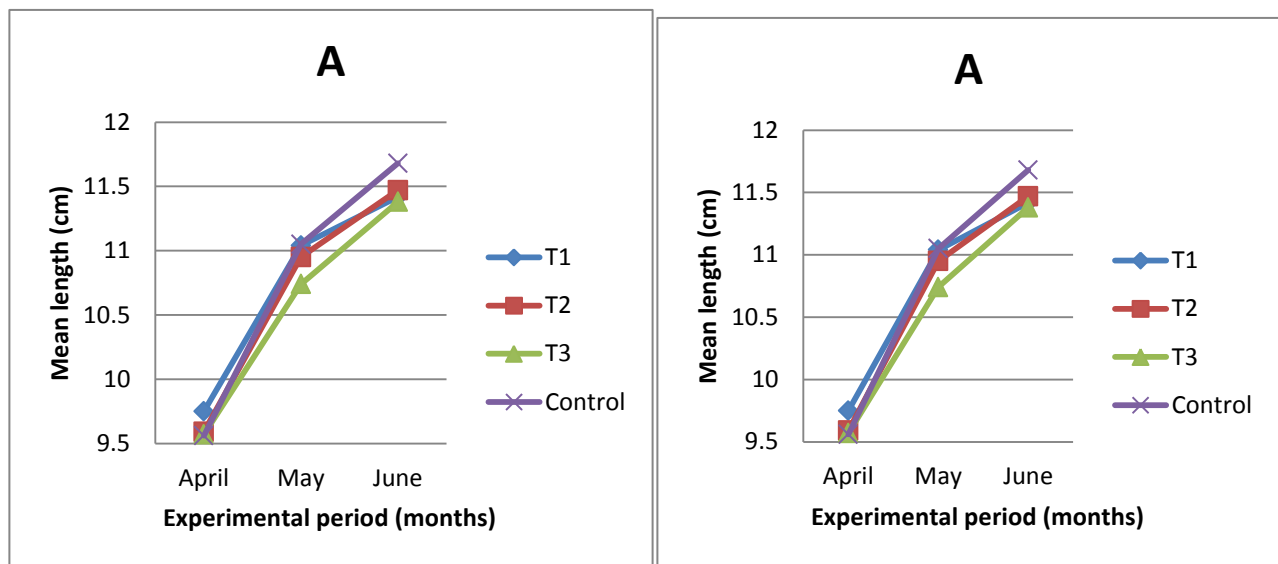


Figure 1: Growth trend of *C. carpio* in length (A) and weight (B) under different inclusion level of *Eichhornia crassipes*

#### Cost analysis of using water hyacinth in place of wheat bran in fish feed

The cost analysis of feed items in all treatments was estimated as shown in Table 4. Labor cost of processing fishmeal was relatively equal in all treatments while processing of water hyacinth differ the cost among T1, T2 and T3 as indicated in the table 4 below. The indicated cost was only the cost of items up to three month data of common carp fish growth. Hence, the value obtained from fish was not evaluated since the fish was not sold. Even though differences in fish growth were not statistically significant in all treatments, it is possible to summarize that, as water hyacinth level increase in treatments, the labor cost increases and growth of fish decreases as compared to the control group.



Table 4: Cost analysis of fish feed used in different treatments.

Treatments	Feed items	% age	Amount used in Kg	Price in birr/Kg	Total Price(ETB)	Labor cost(ETB)	Price of all ingredient (ETB)
T1	Fish meal	20.30	3.05	20	61	85	604.82
	NC	47.4	7.11	35	248.85	-	
	WH	7.46	1.12	-	-	28	
	WB	22.4	3.36	14.5	48.72	-	
	Molasses	20%of WH	0.22	15	3.3	-	
	Salt	1.5	0.22	5	5	-	
	Minerals	1	0.15	833	124.95	-	
T2	Fish meal	20.67	3.1	20	62	90	626.37
	NC	48.20	7.23	35	253.05	-	
	WH	14.33	2.15	-	-	53.75	
	WB	14.33	2.15	14.5	31.17	-	
	Molasses	20%of WH	0.43	15	6.45	-	
	Salt	1.5	0.22		5	-	
	Minerals	1	0.15	833	124.95	-	
T3	Fish meal	20.97	3.14	20	62.8	95	646.92
	NC	48.93	7.34	35	256.9	-	
	WH	20.71	3.11	-	-	77.75	
	Molasses	20%of WH	0.62	15	9.3	-	
	WB	7	1.05	14.5	15.22	-	
	Salt	1.5	0.22	20	5	-	
	Minerals	1	0.15	833	124.95	-	
Control (T4)	Fish meal	20.0	3	20	60	85	589.95
	NC	48.4	7.26	35	254.1		
	WH	0	-	-	-		
	Molasses	0	-	-	-		
	WB	28.0	4.2	14.5	60.9		
	Salt(g)	1.5	0.22	20	5		
	Minerals(g)	1	0.15	833	124.95		

### Conclusion and Recommendations

Based on the analysis of proximate composition, the water hyacinth collected from Hara Denbel/Lake Ziway in September 2020 was poor in protein content (below 18 % CP content) but good in carbohydrate content to be used as energy source.

Incorporation of molasses fermented *Eichhornia crassipes* in *C. carpio* feed is possible up to 75% as energy source without causing significant difference in all growth parameters. The nutrient composition of *Eichhornia crassipes* from Lake Ziway/Hara Denbel should be assessed for seasonal variation.

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# Preliminary Study of Fish Parasite in Lake Belbela

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## Abstract

*A preliminary study of fish parasite in Lake Belbela was carried out from September 2020 to June 2021 by collecting fish species using gillnets of various mesh sizes. A total of 116 fish samples were collected from randomly selected site of the lake. The samples included 74 (63.79%) Nile tilapia (*Oreochromis niloticus*), 11(9.48%) *Cyprinus carpio* and 31(26.72%) *Labeobarbus intermidus*. The fish were examined thoroughly for internal and external parasites. Out of 116 individuals of fishes examined during the study period, 26 (22.41%) were infected with parasites of which 6(23.07%) were *Labeobarbus intermidus*, 5(19.23%) were *Cyprinus carpio* and 15(57.69%) were *Oreochromis niloticus*. The major parasites identified during the investigation include adult *Contraecaecum* (Nematoda), *Eustrongylides* (Nematoda), and metacercariae of *Clinostomum* spp. (Digenea). Among the parasites recorded in the present study, the *Clinostomatid* digeneans and *Contraecaecum* nematodes could be medically important from public health point of view since the parasites can be transmitted to humans by eating raw or smoked fish and hence, fish products should not be consumed raw or smoked .*

**Key words:** Fish parasites, Lake Belbela, Prevalence

## Introduction

The fish sector makes a vital contribution to the food and nutritional security of 200 million Africans and it provides income for over 10 million people engaged in fish production, processing and trade. Moreover, fish has become a leading export commodity for Africa, with an annual export value of US\$ 2.7 billion (FAO, 2003). Africa hosts a great diversity of freshwater fish of which more than 3,000 species have been identified. Yet the benefits are at risk as the exploitation of natural fish stocks is reaching its limit (Skelton, 2001).

Freshwater fish can serve as definitive, intermediate or paratenic (transport) hosts in the life cycles of many species of protozoan, helminthes and crustacean parasites. The parasites usually exist in equilibrium with their hosts as a survival strategy (Bush *et. al.*, 2001). However, in instances where the hosts are overcrowded, such as in fish farms, parasitic diseases can spread very rapidly and cause gross mortalities (Paperna, 1996), losses in productivity in different water bodies and also human diseases in many areas of the world (Paperna, 1980; Balarin, 1986; Lester, 1988; Roberts, 1989 all cited in Eshetu Yimer and Mulualem Eneyew, 2003). They can also spoil the appearance of fish and usually affect the marketability of commercially produced fish, thus raising public health concerns especially in areas where raw fish is eaten (Paperna, 1996). In natural systems, they may threaten the abundance and diversity of indigenous fish species. The relative abundance of endo- and ectoparasites of fish in a particular aquatic system can also be used as an indicator of environmental stress. Ectoparasites, for instance, are more in contact with water; if they are sensitive to a pollutant, there will be less ectoparasite than endoparasites in a polluted system (Avenant-Oldewage, 1991).

One of the main emphases in Ethiopia is to develop capture fisheries and aquaculture to their full potential making a big contribution to national food availability, food security, economic growth, and trade and improved living standards. Few studies were done on fish parasites of natural water bodies in Ethiopia. The habit of raw fish eating is common among fishermen and people in Ethiopia, especially people near to water bodies. A pilot survey conducted on fishery of Lake Belbela indicated that the lake contribute fish to the local community and distant markets. But potential fish parasites that can easily be transmitted to fish consumers are not sufficiently known. The objective of the current study was to assess the prevalence of common economically important parasite of fish in Lake Belbela.

## **Materials and Methods**

### ***Collection of fish species and Parasites:***

Samples of fishes were collected by using different mesh sizes of gillnets from the selected sites of the lake. The gears were set in the afternoon and lifted in the following morning. In addition, fish caught by fishermen were included to the samples in order to increase sample size and to supplement the data on certain aspects of parasite infestation of the fish. Immediately after capture, total length (TL) and total weight (TW) of each specimen were measured to the nearest 0.1 cm and 0.1 g, respectively. Each specimen was then dissected and its sex determined by inspecting the gonads. Maturity stages were rated visually and recorded. Five-point maturity scales were used for this purpose (Holden & Raitt, 1974). The procedure of Paperna (1980) was followed to collect the parasites of the fishes. The parasites that were collected from each fish samples were kept in a plastic bag containing 4% formaldehyde solution. Samples was then be transported to center for further laboratory studies.

### ***Fixation, preservation and identification of parasites:***

The technique, method appearance and procedures of Paperna (1980) and Bykhovskaya-Pavlovskaya (1964) were used as a guideline in fixing, preserving and identification of each parasite specimens. Larva nematodes were fixed in 4 % formalin and later stored in the saline solution. Encapsulated larvae were carefully dissected before the tissue was fixed. Preserved larvae was cleared in lactophenol and then observed under low power magnification. Adult nematodes were fixed in hot formalin to insure their relaxation and preserved in 4% formalin mixed in 1% glycerine to avoid accidental drying.

Parasite Cestodes were fixed in AFA (Alcohol Formalin Acetic acid). For microscopic study, Nematodes were cleared in lactophenol for 24 hours and examined under lower magnification microscopy. In the case of Cestodes, diagnoses were made after being carmine stained and cleared in absolute alcohol followed by 70% alcohol.

### ***Data management and Analysis:***

The data collected during the study period were entered into Microsoft Excel Sheet computer program and were analyzed using descriptive statistics and mean comparison procedure of the Statistical Package for Social Science (SPSS V. 21.0).

## Results and Discussion

### *Occurrence of parasites in sampled fishes:*

Out of 116 individuals of fishes examined during the study period, 26 (22.41%) were infected with parasites; of which 6(23.07%) were *Labeobarbus intermidus*, 5(19.23%) were common carp and 15(57.69%) were *O. niloticus*. The common identified parasites were recorded as, *Contracaecum*, *Clinostomum* and *Eustrongylides*.

### *Prevalence in host sex:*

Sexes of the host fish were assessed to observe their influence on the parasite infection. When analyzing the infection rate of all examined parasites larvae by host sex, out of the fifty eight males examined, 9(15.51%) were infected (Table 1). On the other hand, out of 58 females examined, 17(29.31%) were infected.

Table 1: Prevalence of parasites on sex base of the host (n: 116)

Sex	number of examined	number of infected	%age ( infected)
Male	58 (50%)	9	15.51
Female	58 (50%)	17	29.31

### *Total prevalence of parasite species:*

The prevalence of each parasite in the fish samples showed different rates during the study period. Among the total (n: 116) fish examined, 26 (22.41% of the total fish samples) were positive to the parasites infection; 22(18.96% of the total and 84.61% of the infected) were infected by *Contracaecum* larvae while 17 (14.65% of the total and 65.38% of the infected), and 13 (11.20% of the total and 50% of the infected) were infected by *Clinostomum* spp and *Eustrongylides* respectively (Table 2). The results indicated higher prevalence of *Contracaecum* followed by *Clinostomum* spp and *Eustrongylides* parasites during study period in Lake Belbela.

Table 2: Prevalence of fish parasites recorded among examined fishes.

Sp of parasite	number observed	<i>O. niloticus</i>	<i>L. intermidus</i>	<i>C. carpio</i>
<b>Contracaecum</b>	22	12 (54.5%)	6(27.7%)	4(18.2%)
<b>Clinostomum</b>	17	15(88.2%)	0(0.0%)	2(11.8%)
<b>Eustrongylides</b>	13	10(76.9%)	1(7.7%)	2(15.38%)

The most prevalent adult nematodes were *Contracaecum* spp recovered from pericardial cavity of Nile tilapia and *Labeobarbus intermidus*. Prevalence of *Contracaecum* spp in Nile tilapia from Lake Belbela was 16.2% which is again lower than the works of Eshetu Yimer and Mulualem Eneyew (2003) in Lake Tana (59.8%) and far higher than the findings of Eshetu Yimer *et al.* (1999) which was 2.09% from Lake Chamo. The current study also indicated higher prevalence of parasite infections as compared to the findings of Amare Tadesse (1986) in which the prevalence of *Contracaecum* spp. in Nile tilapia from Lakes Awassa and Chamo was 10.6%. Identification of these larval nematodes to species level is difficult

unless it is supported by DNA sequencing technique linked with their adult identification. Encapsulated larval nematodes are known to cause fibrous capsule (Paperna, 1980) and the non-encapsulated larvae cause extensive tissue damage by migration.

Apart from this, the larval stages of *Contracaecum multipapillatum* were reported as potentially zoonotic parasite in Mexico (Vidal-Martinez *et al.*, 1994). The study revealed that another parasite species, the nematode *Eustrongylides spp.* was also prevalent nematode while *Eustrongylides ignotus* was also reported to be infectious to humans (Guerin *et al.*, 1982; Eberhard *et al.*, 1989 and Wittner *et al.*, 1989 all cited in Barros *et al.*, 2004) and the presence of the genus *Eustrongylides* in mesentery of African catfish from Lake Chamo was indicated by the work of Eshetu Yimer *et al.* (1999).

The findings of Eshetu Yimer and Muluaem Eneyew (2003) on Lake Tana showed the most prevalent digenean parasites of Nile tilapia in the lakes were *Clinostomum spp.* which is totally in agreement with the current finding which shows 14.65% digenean parasites on Nile tilapia.

### **Conclusion and Recommendations**

Apart from economic and public health importance, parasites impair fisheries activity. At the lakes, the harvested fish, fishing equipment and fishermen are loaded on too narrow boat. Therefore, parasites that detach from the fish host bite the bare foot of fishermen causing pain, bleeding and breakage of skin that might allow the entrance of other organisms which may cause anxiety and fear among young fishermen employed in the job. The present study shows, the proportion of parasites differ in prevalence. Thus based on this investigation public awareness should be created on the effect of parasites infestation.

Clinostomatid digeneans and *Contracaecum* nematodes could be potential human health risks of eating uncooked or slightly cooked/smoked fish. So, based on their risk to human being,

Medical survey on occurrence of laryngo-pharyngitis should be done on people eating uncooked/ smoked fish,

Identification of the genera *Clinostomum* and *Contracaecum* to species level is important to check for the occurrence of zoonotic parasites

Focus should be given for capacity building in fish parasitology and pathology by establishing network with international institutes experienced in the field

Consumers should not eat uncooked or slightly cooked fish for which health education should be given for them on the risk of eating raw and partly cooked fish.

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# Analysis of Nutrient Content in Common Carp (*Cyprinus carpio*) and Crucian Carp (*Carassius carassius*) Fish Offal

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## Abstract

*Fishmeal is a product prepared from fishery bycatch and fish by-product and is a nutrient-rich feed ingredient commonly used as in poultry and other domestic animals diet. The current experiment was conducted at Batu Fish and Other Aquatic Life Research Center to analyze the nutrient contents of offal of two commercially important fish species, common carp (*Cyprinus carpio*) and crucian carp (*Carassius carassius*) during July 2020 to June 2021. Offal collected from processing shade of Batu Fishermen cooperative were used to prepare the fishmeal. Offal from both species were processed to fishmeal in two different methods. One group was dried without cooking the offal. The remaining group was cooked, pressed, dried, and ground into fine powder. The fishmeal were then analyzed for proximate composition. The fishmeal prepared from cooked offal of common carp had significantly ( $p<0.05$ ) lower moisture ( $6.66 \pm 0.11\%$ ) potassium, manganese and iron contents; but higher zinc content than that of the uncooked one.. Similarly the fishmeal prepared from cooked offal of crucian carp had significantly ( $p<0.05$ ) lower moisture ( $5.97\pm 0.48\%$ ) and zinc contents -but had significantly ( $p<0.05$ ) higher manganese and iron contents as compared to the uncooked one. Comparing the proximate composition of the two fish species, the fishmeal prepared from cooked common carp offal had significantly ( $p<0.05$ ) higher moisture content, crude protein, potassium, manganese and zinc contents but significantly ( $p>0.05$ ) lower sodium and iron contents as compared to that of the fishmeal prepared from cooked crucian carp. The uncooked common carp fishmeal had significantly ( $p<0.05$ ) higher potassium, manganese and iron contents but significantly ( $p>0.05$ ) lower crude ash content than the fishmeal of cooked Crucian carp offal. It was concluded that fishmeal prepared from cooked offal of common carp has better protein content than that of the fishmeal prepared from crucian carp; and hence the former one is preferred to be used as ingredient in animal feed as a good protein source.*

**Keywords:** Common carp, crucian carp, fish offal, nutrient content

## Introduction

Fishmeal is a generic term produced from a nutrient-rich feed ingredient commonly used for poultry and other domestic animals diet for many decades. It can be made from almost any type of seafood but is generally manufactured from wild-caught, small marine fish that contain a high percentage of bones and oil which are not suitable for direct human consumption. A small percentage of fish meal is rendered from the by-catch of other fisheries, and by-products or trimmings created during processing of various seafood products destined for direct human consumption (Miles and Chapman, 2006). Fish meal preparation and utilization began during the early 1800s in northern Europe and North America based primarily on surplus herring catches. The early industry was geared towards the production of fish meal for high nitrogen and phosphorus fertilizer. Fish meal has become a fertilizer and the high protein content makes it very suitable as an animal feedstuff (Clucas, 1982). It has high levels of essential amino acids such as

methionine and lysine, and it also has a good balance of unsaturated fatty acids, certain minerals (available phosphorus), and vitamins (A, D, and B-complex). Fresh fish offal processed under local condition using locally available materials and dried on rack are appropriate and good in feed quality for livestock.

The composition of fishmeal varies according to the species of fish, method of processing, and whether fillets have been removed for separate markets. Anchovies and menhaden are processed whole, with resulting meals containing 64 and 61% protein, respectively. Marine based ingredients, especially fishmeals, are highly sought after as the protein source of choice for many formulated diets (Allan, 2000). That is because fishmeal provide feeds with high contents of essential amino and fatty acids, and low content in carbohydrates; thus being usually well digested and mainly used by feeds industry as a rich source of protein (Allan, 2000; Hardy, 2006). Fish meal (FM) carries large quantities of energy per unit weight and is a source of high-quality protein and highly digestible essential amino and fatty acids (Zinn *et al.*, 2009). The nutritive value of fish meal varies depending on sources of input, place of harvest and addition of salt for preservation. In Ethiopia, the chemical compositions of tilapia and African catfish were studied by different scholars. However, there is no any information on chemical composition of the fishmeal prepared from common carp (*Cyprinus carpio*) and crucian carp (*Carassius carassius*) fish offal. Though there is limitation of information on chemical composition of these two species of fish, the production and leftover of common carp and crucian carp increases from time to time in different water bodies of Ethiopia including Lake Denbel/Ziway. So, conducting a research on chemical profile of common carp and crucian carp is unquestionable to utilize as fish feed for future times. Understanding the chemical composition of various fishmeals used in animal or aqua feed are essential for formulating artificial diets (Kinh *et al.*, 2011). The aim of this study was to determine the chemical composition of fishmeal prepared from offal of common carp and crucian carp fish species through cooking and uncooked processing.

## **Materials and Methods**

### ***Sample preparation***

The raw materials used for preparation of fishmeal were common carp and crucian carp offal collected from fish processing shade of Ziway-Batu fishermen cooperative. The offal consisted of parts of discarded fish which include: fish bone, head, skin, internal viscera, the digestive tract, liver, pancreas, spleen, gonads and air sac, parts that are not usually used for human consumption. Offal from both species was categorized into two groups. One group was dried without cooking. The remaining group was cooked according to the procedure described by Abera Degebassa *et al.* (2008). *It involves* cooking, pressing, drying, and grinding of fish or fish waste into a solid.

**Cooking:** The purpose of the heating process is to liberate the oil from the fat depots of the fish, and to condition the material for the subsequent treatment. The fresh fish offal was cooked-until the flesh opaque and separates to about 95 °C -100 °C for about 30 minutes (FAO,1986). In this step, oil from fat deposits and moisture content librated and un-wanted materials was sterilized.

The cooked offal was left to settle in the barrel over night. That is from 6 pm to 8 am (morning) to separate the oil, water and solid components.

**Pressing:** The purpose of the press is to squeeze out as much liquid as possible from the solid phase. This is important not only to improve the oil yield and the quality of the meal, but also to reduce the moisture content of the press cake as far as possible, thereby reducing the fuel consumption of the dryers and increasing their capacity. The pressed by-products were spread on the laminated tin on rack for drying in the sun.

**Drying:** The purpose of the drying process is to convert the wet and unstable mixture of press cake, decanter sludge and concentration into a dry and stable fish meal. In practice, this means drying to a moisture content below 12%, which generally may be considered low enough to check microbial activity. This drying is done by heating the material to a temperature where the rate of evaporation of the water is considered satisfactory.

**Grinding, packaging and Storing:** The dried meal both from (cooked and uncooked) are ground using Laboratory miller to appropriate size. The ground powder was packaged using polyethylene bags stored in dessicator until analyzed.

#### ***Estimation of total carbohydrate***

The percentage of carbohydrate was calculated using the formula:  $100 - (\text{percentage of ash} + \text{percentage of moisture} + \text{percentage of fat} + \text{percentage of protein})$  (Indrayan *et al.*, 2005).

#### ***Estimation of nutritive value /Energy/***

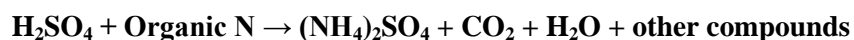
Nutritive values of the samples were determined by multiplying the values obtained for protein, fat and carbohydrate by 4.00, 9.00 and 4.00, respectively and adding up the values (Indrayan, *et al.*, 2005).

#### ***Determination of proximate composition***

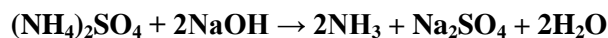
Moisture percentage was determined by weighing every Petri dish and the wet sample. Consequently sample was dried in an oven at 105 °C for 3 hours. Then Petri dish along with dried sample was placed in desiccators for 30 minutes. Eventually, the weight of Petri dish and dried sample was weight (AOAC, 2005). The moisture percentage was calculated using the following formula:

$$\text{Moisture \%} = 100 * ((\text{wet sample weight} - \text{dry sample weight})/\text{wet sample weight})$$

Crud protein percentage was determined through the Kjeldahl method according to (AOAC, 2005). VELP DK Series Digestions Unit and a VELP UDK 159 Automatic Digestion were used subsequently. Every Kjeldahl flask was prepared to add 2 catalysts for digestion. This included 1 g of the dried sample, 0.2 g CuSO<sub>4</sub> and 7 g of K<sub>2</sub>SO<sub>4</sub>. For the actual digestion, 12 ml of H<sub>2</sub>SO<sub>4</sub> was added also. Digestion was completed at 420°C for 1 hour. Eventually the sample was cooled down to 50°C. The sulfuric acid converts all nitrogen in organic form into inorganic form that is stable and suitable for analysis:



The nitrogen was separated from the digested mixture by steam distilling with the UDK 159 in order to extract ammonia from the alkaline solution. Sodium hydroxide (40 %) was added to raise the pH and convert solid  $\text{NH}_4^+$  into gaseous  $\text{NH}_3$ . The distilled nitrogen was then trapped by adding boric acid.



For the final colorimetric titration, hydrochloric acid was added in order to react with the ammonia and the indicators. The volume of titrant that was needed to reach the end point, allows to automatically calculating the amount of nitrogen, expressed as % N.

Crude fat was determined using Soxhlet fat extractor, consisting of a rounded-bottom flask, an extraction chamber and a condenser. For 200 ml extraction chambers, 50 ml of diethyl ether was used as extraction solvent. Approximately 2 grams of dried sample in a filter was placed into the extraction chamber to conduct the fat extraction. The rounded-bottom flask was weighed before each sample in case some fat remained from the previous sample. The sample was subjected to 3 hours of extraction. Afterwards the rounded-bottom flasks containing the fat was placed in the oven to eliminate any remaining petroleum diethyl ether and was weighed again (AOAC, 2005). The fat percentage will be calculated using the following formula:

$$\text{Fat \%} = 100 * ((\text{weight rounded-bottom flask \& fat} - \text{weight rounded-bottom flask}) / \text{dry sample weight}).$$

To determine crude ash, the sample was ashed in order to be able to analyse the mineral content afterwards. Two grams of the dried samples was placed in crucible cups that were weighed beforehand. Prior to placing the samples in the oven, they were charred on hot plates for 30 minutes to prevent excess smoke production. The ashing itself will be done by placing the crucible cups in a 550 °C oven for 8 hours (AOAC, 2005). The following formula was used to calculate the ash percentage:

$$\text{Ash \%} = 100 * (\text{weight ash} / \text{weight dry sample})$$

### *Data analysis*

Data of proximate composition (moisture, protein, fat and ash) of cooked and uncooked common carp and crucian carp was analyzed using t-test. Mean values were separated at 0.05 level of significance where significant differences were detected.

### **Results and discussion**

The proximate composition and minerals content of common carp both cooked and uncooked is presented in Table 1.

Table 1. The proximate composition and minerals contents of fishmeal prepared from cooked and uncooked common carp offal.

Parameters	Cooked	Uncooked	p-value
Moisture content	6.65±0.11	7.50±0.33	0.039
Crude Protein	64.23±1.92	63.41±2.36	0.862
Crude Fat	6.72±0.624	9.1158±0.5061	0.125
Crude Ash	25.94±0.16	21.45±0.13	0.00
Gross energy (Kcal/ 100g)	317.36	335.69	
Sodium	2518.21±10.25	2472.34±4.077	0.074
Potassium	8579.66±44.36	8965.26±35.12	0.001
Manganese	54.31±0.39	83.11±0.9063	0.000
Iron	665.89±19.18	1707.72±40.47	0.003
Zinc	531.76±1.80	369.74±1.461	0.000
Magnesium	2570.35±97.96	2286.87±50.08	0.143

There was statistically significant difference in moisture content, crude ash, potassium, manganese, iron and zinc contents between the fishmeal prepared from cooked and uncooked common carp offal (Table 1). The fishmeal prepared from cooked common carp offal had significantly ( $p < 0.05$ ) lower moisture content ( $6.6558 \pm 0.1133$ ) as compared to uncooked one ( $7.5028 \pm 0.3291$ ). Similarly the potassium, manganese and iron contents in cooked fishmeal were significantly ( $p < 0.05$ ) lower than the content in fishmeal prepared from uncooked offal. Protein, fat, sodium and magnesium contents were not statistically different ( $p > 0.05$ ) between the fishmeal prepared from cooked and uncooked offal while zinc content was statistically higher in cooked one.

In the present study, the moisture content is far below 15 %. that fishmeal could have shelf life over one year (Poulter *et al.*, 1982) . Cooking increases the release of liquor, hence, the moisture content of cooked fishmeal was lower than that of uncooked fishmeal. This corroborates the reason why cooked offal has lower moisture content as compared to sun dried offal (Ward *et al.*, 1977). Cooking has no effect on the concentration of Grouper (*Epinephelus morio*), red snapper (*LuGanus campechanus*), Florida pompano (*Trachinotus carolinus*) and Spanish mackerel (*Scomberomorus macuhtus*) of microelement, zinc, copper, iron and manganese (Gall *et al.*, 1983; Marimuthu *et al.*, 2012).

Crude ash, crude protein and crude fat contents of cooked fish increased due to rise in dry matter contents (Uran and Gokoglu, 2014). Accordingly, the increase in ash, protein and fat content found in cooked silver catfish fillets is explained by the reduction in moisture. While cooking methods affected mineral content of anchovy, cooking temperature did not affect. Cooking methods reduced moisture and increased the protein content in silver catfish (*Rhamdia quelen*) (Weber *et al.*, 2008). Na, Mg, and Zn contents of cooked fish fillets significantly decreased in Red mullet (*Mullus barbatus*) (Koubaa *et al.*, 2012). In comparison to raw fish fillets, when grass carp (*Ctenopharyngodon idella*) was cooked there was an increase in protein, lipid and ash contents. Na, K, Mg, P and Zn contents of boiled fish fillets significantly decreased (Golgolipour *et al.*, 2019). Fish cooking, considering that Mg, P, Zn and Mn contents decreased in almost all cooking methods of rainbow trout (*Oncorhynchus mykiss*) (Gokoglu *et al.*, 2004). The protein and ash contents increased in all cooked fish. The moisture content of cooked fish decreased. Mineral levels were affected by cooking methods in African catfish (Ersoy and Özeren, 2009).

Table 2. The proximate composition and minerals contents of cooked and uncooked crucian carp

Parameters	Cooked	Uncooked	P-value
Moisture content	5.97±0.48	7.03±0.88	0.009
Crude Protein	58.88±0.67	57.43±6.35	0.826
Crude Fat	7.36±0.719	10.03±0.71	0.150
Crude Ash	25.92±0.51	26.26±0.76	0.168
Gross energy (Kcal/ 100g)	301.74	320.04	
Sodium	2705.20±8.19	2561.60±28.17	0.05
Potassium	8071.77±60.05	8466.50±157.13	0.168
Manganese	42.56±1.87	53.31±0.42	0.04
Iron	1992.42±21.9	676.25±19.02	0.001
Zinc	405.84±4.64	550.92±10.22	0.01
Magnesium	2316.86±46.62	2797.22±76.42	0.044

The moisture content, sodium, manganese, iron zinc and magnesium contents were statistically significantly different between the fishmeal prepared from cooked offal of crucian carp and the uncooked offal meal (Table 2). Cooking or heating has no statistically significant ( $p>0.05$ ) effects on protein contents, fat, ash, Sodium and Potassium of fishmeal. The cooked crucian carp fishmeal had significantly ( $p<0.05$ ) lower moisture and zinc contents but higher manganese and iron contents as compared to uncooked one. .

In Table 1 and 2, the moisture content and fat content of fishmeal prepared from cooked offal are lower as compared to the uncooked ones both in common carp and crucian carp. The reason behind low moisture content and fat content in cooked fish offal is that heat ruptures fat deposits to release oil and pressing drained out water content. Cooking coagulate protein thereby liberating bound water and oil, separation by pressing yield press cake which contains 60-80% of the oil-free dry matter (protein, bones) and liquid phase (press liquor) which contains water, oil, dissolved and suspended protein, vitamins and minerals. Pressing of the cooked offal is done to separate the bulk of the liquid fraction (press liquor) from the solid parts (press cake). This can be attained by pressing tightly the lid and tilting barrel containing cooked offal, so that the liquid fractions drain out. The main purpose of pressing is to squeeze out as much liquid (water and oil) as possible from the solid phase. Pressing helps to accelerate the drying process.

The decrease in moisture content after heat treatments is caused by partial water loss through evaporation (Weber *et al.*, 2008). Decreased moisture content has been described as the most important change causing significant protein increase in cooked fish fillets. Ersoy and Ozeren (2009) pointed out that the increase in protein, fat, and ash contents in fish after different cooking methods could be explained by water reduction, indicating an inverse relationship between water content and other nutritional components.

Table 3. Proximate and Minerals contents of Cooked Common carp and Crucian carp

Parameters	Common carp	Crucian carp	P-value
Moisture Content	6.66±0.65	5.97±0.05	0.008
Crude Protein	64.23±1.92	58.88±0.67	0.05
Crude Fat	6.72±0.44	7.36±0.07	0.430
Crude Ash	25.94±0.16 <sup>a</sup>	25.92±0.30 <sup>a</sup>	0.934
Gross energy (Kcal/ 100g)	317.36	301.74	
Sodium	2518.21±10.26	2705.20±8.19	0.009
Potassium	8579.66±44.36	8071.77±60.06	0.034
Manganese	54.31±0.39	42.56±1.87	0.017
Iron	665.89±19.18	1992.42±21.9	0.001
Zinc	531.75±1.80	405.84±4.64	0.002
Magnesium	2570.35 ± 97.96	2316.86±46.61	0.205

Fish species has statistically significant effect on the moisture content, sodium, potassium, manganese, iron and zinc of fish (Table 3). Fish species has no statistically significant ( $p>0.05$ ) effects on protein, fat and ash. The cooked common carp fishmeal had significantly ( $p<0.05$ ) high moisture content, crude protein, potassium, manganese and zinc as compared to cooked crucian carp. However, cooked common carp fishmeal had significantly ( $p>0.05$ ) low Sodium and Iron as compared to cooked Crucian carp.

The result obtained revealed, the crude protein content of tilapia and catfish is about (50.8% and 45.6%), fat content (9.2 and 8.8%), ash (29.5% and 37.6 %), the moisture content (12.8% and 15.4%), respectively (Abera Degebassa *et al.*, 2008). In the present study, moisture content of Common carp and Crucian carp is  $6.6558\pm0.654$  and  $5.9731\pm0.048$ , protein content ( $64.23 \pm 1.924$  and  $58.8839 \pm 0.6717$ ) and fat is ( $6.715\pm0.4415$  and  $7.3563\pm0.0719$ ). Fishmeal contains typically 60 to 72 percent protein, 10 to 20 percent ash, 5 to 12 percent fat and has a high content of the fatty acids EPA and DHA; more commonly referred to as long chain omega-3s (Crexi *et al.*, 2009). The protein content of Common carp and Crucian carp is higher than Nile tilapia and African Catfish (Abera Degebassa *et al.*, 2008). Common carp has significantly ( $p<0.05$ ) higher moisture content as compared to Crucian carp. The protein content and fat content was statistically not affected ( $p>0.05$ ) by fish species. Even though there is no statistically significant difference in protein content between Common carp and Crucian carp, the protein content of Common carp is higher than Crucian carp. This may be attributed to differences in the feeding habits, age, sex, breeding conditions and seasonal variations. These variations are closely related to the processing methods of raw material, food shortage, or physiological factors such as spawning or migrations, influence the chemical composition, occurring a higher variation of the lipid fraction (Crexi *et al.*, 2009). Crucian carp has significantly higher ( $p<0.05$ ) ash content as compared to Common carp. Study conducted on the effects of cooking temperatures (55, 65, 75, 85, 95, and 100 °C) on sensory properties and protein hydrolysates were studied in crucian carp (*Carassius auratus*) soup. A cooking temperature of 85 °C was preferred for more excellent flavor and higher nutritional value of crucian carp (Zhang *et al.*, 2013).

In a study conducted to determine the effect of different cooking methods on the nutritional composition of salmon and Chilean jack mackerel fillets, it was observed that protein content in both salmon and Chilean jack mackerel significantly increased under the different cooking methods (Bastias *et al.*, 2017).

Table 4. Proximate and Mineral contents of Uncooked Common carp and Crucian carp

Parameters	Common carp	Crucian carp	P-value
Moisture Content	7.50±0.19	7.03±0.05	0.097
Crude Protein	63.41±2.36	57.43±6.35	0.517
Crude Fat	9.12±0.04	10.03±0.71	0.405
Crude Ash	21.45±0.13	26.26±0.44	0.004
Gross energy (Kcal/ 100g)	335.68	320.03	
Sodium	2472.34±4.08	2561.6±28.17	0.094
Potassium	8965.27±35.12	8466.50±157.14	0.060
Manganese	83.11±0.91	53.301±0.43	0.001
Iron	1707.72±40.47	676.25±19.017	0.001
Zinc	369.74±1.46	550.92±10.22	0.002
Magnesium	2286.87±50.08	2797.22±76.42	0.047

Fish species has statistically significant effect on crude ash, manganese, iron and zinc of fish (Table 4). Fish species has no statistically significant ( $p>0.05$ ) effects Moisture content, protein, fat, Sodium and Potassium of uncooked fish. The uncooked common carp fishmeal had significantly ( $p<0.05$ ) high Potassium, Manganese and Iron as compared to uncooked Crucian carp. However, uncooked common carp fishmeal had significantly ( $p>0.05$ ) low Crude ash as compared to cooked Crucian carp. Al-Zahiri, *et al.*, 2021 studied the effect of dried and cooked common carp offal to feed common carp during which he observed that the crude protein (62 %) content was similar with the present finding, however, crude fat (14 %) and crude ash (9 %) differs from present study.

### Conclusion and recommendations

From the present study, it was concluded that cooking has statistically significant effect on the moisture content, fat content and ash contents. Cooking or heating has no statistically significant effects on protein contents of fishmeal. Common carp has significantly higher moisture content as compared to Crucian carp, however, the protein content and fat content was statistically not affected by fish species.

It is recommended that process control is necessary for producing high quality fishmeal. Excess temperature for prolonged periods in the cooking, evaporating and drying should be avoided since fish protein is sensitive to excessive heat. When the flesh is cooked at high temperatures, the proteins on the surface of fish flesh coagulated. On the other hand, when the cooking temperature is lower, the denaturation of muscle protein in the fish flesh might not complete and the soluble protein hydrolysates had not been dissolved sufficiently. Cooked offal of common carp has better protein content than that of crucian carp, and hence it is preferred to be used as an animal feed..

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