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Dairy Animals Research Results

Effect of timing of Artificial insemination on fertility of cows and calf sex ratio in Arsi cows

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Abstract

An on-station study was conducted to investigate the effect of the interval between artificial insemination (AI) and the beginning of estrus on the fertility and sex ratio of offspring in Arsi cows. Standing to be mounted was considered the beginning of estrus, and artificial inseminations were made 6 h (Group 1), 12 h (Group 2), 18 h (Group 3) and 24 h (Group 4) after the onset of estrus in cows. Overall, 50 cows with parity between 2 and 4, body weight ranging from 250 to 300 kg were used for the purpose of this study. Ovulation was synchronized by administration of PGF₂α hormone. Pregnancy diagnosis was performed by rectal palpation at 90 to 120 days post AI. Pregnancy rates were 60%, 60%, 60% and 50% in Groups 1, 2, 3 and 4, respectively. The highest female ratio of offspring was obtained in Group 1 (6h) with significant difference (P=0.041). The highest ratio of male calf was recorded in group 4. The number of artificial inseminations per pregnancy was within the economical limits in all groups. In conclusion, the results of this study indicate that the interval between the onset of estrus and artificial insemination time alter the sex ratio of offspring in Arsi cows without affecting fertility parameters.

Key words: Artificial insemination time; fertility rate; calf sex ratio; Arsi cows

1. Introduction

Artificial insemination (AI) has been defined as a process by which sperm is collected from the male, processed, stored and artificially introduced into the female reproductive tract for the purpose of conception (Morrow, 1985; Webb, 2003). When AI was being developed and confirmed, there were several studies that were designed to determine the optimal time of AI in relation to estrus (Aschbacher *et al.*, 1956). The figures suggested that optimal pregnancy rate per AI would be achieved from mid-estrus until a few hours after the end of estrus. Since then, the recommended practice has been inseminating cows 12 h after the first observed estrus (a.m.- p.m. breeding). However, because of the variability of interval between the onset and the observation of estrus, it is difficult to define the ideal time of AI in relation to ovulation (Foote, 1979).

In Ethiopia, there is high demand for female crossbred calves by dairy producers from artificial insemination services (Bekele, 2005). However, due to different reasons the number of improved heifers is in the country is very low. The use of modified flow cytometry to separate the X and Y sperm populations (sorted sperm), and using the corresponding type to perform AI is a proven and reliable techniques for selecting the gender of offspring. However, it is still the most expensive method for commercial applications (France *et al.*, 1992; Seidel *et al.*, 1999). Sex ratio manipulation can reasonably improve the effectiveness of selection and genetic improvement programs, through the differential increment of males or females born after Artificial insemination (AI) (Seidel, 2003).

Several efforts have been made to alter the sex of calves by varying time of insemination in different breeds of cattle and environments (Pursley *et al.*, 1998; Martinez *et al.*, 2004). It has been recommended that early inseminations (i.e. far before ovulation) would result in more female calves whereas late inseminations (i.e. close to ovulation) would result in more male calves, due to different timing of capacitation and survival time of the X- and Y-chromosome bearing spermatozoa in the female reproductive tract (Martinez *et al.*, 2004). However, there is lack of information on suitable insemination time after onset of estrus to alter the ratio of sex in new born calves without affecting fertility of cows in Arsi cattle. Therefore, the objective of the present study was to evaluate the effect of various intervals of AI on the fertility of cows and sex ratio of calves in Arsi cows.

2. Materials and methods

2.1 Study area

The study was conducted at Adami Tulu Agricultural Research center located at 167km south of Addis Ababa and situated at latitude of 7° 9' N and 38° 07' E longitude in semi-arid middle rift valley of Ethiopia. The area is situated at 1500 meters above sea level and the soil type of the area is fine, sandy loam with sand, clay in the proportion of 34:48:18 respectively. The average annual rain fall is 760mm. The minimum and maximum temperature are 12.6 and 27°C, respectively (ATARC, 2003).

2.2. Experimental animals, feed and housing

Arsi cows reared on station were selected based on their age, parity and body condition. They were used as a dam line for the study. The semen of different sire of Holstein Frisian breed was used as a sire line. The study was conducted on 50 Arsi cows with parity between 2 and 5 and average body weight of 280 kg. The study was conducted over a period of 18 months (from January 2019 to June 2020). The cows were allowed to graze natural pasture and concentrates were given in the morning and afternoon according to production level. The animals were housed in free stalls with a concrete slatted floor.

2.3. Synchronization, estrus detection and artificial insemination

Experimental animals were checked for pregnancy through rectal palpation before injecting with hormone PGF₂α. Those which were negative to pregnancy test were injected with PGF₂α hormone for synchronization. Estrus was observed by experienced herders and observation was held early in the morning, mid-day, afternoon and mid-night. The onset of estrus was determined when the cows stood to be mounted. A standard semen handling and insemination procedure recommended by IAEA (2005) was used to inseminate animals. Artificial inseminations were carried out by artificial inseminators with standard methodology for cattle. Semen from Holstein Friesian bulls (progeny tested) were deposited in the body of the uterus. The cows were inseminated with frozen-thawed semen 6 h (Group 1), 12 h (Group 2), 18 h (Group 3) and 24 h (Group 4) after the detected onset of estrus. Finally, the cows were examined by rectal palpation for pregnancy between 90 and 120 days after the artificial inseminations. The sex of calves was identified after delivery (9months later).

2.4. Statistical analysis

Data were analyzed by using the Statistical Package for the Social Sciences (SPSS 20). Chi-squared tests were used to compare pregnancy rate, the sex ratio of offspring and the number of artificial inseminations per pregnancy. Statistical significance was taken when $P < 0.05$.

3. Results and discussion

Table 1 summarizes the results of sex ratio and fertility studies. The sex ratios (female/male) were 75%/25%, 66.7%/33.3%, 50%/50% and 80%/20% in Groups 1, 2, 3 and 4, respectively.

Table 1. The effect of artificial insemination timing on the sex ratio of offspring, pregnancy rates and artificial insemination per pregnancy of Arsi cows (n=50)

AI time (Groups)	Pregnancy rate (%)	Sex ratio (%) (female/male)	Number of AI per pregnancy
Group 1 (6 h)	60	75/25 ^a	2.02
Group 2 (12 h)	60	66.7/33.3 ^{ab}	1.33
Group 3 (18 h)	60	50/50 ^{ab}	1.5
Group 4 (24 h)	50	20/80 ^b	1.6
Overall	58	58.6/41.4	1.7
P value		0.041	

The present sex ratio study showed that there are significant differences ($P < 0.05$) among group 1 and others with the highest female ratio was obtained in Group 1. On the other hand, the highest male ratio was detected in Group 4. When insemination is carried out beyond 18 h from the onset of the estrus, the percentage of males increases significantly. There was no significant difference ($P > 0.05$) in pregnancy rate among treatment groups. This indicates that it is possible to manipulate the sex ratio of the offspring in Arsi cows with timing of AI without affecting the fertility of cows.

Researchers have reported many conflicting results about the effect of the timing of artificial insemination on the sex ratio of offspring in dairy cows (Rorie *et al.*, 1999; Martinez *et al.*, 2004). Similar to the present study Martinez *et al.* (2004) reported that the percentage of females in the offspring could be increased by the inseminations performed in the first 18 h after the onset of estrus. In contrast, Orkun *et al.*, 2005 reported that artificial inseminations performed at different times in the first half of the estrus period did not alter the sex ratio of offspring in dairy cows. The difference between the results of these studies could be due to the differences in the breed of animals, difference in estrus detection and the quality of semen used for inseminations.

This difference in sex ratio of calves regarding to the time of insemination relative to the estrus on set can be explained considering that there are many physiological differences between Y and X spermatozoa. Rohde *et al.* 34; found that Y chromosome bearing sperm progress more quickly through cervical mucus than those carrying an X chromosome bearing sperm. There is a process of sperm selection in the oviduct, in which spermatozoa interact with the oviductal epithelium, forming a reservoir at the uterotubal-isthmus junction and undergo capacitation (Rodriguez *et al.*, 2001; Hunter, 2001).

Martinez *et al.* (2004) reported that the high percentage of female calves when cows are inseminated within the first 18 h from the onset of estrus can be explained by the fact that Y chromosome bearing sperm in the isthmus would achieve capacitation earlier than X chromosome bearing sperm, release from the oviductal epithelium and reach the fertilization place long before the ovulation.

In the present study there was no significant difference in conception rate between different insemination times. Contrary to the present study, Dransfield *et al.* (1998) found the highest conception rate between 8 and 12 h after the onset of estrus. Although 15 h is within the optimal interval, the pregnancy rate differences between these results could be due to the estrus detection method differences. The mean artificial insemination per pregnancy detected in the present study is within the economical limits advised for healthy herds (Herman *et al.*, 1994).

The results of the present study indicate that the percentage of females in the offspring can be increased by performing the AI within the first 12 h from the onset of the estrus, whereas delaying the AI for 24 h significantly increases the percentage of males in Arsi cows.

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Handling, Utilization and Processing of Camel Milk in Borana Zone, Southern Oromia, Ethiopia

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Abstract

The study was conducted to assess traditional camel milk and camel milk products handling, preservation and processing of Borana area as well as evaluate processing methods and quality of traditional and laboratory prepared fermented camel milk (Chuuchoe). This study was carried out using survey and laboratory experiments. For the survey, 132 respondents were selected through purposive sampling technique and interviewed on various aspects of camel milk and camel milk products using a single-visit multiple-subject diagnostic survey. Survey result revealed that the majority of camel dairying of labor gender division was done by women. Result showed pastoralists and agro-pastoralists hygienic handling of camel milk and milk products was poor. Respondents reported that they preserved camel milk by washing and smoking milk vessels, keeping milk in a cold place and processing into other products. All most all respondents use camel milk mainly in its raw state for home consumption. Most of respondents in the study area traditionally process camel milk into other camel milk products mainly during surplus milk production. Major fermented sour camel milk produced by respondents locally named Chuuchoe. Laboratory experiments on physicochemical and microbial quality of raw camel milk and Chuuchoe were analyzed. Results specified values of camel milk pH, TA, protein, fat, lactose, SNF, ash and total solids were 6.62 ± 0.07 , 0.14 ± 0.02 , 3.15 ± 0.18 , 3.25 ± 0.27 , 4.62 ± 0.54 , 8.5 ± 0.55 , 0.72 ± 0.02 and 11.74 ± 0.56 , respectively. Average counts of total viable count bacteria, fecal coliforms and yeast and molds were 7.32 ± 0.12 , 7.12 ± 0.16 and $6.33\pm 0.22 \log_{10} \text{cfu mL}^{-1}$, respectively. In comparisons of home and laboratory made Chuuchoe, significant differences ($P < 0.01$) were observed in physicochemical properties and microbial quality. In conclusion, handling and processing of camel milk is poor. Thus, improvement of camel milk hygienic handling and processing should be launched.

Keywords: Camel milk, hygienic handling, traditional processing, utilization

1. Introduction

One-humped camel (*Camelus dromedarius*) plays an important role as a primary source of subsistence in the lowlands of Ethiopia. Camel lives in areas which are not suitable for crop production and where other livestock species hardly thrive. The majority of camel is found in the eastern and southern parts of Ethiopia. Pastoralists depend on camel production for their livelihood due to the outstanding performance of camel in the arid and semi-arid lowland areas of Ethiopia where animal feeds and water are limited. In these areas, camel is generally kept for milk production and produce milk for a longer period of time even during the dry season when milk from cattle is scarce. The major ethnic groups owning camels in Ethiopia are the Beja, Rashaida, Afar, Somali and Borana. Camel has a significant contributions to the

livelihood of the pastoralist society who have little alternative mode of production system, particularly for milk production (Bekele *et al.*, 2002; Workneh, 2002; Eyassu, 2007).

Camel milk plays a vital role in household food security, prevention of malnutrition and acts as a source of cash to camel keepers and traders. It is an essential constituent of human diet in many parts of the world as well as in the pastoral and agro-pastoral areas of Borana zone. It was observed that camel milk has essential nutrients found in bovine milk composition which has valuable nutritional properties; however, it contains a high amount of antibacterial substances, vitamin C, and nutrient than bovine milk (Ramet, 2001; Haddadin *et al.*, 2008; Barłowska *et al.*, 2011).

Pastoralists commonly claim that camel milk is difficult to process into products and is only suitable for drinking as fresh or sour milk. However, currently, the possibility of producing various products from camel milk including soft cheese, yoghurt and butter has been reported (Berhe *et al.*, 2017). Milk is an ideal medium for the growth of microbes and loses its quality within a short period of time if not preserved in some way. Poor handling and processing practices of milk usually result in undesirable products. Spontaneous nature of milk fermentation process common in arid areas can result in undesirable products that are sometimes even risky or dangerous for human health (Farah *et al.*, 2007). Traditional fermented camel milk has different names and processing methods in different countries. It is known as '*Dhanaan*' in eastern Ethiopia (Eyassu 2007), '*Ititu*' in Karayu area of Ethiopia (Kedija *et al.*, 2008), '*Suusac*' in Kenya, '*garris*' in Somalia (Farah *et al.*, 2007) and '*Shubat*' in Turkey (Brezovecki *et al.*, 2015).

Camel milk production is facing high post-harvest quality deterioration and milk is wasted due to spoilage and quantity losses during the rainy seasons when production is high (Odongo *et al.*, 2016). It is frequently reported that surplus of camel milk is produced in the Borana pastoral and agro-pastoral areas during the rainy season (Dejene, 2015). However, it was not studied in depth, especially regarding camel milk handling, preservation and processing into different milk products and shelf-life of camel milk and camel milk products as compared to milk of cow under pastoral condition of Borana zone.

Camel milk handling practices at producers, traders, collection points, preservation, processing, poor infrastructure, unreliable transportation are the major constraints. In Ethiopia, including Borana zone, not only at different dairy cooperatives but also in researches, camel milk post-harvest handling and processing have not been given much attention. Camel milk is one of the economically important livestock products to improve the socio-economic status of camel owners, traders and dairy processing cooperatives. Although, despite the important contribution of camel milk to pastoralists living in the lowlands of the Borana zone, little is known about the properties, preservation, processing, shelf-life and keeping quality of camel milk at the high ambient temperatures prevailing at the production area. In addition, identifying traditional methods and types of camel milk fermentation is a paramount for camel milk quality improvement and product development.

Currently, climate change is a worldwide phenomenon throughout the world as well as in Borana agro ecological areas (Dirriba *et al.*, 2020). Consequently, this climate change is causing livestock death specially cattle following recurrent drought. Because of this, Borana communities are diversifying their livelihood towards camel dairy production and the demand of camel milk and milk product is also

increasing (Dejene, 2015). Only camel milk production is not enough to improve the livelihood of the producers thus they convert camel milk into diversified milk products to minimize spoilage and postharvest losses, especially during surplus milk product. Therefore, study is needed to address the information gaps on handling, utilization and processing of camel milk. Besides, traditional methods and types of camel milk fermentation and perception of pastoral and agro-pastoral community toward camel milk processing are vital issues for further interventions to design and develop appropriate camel milk quality improvement and camel dairy product technologies with the following objectives.

Objectives

- ❖ To assess handling, urination and processing of camel milk in the study area
- ❖ To evaluate processing methods and quality of traditional and laboratory prepared fermented camel milk

2. Materials and Methods

2.1. Description of the study area

The study was conducted in Yabello, Gomole and Moyale districts of Borana zone, southern Oromia, Ethiopia. These districts are located on Addis-Moyale road. The Borana plateau is the portion of the Southern Ethiopia rangelands which its climate is generally semi-arid with annual rainfall range of 500 mm in the South and 700 mm in the North, the altitude ranges from 1000m in the South to 1500m in the Northwest, 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, annual mean daily temperature varies from 19 to 24°C. There are four major seasons identifies the Borana plateau. These include: (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).

2.2. Methods of conducting experiments and data collection

The study was conducted in Gomole, Yaballo and Moyale districts as well as at Yabello Pastoral and Dryland Agriculture Research Center in the laboratory. This study was conducted in two phases which had survey and laboratory parts.

2.2.1. Assessment of camel milk handling, utilization and processing.

Survey was conducted to assess traditional camel milk handling, preservation and processing under pastoralists and along milk supply in Borana Zone. Three well known districts in camel production potential from pastoral area were purposively selected. From each district, milk-supplies and two Pastoral Associations (PAs) were selected. The selection of PAs from each district of Yabello, Gomole and Moyale that had good camel production potential were selected again using purposive sampling technique. Households and milk-supplies at each district were selected based on accessibility of the village and willingness of the camel owners and milk-supplies to take part in the interviews. Information about consumption pattern, preference and importance of camel milk, traditional processing and preservation methods, types of fermented camel milk, types of plant used for processing, types of spoilage

and shelf life of and milk products were collected from respondents by means of semi-structured questionnaires.

In order to develop effective interventions, it is necessary to understand what and how pastoralists and milk-supply cooperatives carry out the kind of camel milk and milk products in a given or particular way. Thus, to make use of existing sources of information, both secondary and primary information were used in the study. To collect required information for this study, a combination of different techniques were applied. Secondary information was collected from zone and district offices of pastoral and agro-pastoral. Moreover, relevant literatures and documents were consulted to provide technical background and to develop a basic understanding of how camel milk and milk products handling and processing operated in the study areas.

Handling, preservation and processing of camel milk under pastoral and along milk-supplies in Borana zone were assessed by using a single-visit multiple-subject diagnostic survey (ILCA 1990). A total of 132 respondents including milk-supply were selected using purposive sampling technique.

2.2.2. Evaluation of traditional practices and laboratory evaluation of fermented camel milk

2.2.2.1. Evaluation of processing methods, composition and microbial quality of traditional and laboratory prepared fermented camel milk

Plant materials used for fermentation of camel milk and processing were identified. Traditionally, processed fermented camel milk samples were collected for analyzing from selected pastoralists. Fresh camel milk samples used to prepare fermented camel milk for evaluation as traditional methods in the laboratory were also collected from pastoralists.

Fermented (500 ml) and raw camel milk (250 ml) samples were taken aseptically from 10 camel milk producers of Gomole and Yabello districts and placed into separate sterile universal bottles. Then the samples were labeled and transported by placing in icebox to the laboratory of Yabello Pastoral and Dryland Agriculture Research Center. The composition and microbial quality of camel raw milk were analyzed immediately after collecting soon at the arrival at laboratory, whereas composition and microbial quality of fermented camel milk samples were analyzed within 24 h of collection and processing.

2.2.2.2. Physicochemical analysis of raw and fermented camel milk

Physicochemical properties (pH, fat, protein, lactose, total solids and ash) of raw camel milk were analyzed using Lacto-scan model (Lactoscan SA, 8900 Nova Zagora Bulgaria). On the other hand, physicochemical properties of fermented camel milk were analyzed following standard procedures of AOAC (1995). The pH was measured by a digital pH meter. Acidity (% lactic acid) was determined by titrating 9 ml of sample with 0.1N NAOH solution. Crude protein (%N x 6.38) was determined by the Kjeldahl method. Fat was analyzed by Gerber method. Total solids and ash were determined according to Richardson (1985). Solids-not-fat content was determined by subtracting the percentage fat from % total solids (O'Mahony, 1985). Lactose was analyzed using proximate analyzing methods.

2.2.2.3. Microbiological analysis of raw and fermented camel milk

Microbiological counts of milk samples and considered were total plate count, coliform count (CC) and yeast and mould count (YMC). For determination of these counts, peptone water was sterilized by autoclaving at 121⁰C for 15 minutes. Similarly, the total plate count agar used for determination of total viable organisms and potato dextrose agar used for the determination of yeast and mould count were sterilized by autoclaving at 121⁰C for 15 minutes, while the violet red bile agar used for determination of CC was sterilized by boiling (Richardson, 1985). The Medias used for all the three counts were prepared according to the guidelines given by the manufacturers. Each count was done in duplicate.

2.2.2.3.1. Total viable bacteria count

Total bacteria count (TBC) was made by adding 1ml of milk sample into sterile test tube having 9ml of peptone water. After thorough mixing, the sample was serially diluted up to 1:10⁻⁵ dilution level and duplicate samples (1ml) were pour plated using 15 - 20ml of molten standard plate count agar. The plated sample was allowed to solidify and then incubated at 32⁰C for 48 hours. Colony count was made using colony counter (Marth, 1978).

2.2.2.3.2. Coli form count

One ml of milk sample was added into sterile test tube having 9 ml of peptone water. After mixing, the sample was serially diluted up to 1: 10⁻³ dilution level and duplicate samples (1ml) were pour plated using 15 - 20ml of molten Violet Red Bile agar. After thorough mixing, the plated sample was allowed to solidify and then incubated at 30⁰C for 24 hours. Finally, colony count was made using colony counter (Abdel-Rahman *et al.*, 2009). Typical dark red colonies were considered as coli forms.

2.2.2.3.3. Yeast and mould count

One ml of milk sample was added into sterile test tube having 9 ml of peptone water. After mixing, the sample was serially diluted up to 1: 10⁻³ dilution level and duplicate samples (1ml) were pour plated using 15 - 20ml of molten potato dextrose agar. After thorough mixing, the plated sample was allowed to solidify and then incubated at 25⁰C for 5 days. Finally, colony count was made using colony counter (Marth, 1978).

2.2.2.3.4. Viable lactic acid bacteria count for fermented camel milk

The lactic acid bacteria count was conducted by the pour plate method, with an overlay, adding 1 mL of fermented camel milk diluted inoculums and adding 15-20 ml of MRS agar in the Petri dishes. After solidification of the medium, an overlay was added, seeking the creation of a 15% CO₂ atmosphere, followed by incubation at 30⁰C for 5 days. After the required incubation time, the count was conducted in Petri dishes that presented between 25 and 250 colonies (Tebaldi *et al.*, 2007).

2.2.2.3.5. Microbiological count of fermented camel milk

A Standard Plate Count Agar, MRS agars, Violet Red Bile Agar and Potato Dextrose Agar were used for total bacterial count, Lactic acid bacteria, coli form count and yeast and mould count, respectively. All counts were determined according to Richardson (1985) as the same as milk indicated above.

2.3. Statistical Analyses

The collected survey data through key informants' interview were analyzed using descriptive statistics by using SPSS version 20 software. Microbial count data were first transformed to logarithmic values (\log_{10}) before statistical analysis. Physicochemical properties and the transformed microbial count data of raw and fermented camel milk samples were analyzed using the Analyzing of Variance (ANOVA) technique of Completely Randomized Design (CRD) by SAS version 9.0 (SAS, 2004). Each experiment was performed in three replicates.

3. Result and Discussion

3.1. General information of respondents and camel handling

General information of respondents was indicated in Table 1. Most of the respondents in the study areas were females (67%) and the remaining (33%) were males. Age distribution of respondents were 18-30 (37%), 31-45 (40%), 46-65 (36%) and >65 (19%) years old. Majority of respondents were illiterate 116 (88%); whereas 10.5% and 1.5% were in grade 1-8 and 9-12, respectively (Table 1).

Table 1. General information of respondents

	Sex		Age		Educational level			
	N	%	N	%	N	%		
Male	43	33	18-30	37	28	Illiterate	116	88
Female	89	67	31-45	40	30	1-8 grade	14	10.5
			46-65	36	27	9-12 grade	2	1.5
			>65	19	15			
			Total	132	100	132	100	132

The survey result revealed that the majority of camel dairying labor gender division was done by women (Table 2). It was indicated that women are generally responsible for milking (61.5%), processing 81.1%), cleaning milk vessels (85%) and marketing of camel milk (80) where accomplished by women. On the other hand, herding (77%) and barn clearing (51%) were performed by adult men and men (husband), respectively.

Table 2. Gender division in camel dairying responsibility

Household members	Milking N (%)	Processing N (%)	Cleaning N (%)	Selling N (%)	Herding N (%)	Barn clearing N (%)
Adult men	2 (1.5)	0	0	0	102 (77)	60(45.45)
Adult female	34 (26)	25 (18.9)	20 (15)	26 (20)	16 (12)	5 (3.79)
Men	15 (11)	0	0	0	14 (11)	67 (50.76)
Women	81(61.5)	107 (81.1)	112 (85)	106 (80)	0	0
Total	132 (100)	132 (100)	132 (100)	132 (100)	132 (100)	132 (100)

According to respondents, all most all 132 (100%) barn types of camel were fenced. Pastoralists use bush fences that do not have roof by separating different age category of camels. They made separate fence for calves. The purpose of making house for calves separating from young breeding male calves is to get

morning milk while it is used to control mating for young breeding male. Result of the survey showed that 70 (53.03%) of respondent cleared their camel barn once a month while 40 (30.30) and 22 (16.67) of them cleared once a week and do not clean other than changing fence, respectively.

Table 3. Barn facility and cleaning

Variables	Districts			
	Yabello N (%)	Gomole N (%)	Moyale N (%)	Overall N (%)
Types of barn				
Housed	0	0	0	0
Fenced	45 (34.1)	40 (30.3)	47 (35.6)	132 (100)
No barn	0	0	0	0
Frequency of barn clearing				
Once a week	13 (9.85)	15 (11.36)	12 (9.10)	40 (30.30)
Once a month	27 (20.45)	20 (15.15)	23 (17.42)	70 (53.03)
Do not clean	5 (2.27)	5 (2.27)	12 (4.01)	22 (16.67)

3.2. Camel production

Scholars stated, originally, Borana communities are known for their indigenous cattle production where they have a long experience and a very strong attachment to cattle rearing. Camel production becomes the most common livestock species where most of the Borana pastoralists were commonly need to have. On the other hand, camel production was, in some case like traditional taboo, exclusive to the Borana except Gabbra people. However, now days the proportion of Borana people keeping camels is increasing, even if it is not in accordance with their indigenous knowledge and habits. As a result, camel populations increased Borana zone over same time (Dejene, 2015)

Recently, different factors have been induced suppressor impacts on livestock production system of Borana pastoralists. However, cattle have a priority demand in pastoral areas, now a day the preference pastoralists have changed especially due to drought. Respondent reported that camel and goat become the most important livestock types more importantly as much as cattle only due to climate change. The most reason behind this priority and purpose of choice to produce camel is for milk production (85%) during dry season, drought resistance (64%), good price (59%), meat production (58%) and wealth status (49%) indicated as in Table 4.

Table 4 Purpose of keeping camel

Variables	N	Valid Percent	Rank
Milk production	112	85	1
Drought resistance	85	64	3
Good price	78	59	2
Meat production	76	58	4
Wealth status	65	49	5

Table 5. Frequency of milking and production potential of camel

Variables	Season				Overall mean
	LDS	LRS	CDS	SRS	
Frequency of milking	N (%)	N (%)	N (%)	N (%)	-
One times	0	0	0	0	-
Two times	132 (100)	51(38.64	132 (100)	63 (47.73	-
Three times	0	81(61.36	0	69 (52.27)	-
Milk yield L/day (Mean ± SD)	3.79±0.76	6.39±1.39	4.04±0.73	6.30±1.47	5.13±1.10
Lactation length (months)					12.01±3.00

LDS = Long dry season, LRS = Long rainy season, CDS = Cool dry season, SRS = short rainy season and SD = standard deviation

In Borana area, southern Ethiopia, pastoralists and agro pastoralists keep mixed livestock species (livestock diversification) each for a particular purpose. The domestic livestock species kept by them for milk production in the study areas include camels, cows, goats and sheep. Among these, cows and camels are the major milk producing animals in the area. Cow milk followed by camel milk was highly preferred by the pastoralists in the study sites. With regard to preference, cow, camel, goat and sheep milk ranked first, second, third and fourth, respectively (Table 6).

According to the respondents view, milk type from each species has its own unique characteristics and properties. Pastoralists and agro-pastoralists gave many reasons for preference of milk types of their domestic animals. They stated that cow milk can be processed into other milk products easily whereas processing of camel milk into other dairy products is difficult. Cows' milk tend to make people fat, that causes obesity but camel milk gives strength, endurance and stamina. Unlike cows' milk, camel milk has medicinal values and can be used to treat a number of ailments in human beings. The informants also indicated that cows' and sheep milk have high fat content than camel milk and thus suitable for butter-making.

In the study area, camel milk is largely consumed in its raw state without being subjected to any sort of processing treatment. All the households interviewed reported that they use camel milk when it is fresh (Table 6). This observation is in agreement with that reported earlier by Alhadrami (2003) and Eyassu (2007) who indicated that camel milk is consumed fresh in most camel rearing societies. On the other hand, pastoralists and agro-pastoralists exercised making of fermented camel milk (Chuuchee) and butter from camel milk alone or by mixing it with cow or goat milk (Table 6).

Table 6. Preference of milk type, consumption and marketing of camel milk

Preference of milk type	Valid percent	Rank
Cow	46.21	1 st
Camel	39.39	2 nd
Goat	12.12	3 rd
Sheep	2.27	4 th
Consumption of camel milk	N	Proportion (%)
Fresh raw milk	132	100
Chuuchee (sour milk)	62	49.97
Butter	9	6.87
Blended with other milk type	32	23.48

3.3. Camel milk hygienic handling and processing

3.3.1. Camel milk handling and preservation

The survey result revealed that from the total of 132 respondents 96.97% of them did not take training on camel milk hygienic handling and utilization. Study showed that 68.18% of respondents practiced washing hand before milking (Table 7). In the study area, the majority of respondents (72.73%) use pond water while 12.88% and 14.39% bore holes and tape water, respectively for camel dairying activities. Study result indicated that almost all pastoralists and agro-pastoralists heat or warm water before milking to wash milking vessels and clean and smoke milk containers regularly before milking. Respondent explained that the purpose of smoking milk vessels is give flavor and increase shelf life (Table 7). The major traditional methods of Borana pastoralists and agro-pastoralists used to preserve camel milk in the study area include washing and smoking milk vessels (57.5%), keeping milk in a cold place (18.94%) and processing (23.48%) into other products particularly Chuuchee (Table 7).

Table 7. Camel milk handling

Variables	Districts			
	Yabello N (%)	Gomole N (%)	Moyale N (%)	Overall N (%)
Previous training on camel milk				
Yes	1 (0.75)	2 (1.52)	1 (0.75)	4 (3.03)
No	44 (33.33)	38 (28.78)	46 (34.85)	128 (96.97)
Washing of hand before milking				
Yes	30 (22.73)	28 (21.21)	32 (24.24)	90 (68.18)
No	15 (11.36)	12 (9.09)	15 (11.36)	42 (31.82)
Source of water for milking activity				
Pond	35 (26.51)	34 (25.76)	27 (20.45)	96 (72.73)
Bores holes	2 (1.52)	6 (4.54)	9 (6.82)	17 (12.88)
Tap water	8 (6.06)	0	11(8.33)	19 (14.39)
Heat water before milking				
Yes	45 (34.09)	29 (21.97)	46 (34.86)	132
Cleaning and smoking milk containers regularly				
Yes	45 (34.09)	40 (30.30)	47(35.61)	132
Purpose of milk vessels smoking				
Give flavor and increase shelf life	45 (34.09)	35 (26.52)	47 (35.61)	132

Table 8. Traditional preservation methods

Traditional preservation methods	Number and proportion (%) of responses
Washing and smoking milk vessels	76 (57.58)
Keeping milk in a cold place	25 (18.94)
Processing (Chuuchee)	31(23.48)

As indicated in figure 1 most milk containers are generally fumigated with burned woods of trees namely *Premnaschimperi*, *Olea Africana*, *Acacia brevispica* and *Faurea speciosa*. Smoking milk containers has been reported to exert anti-microbial properties and prolong the shelf life of cow milk (Ashenafi 1996). Compounds released from these tree species during smoking of the milk containers may in part be responsible for the longer shelf life of camel milk observed in the present study. Pastoralists and agro-pastoralists in the study area used different milking vessels. Accordingly, 79%, 29%, 21% and 3% of respondents used wood, gorfa, plastic, and okole, respectively for milk vessels (Figure 2). In addition, pastoralists and agro-pastoralists also used garican (89.4%) and sororo (10.6%) vessels for shipping camel milk to market (Figure 3).

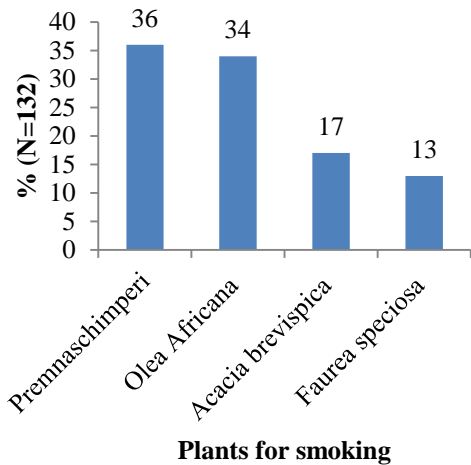


Figure 1. Mostly used plant for smoking

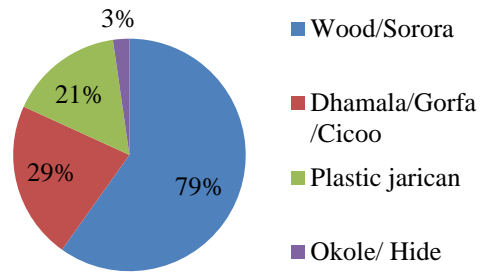


Figure 2. Types of milking vessels

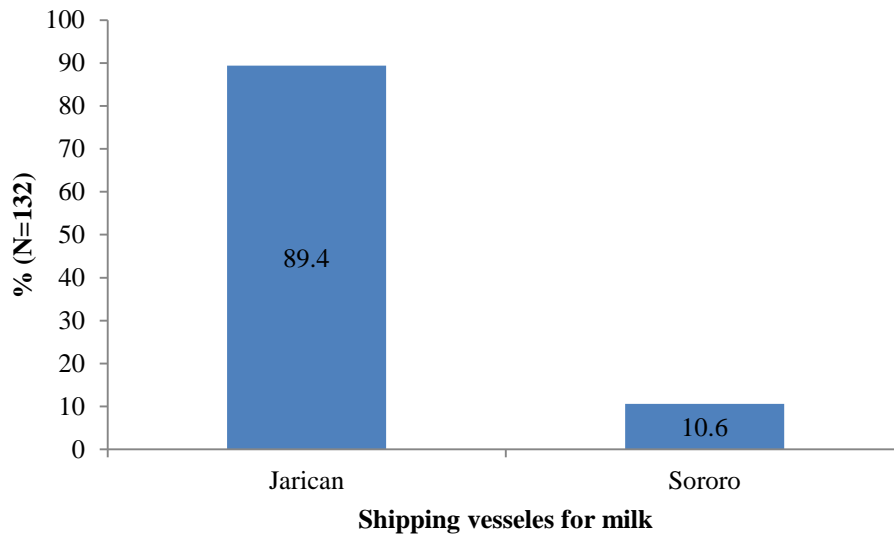


Figure 3. Types of vessels for shipping milk to market

3.3.2. Traditional camel milk processing

The survey result shows that all most all respondent use raw milk for home consumption in the study area. The majority (80.30%) of respondent did no boil milk before consumption. Most of pastoralists and agro-pastoralists (77.27%) in the study area traditionally process camel milk into other camel milk products mainly during surplus milk production (Table 9). Camel milk products that are traditionally made by pastoralists and agro-pastoralists in the study area are indicated in Table 9. Pastoralists and agropastoralists in the study area produce naturally fermented sour camel milk called Chuuchee. Chuuchee is made by placing fresh camel milk in a clean/smoked container and keeping it in a warm (ambient temperature) to allow spontaneous fermentation. Chuuchee is said to have a shelf life of about 3 months. Similar products like *dhanaan*, *shubat* and *gariss* are traditionally made from camel milk were reported from Ethiopia Somali (Eyasu, 2007), Kenya and Sudan (Alhadrami 2003), respectively.

Chuuchee is made by spontaneous fermentation without using a starter culture. Some respondents stated that they mix camel milk with goat milk to make viscous Chuuchee than alone make it from camel milk. Research report showed that the quality of *susac*, fermented camel milk, improved using selected mesophilic lactic starter cultures rather than spontaneous fermentation; the resulting fermented milk had a uniform taste and a longer shelf life (Lore *et al.*, 2005). Isolation and identification of microorganisms that are responsible for the fermentation and production of the indigenous fermented camel milk product, Chuuchee, would help to develop a commercial starter culture and to standardize the manufacturing method for this product in the future.

The majority of the respondents reported that it is difficult to make butter from camel milk. However, small proportion of pastoralists and agro-pastoralists reported that they mix camel milk with cows and goats milk when intentional to make butter (Table 9). Few respondents revealed that butter can be made from camel milk when fermentation is undergone for 7 days. Production of butter from camel milk cannot be achieved easily because camel milk shows little tendency to cream up and also because the fat in camel milk is firmly bound to the protein (Rao *et al.*, 1970). Factors that affect manufacture of butter and optimization of churning and cream separation processes from camel milk may help alleviate the difficulty of butter-making from camel milk.

Table 9. Camel milk processing

Variables	Districts			
	Yabello N (%)	Gomole N (%)	Moyale N (%)	Overall N (%)
Use raw milk for home consumption				
Yes	45 (34.09)	40 (30.30)	47 (35.61)	132 (100%)
Boil milk before consumption				
Yes	3 (2.27)	11(8.33)	12 (9.09)	26 (19.70)
No	42 (31.82)	29 (21.97)	35 (26.52)	106 (80.30)
Camel milk processing				
Yes	29 (21.97)	32 (24.24)	41(31.06)	102 (77.27)
No	16 (12.12)	8 (6.06)	6 (4.55)	30 (22.73)
Type of camel milk products processed				
Chuuchee	10 (7.58)	13 (9.85)	39 (29.55)	62 (46.97)
Butter	2 (1.52)	4 (3.03)	3 (2.27)	9 (6.82)
Blending	15 (11.36)	14 (10.61)	2 (1.52)	31(23.48)

3.4. Camel milk marketing

Apart from its food value, pastoralists and agro-pastoralists generate income from sale of camel milk. Almost all respondents 132 (100%) reported that they sale camel milk to generate income. There are no cooling facilities in the market of study area. Survey result indicated that 75 (56.82) and 73 (55.30) of respondents reported there were spoilage of raw milk due to market problem and problem of marketing fresh milk, respectively (Table 10). Pastoralists and agro-pastoralist sale their camel milk to individuals

(44 (33.33%)), retailers (72 (54.55%)) and hotel/cafeteria (16 (12.12%)). Study result revealed that 120 (90.91%) of respondents buyers did not put milk quality criteria; whereas 12 (9.09%) gave attention on quality milk for buying (Table 10). All the households interviewed in the present study reported that they sale camel milk implies that camel milk has high demand in the market. Even though, camel milk has high demand in the area, camel milk marketing is constrained by price fluctuation, cooling facilities and well-organized transportation and marketing systems.

Table 10. Camel milk marketing

Variable	Districts			
	Yabello N (%)	Gomole N (%)	Moyale N (%)	Overall N (%)
Sell fresh whole milk				
Yes	45 (34.09)	40 (30.30)	47 (35.61)	132
Cooling at market				
No	45 (34.09)	40 (30.30)	47 (35.61)	132
Problem of marketing fresh milk				
Yes	24 (18.18)	20 (15.15)	31(23.48)	75 (56.82)
No	21(15.91)	20 (15.15)	16 (12.12)	57 (43.18)
Spoilage of raw milk due to market problem				
Yes	21 (15.91)	22 (16.67)	30 (22.73)	73 (55.30)
No	44 (33.33)	18 (13.64)	17 (12.88)	79 (59.85)
For whom do you sale your fresh milk				
Individuals	12	15 (11.36)	17 (12.88)	44 (33.33)
Retailers	33 (25)	19 (14.39)	20 (15.15)	72 (54.55)
Hotel/Cafeteria	0	6 (4.55)	10 (7.58)	16 (12.12)
Buyers put quality criteria				
Yes	5 (3.79)	3 (2.27)	4 (3.03)	12 (9.09)
No	40 (30.30)	37 (28.03)	43 (32.57)	120 (90.91)

3.5. Camel milk along supply

This particular assessment of camel milk hygienic handling and processing along milk supply covers Yabello, Gomole and Moyale districts of Borana zone with the specific dairy marketing sites of Yabello, Surupa and Moyale towns. The major sources of raw camel milk supply to traders in Gomole district at Surupa are producers and collectors around Surupa. Traders and collectors check quality of milk by using organoleptic test (watching cleanliness and tasting by tongue). It was stated that milk that below the acceptable limit was rejected. There was no quality based standard payment for milk; however, payment was made as market price for acceptable milk by sensory evaluation. As reported by the traders and other key informants in the area; milk quality problem is not much common except slight poor milk handling at producers and collectors as well as camel milk price fluctuation.

Collectors and traders clean and fumigate their milk Jericans using water and locally available wood chips to reduce deterioration of milk. There were no transportation access and cooling facilities for raw camel milk. Traders supply collected camel milk from Surupa to Moyale in major volume and also to Bule Hora in small quantity depending on seasonal camel milk production. The main agents of raw camel milk supplier to Yabello town were producers, collectors that encampment around Yabello and some traders from Gomole district. Milk supplied to Yabello is consumed and used in the town without shipping to other area since Yabello is capital city of Borana and has high population than other towns in the study areas.

The terminal milk supply for camel milk and milk products in the study area was Moyale. The main suppliers of milk to Moyale are producers in the nearby Moyale town, traders from different parts of Borana and brokers at Moyale. The problem of camel milk is variation of price of milk and price fluctuates greatly seasonal. During surplus production, at wet season, milk left from selling was changed to other camel milk product Chuuchee and sold as sour camel milk. As to traders and brokers perception supply of raw camel milk is inadequate during dry season and the problem of supply shortage is severe during and after incidence of drought. Quality problem is also more common during dry than wet season. However, modes of transportation used are the basic causes of quality deterioration as the opinion of traders and brokers. Prior to milk storage, traders and brokers clean their milk Jeri cans with boiled water and then fumigate with locally available wood chips.

According to camel milk collectors, traders and key informants, adulteration and lack of cooling facilities are the major problems of milk quality at milk supply of the study area. Milk and milk products are very susceptible to adulteration. In the study area respondents revealed that camel milk adulteration increases as the product is moved to market from areas where closer to the pre-urban and urban centers. May be there is less adulteration at production level. Water is used as substance for milk adulteration. In addition, in the study area, lack milk collection equipment like stainless steel, market milk selling shed, quick spoilage of milk due to hot environment, seasonality of milk supply in which excess supply in wet season and extremely low supply in dry season are the main constraints.

3.6. Shelf life and spoilage of camel milk and milk products

The majority of the respondents reported that fresh camel milk can be kept unspoiled for about 2 days (Table 11). This is much shorter than the shelf life of camel milk reported previously by Yagil *et al.* (1984) and Eyassu (2007) indicated that took 7 days for camel milk to sour. When compared to cow milk, camel milk has longer shelf life than cow milk. The better keeping quality of camel milk suggests that it probably contains compounds or substances with strong anti-microbial properties. The majority of respondents stated that Chuuchee can stay for a week whereas some of them revealed that Chuuchee can stay up to 3 months. According to respondents the shelf life of butter was 1 day. However it can be stay for 7 days if it can be kept in quality. The most common types of spoilage that occur in camel milk include souring, ropiness and whey separation (syneresis). These defects are similar to the types of spoilage that occur in cow milk.

Table 11. Shelf life and spoilage of camel milk and milk products

Product type		Shelf life (%)			
Raw milk	1 day (5.30)	2 days (78.79)	3 days (15.91)	-	-
Chuuchee	1 week (50.76)	2 weeks (17.42)	1 month (3.79)	2 months (3.03)	3 months (2.3)
Butter	1 day (4.58)	1 week (2.27)	-	-	-
Spoilage types		N (%)			
Whey separation		20 (15.15)			
Ropiness		30 (22.73)			
Souring		82 (62.12)			

3.7. Therapeutic properties of camel milk

The other benefit of camel milk is its curative value against a number of human diseases. Pastoralists and agro-pastoralists stated that camel milk is used to treat a number of sicknesses in human beings (Table 12). The respondents reported that camel milk is used to treat backbone pain, diarrhea, malaria disease, heart disease, respiratory deceases, and women uterus contracts and shrinks (post delivery). According to the pastoralists and agro-pastoralists prospect, the revealed beneficial property of camel milk is attributed to the fact that camels browse on various plant species and active agents with therapeutic properties from these plant species are secreted into the milk of camels. The medicinal value of camel milk has also been reported by other authors (Yagil 1985; Eyassu, 2007).

Table 12. Therapeutic use of camel milk

Type of disease	Rank
Women uterus contracts and shrinks (<i>abdominal pain</i>)	1
Respiratory deceases	2
Malaria disease	3
Backbone pain	4
Diarrhea	5
Heart disease	6

3.8. Evaluation of raw and fermented camel milk (Chuuchee)

3.8.1. Physicochemical and Microbial Quality of Camel Milk

3.8.1.1. Physicochemical properties of camel milk

Average values for physicochemical properties of camel milk samples are placed in Table 13. The average value of the pH and titratable acidity were 6.62 ± 0.07 and 0.14 ± 0.02 , respectively. The pH values and titratable acidity of some samples may be explained by the production of lactic acid by microbial flora, in particular lactic bacteria, during raw camel milk storage and transportation to the laboratory. Result showed that protein, fat, lactose, SNF, ash and total solids contents of camel milk for preparing Chuuchee were 3.15 ± 0.18 , 3.25 ± 0.27 , 4.62 ± 0.54 , 8.5 ± 0.55 , 0.72 ± 0.02 and 11.74 ± 0.56 , respectively. The chemical composition of camel milk is relatively similar to that of cow milk. The present study is in line with the finding of Farah and Fischer (2004) stated that camel milk composition ranges are: 9.08-14.4% total solids, 2.7-4.5% protein, 3.2-5.5% fat 4.0-5.6% lactose and 0.63-0.9% ash. The distribution of the

values found for chemical composition among camel milk samples could be explained by the effect of various factors such the camel breed, stage of lactation, age and health status, herd management practices and environmental conditions (Al Haj and Al Kanhal, 2010). The average value of camel milk sample density in the present experiment was $1.026 \pm 0.01 \text{ g/cm}^3$. This is similar to the value 1.023 reported by Siboukeur (2007) for Algerian camel milk. Camel milk densities were related to differences in the frequency of camel watering (Rahli *et al.*, 2013).

Table 13. Chemical composition of camel milk

Composition	Mean	SD
pH	6.62	± 0.07
TA (%)	0.14	± 0.03
Protein (%)	3.15	± 0.18
Fat (%)	3.25	± 0.27
Lactose (%)	4.62	± 0.54
SNF	8.50	± 0.55
Ash (%)	0.72	± 0.02
TS (%)	11.74	± 0.56
Density (g/cm^3)	1.026	± 0.01

3.8.1.2. Microbiological profile of raw camel milk

Results obtained by enumeration of different microbial flora of raw camel milk samples are shown in Table 14. The average of total viable count bacterial flora was $7.32 \pm 0.12 \text{ cfu mL}^{-1}$. Average counts of fecal coli-forms and yeast and molds were 7.12 ± 0.16 and $6.33 \pm 0.22 \text{ cfu mL}^{-1}$, respectively. The analyzed samples contained high levels of microbial floras. These results indicate strong microbial contamination of the camel milk samples studied. This can be due to noncompliance with hygienic conditions and rules during production and milking. It should be noted that the camel farming in Borana areas is extensive and the camel herds move during the year in search of feed, and this makes access to water and control of milking conditions very difficult.

Table 14. Microbial count ($\log_{10} \text{ cfu /mL}$) of raw camel milk

Microbial groups	Mean \pm SD
TVCB	7.32 ± 0.12
Coli form	7.12 ± 0.16
Yeast and mold	6.33 ± 0.22

3.8.2. Physicochemical properties and microbial quality of fermented camel milk

3.8.2.1. Physicochemical composition of Chuuchee

Physicochemical properties of home and laboratory made Chuuchee were determined and the results are shown in Table 15. Significant differences ($P < 0.05$) were observed in physicochemical properties between home and laboratory made Chuuchee. Physical properties of fermented dairy product play an important role in determining its quality. Lower pH value was recorded for home made Chuuchee than pH of laboratory made Chuuchee. On the other hand, higher titratable acidity was observed from home made Chuuchee than titratable acidity of laboratory made Chuuchee. The variations in pH and acidity for

Chuuchee could be attributed to the difference in hygiene of the actual milking and the total microbial count and original lactic acid bacteria of the milk that had effects on the final result of spontaneous fermentation of milk (Al and Kanhal, 2010).

Camel milk Chuuchee made at home had significantly higher Protein, Fat, Lactose, SNF, TS and Ash contents than Chuuchee prepared at laboratory (Table 15). The variation of components and total solids incorporated into the Chuuchee might be attributed to the original milk composition and processing condition of Chuuchee preparation. Most of the total solids parts are progressively concentrated into the Chuuchee according to processing and the way used to drain the liquid by slow acidification occurrence of *syneresis* which made spontaneous expulsion of liquid (Walstra *et al.*, 2006). In addition, acidification process plays a key role in eliminating the colloidal minerals of the casein micelles. Consequently, the final solubility level of calcium and phosphorus determines the draining rate and, in turn, the total solids content of the Chuuchee.

Table 15. Chemical composition of fermented camel milk

Composition	Mean \pm SD		Sig.
	Tradition	Lab	
pH	3.81 \pm 0.08	4.08 \pm 0.10	***
TA	1.79 \pm 0.04	1.64 \pm 0.07	***
Protein	6.23 \pm 0.30	5.59 \pm 0.24	***
Fat	7.09 \pm 0.16	6.55 \pm 0.23	***
Lactose	1.49 \pm 0.03	1.39 \pm 0.06	***
SNF	9.61 \pm 0.22	8.93 \pm 0.31	***
TS	15.84 \pm 0.51	14.52 \pm 0.37	***
Ash	1.03 \pm 0.05	0.98 \pm 0.04	**

3.8.2.2. Microbial quality of fermented camel milk

Microbial counts of Chuuchee are shown in Table 16. Results revealed that higher total bacteria, lactic acid bacteria and yeast and mold counts were recorded from home made Chuuchee than in comparison to those Chuuchee made in laboratory. However, there was no coliform recorded from home made Chuuchee than in comparison to those Chuuchee made in laboratory. These variations of microbial counts can be attributed to the fermentation process and acidic pH of the Chuuchee samples. The result has shown that coliforms did not survive in the home made of traditional fermented camel milk than laboratory made fermented camel milk. It has been reported that coliforms are unlikely to grow when the pH of milk has been brought down by lactic acid bacteria to < 4.5 (Walstra *et al.*, 2006) and also smoked milk containers has been reported to exert anti-microbial properties (Ashenafi 1996). In addition the growth of coliforms is slow in the smoked containers (Tesfemariam *et al.*, 2017).

Table 17. Microbial count (\log_{10} cfu /mL) of tradition and laboratory made fermented camel milk

Microbial groups	Tradition	Lab	Significant level
Total bacteria count	4.85±.05	4.59±0.03	**
Coliform	0	4.42±0.08	***
Lactic acid bacteria	5.12±0.02	4.92±0.06	**
Yeast and mold	5.09±0.11	4.76±0.18	***

4. Conclusion and recommendation

The hygienic practice during camel milk production in the study area was poor and camel milk handling practices were also susceptible for contamination. In the study area, the majority of respondents use pond water which is poor in water quality for camel dairying activities. Yet, in the study area most of the respondents were not implemented proper hygienic practices except hand washing before milking. The major traditional methods of Borana pastoralists and agro-pastoralists used to preserve camel milk in the study area were practices smoking and washing milking and storage vessels. All most all respondents use raw milk for home consumption in the study area. The majority of respondent did no boil milk before consumption. Most of pastoralists and agro-pastoralists in the study area traditionally process camel milk into sour camel milk Chuuchee and other camel milk products mainly during surplus milk production. According to camel milk users and key informants, adulteration and lack of cooling facilities are the major problems of milk quality at milk supply of the study area. In addition, lack milk collection equipment like stainless steel, market milk selling shed, quick spoilage of milk due to hot environment, seasonality of milk supply in which excess supply in wet season and extremely low supply in dry season are the main constraints.

Pastoralists and agro-pastoralists generate income from sale of camel milk apart from its food value. Almost all respondents reported that they sale camel milk to generate income. All the households interviewed in the present study reported that they sale camel milk implies that camel milk has high demand in the market. Even though, camel milk has high demand in the area, camel milk marketing is constrained by price fluctuation, cooling facilities and well-organized transportation and marketing systems. The high ambient temperature prevailing in the area which coupled with lack of cooling facilities reduces the shelf life of the milk and thus makes delivery of raw camel milk to the market difficult. Establishment of milk collection centers and introduction of small-scale milk processing plants might help to solve the marketing problem of camel milk in the area.

Chuuchee is made by spontaneous fermentation without using a starter culture. Some respondents stated that they mix camel milk with goat milk to make viscous Chuuchee than alone make it from camel milk. Chuuchee made traditionally was found poor quality and this was attributed to the poor hygienic conditions followed during handling and preparation of Chuuchee. This calls for the need for teaching pastoralists and agro pastoralists to follow proper hygienic practices during milking, milk handling and preparation of Chuuchee. Lactic acid bacteria (LAB) responsible for fermentation of camel milk and production of Chuuchee were not isolated. Thus, isolation of LAB responsible for Chuuchee production deserves detail study in the future.

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The Effects of Blending Camel Milk with Cow, Goat and Sheep Milk on Yield and Quality of Butter in Borana Zone of southern Oromia, Ethiopia

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Abstract

In this study three experiments were conducted at Yabello Pastoral and Dryland Agricultural Research Center (YPDARC) to evaluate the effects of blending camel milk with cow, goat and sheep milk on butter yield and quality. Each of the experiment comprised 5 treatments. Experiment one comprised 5 treatments, that was , pure camel milk (T1), treatment 2, 3 and 4 comprised blending of camel milk with cow milk at proportion of 25, 50 and 75%, respectively, and treatment 5 was pure cow milk. Experiment 2 and 3 were arranged in the same fashion except goat and sheep milk samples were specifically used in experiment 2 and 3 instead of cow milk. The experiments were laid out in completely randomized design. Results showed that camel milk contains all the essential nutrients present in cow`s milk and also has similar composition except fat and total solids. Higher total viable bacteria count (TVBC) was registered for fresh camel milk than for raw cow milk. There were significant differences ($p < 0.05$) among treatments in butter yield recovery. In all levels of camel milk blended with cow milk, higher butter yields were obtained than from pure camel milk, whereas butter yield from pure cow milk was higher than any of those from the other treatments. Statically higher gross composition of pH, fat, SNF, lactose, ash and total solids were recorded for goat milk than for camel milk. Higher coliform bacteria and yeast and mold counts were recorded for camel milk than for goat milk. Butter yield from a blended of 25% camel milk and 75% goat milk (T4) was higher than butter yield of other treatments. Statically higher gross composition (except lactose) was recorded for goat milk than for camel milk. Poor microbial quality was recorded for sheep milk than for camel milk. Higher butter yield was obtained from pure sheep milk than from camel and sheep milk. Generally, blending of camel milk with cow, goat and sheep milk at any level gave better butter yield than pure camel milk.

Keywords: Butter, Camel milk, Churner, Microbial quality, Physicochemical

1. Introduction

The one-humped camel (*Camelus dromedarius*) is an important livestock species uniquely adapted to hot and arid environments (FAO, 2010). The majority of camels in the Ethiopia are found in the drier areas of the Eastern and Southern part of the country. Camels are kept, among other things, mainly for milk production in the pastoral areas. They produce milk for quite longer period, even during dry periods compared to cattle (Kurtu, 2003). Dromedary camels are naturally browsers, thrive on sparse pasture and produce milk where other domesticated animals would virtually starve (Zubeir *et al.*, 2010). This characteristic makes the lactating camel a very valuable animal for the survival of camel herders and their family in harsh environments. Camel milk is an important component of human diet in many parts of the world. It is considered as an important source of protein for the people living in the arid lands of the world (Legesse *et al.*, 2017). The present knowledge about the milk production potential of camels is very

limited. However, a healthy camel on good feed can produce 2000 L of milk per lactation period (Knoess *et al.*, 2008).

Milk and dairy products are part of a healthy diet which, besides cow's milk, sheep's and goats' milk are involved. Cow's milk has long been considered a highly nutritious and valuable human food and is consumed by millions of people daily in variety of products. Goat has been referred as the "poor man's cow" due to its great contribution to the health and nutrition of the landless and rural poor. Goat milk differs from cow or human milk in having better digestibility, alkalinity and buffering capacity (Morteza *et al.*, 2017; Mohapatra *et al.*, 2019).

It is commonly believed that camel milk is difficult to process into products and is only suitable for drinking as fresh or sour milk. However, currently, the possibility of producing various products from camel milk including soft cheese, yoghurt, and butter has been reported (Berheet *et al.*, 2017). Pastoralists claim that it is difficult to churn camel milk to make butter (Yagil, 1982) and further stated that butter from camel milk cannot be obtained so easily using the traditional churning methods because camel milk shows little tendency to cream up and the fat in camel milk is firmly bound to the protein (Rao *et al.*, 2011). On the other hand, reports revealed that butter can be made from camel milk by churning fresh or soured camel milk at 24 to 25°C (Farah *et al.*, 2007; Berhe *et al.*, 2013). In the Algerian Sahara, there is a popular butter made from camel milk and is called Shmen or Semma (Mourad and Nour-Eddine, 2006). In this region, fresh camel milk butter is difficult to preserve because it usually contains many impurities (sand, hair, etc.) and rapidly becomes rancid. The Tuaregs (nomad tribe of Sahara) improve the shelf life of camel milk butter by transforming it into clarified butter oil (Shmen). This product has been playing a major role in the diet of Tuareg communities in the Sahara, and today, there is a special demand for this product among consumers (Mourad and Nour-Eddine, 2006).

In pastoral areas, a large amount of camel milk is produced but butter making from camel milk is difficult due to the inherent characteristics of the milk. In addition to camel milk, milk from cow and other small ruminants particularly goat and sheep is also available in pastoral areas. Thus, there are possibilities to make butter from camel milk by blending it with cow and goat's milk. Hence, knowledge of the factors that influence butter making and the possibilities of churning camel milk to make butter are very important aspects of camel milk processing for enhancing the product and value addition of camel milk that will subsequently enrich the diets and income of the pastoralists. Butter is among dairy products produced in Borana area. Butter has long shelf life as compared to fresh milk, especially when heated to higher temperature (100 to 120°C) for 30 min. It can stay for several months without spoilage (Lejko *et al.*, 2009). Camel milk butter is also used in the preparation of nutritious and medical soups. Also the by-product of butter, that is, buttermilk, is used as a functional ingredient in many food products such as salad dressings, pasta sauces, chocolate, cheese seasonings, ice cream mixes and yoghurt (Fox *et al.*, 2000). Butter making from camel milk has multi dimensional advantages for the pastoral communities of the study area, especially during surplus milk yield. Therefore, this research work was conducted with the following objectives.

Objectives:

- To evaluate the effectiveness of butter obtained from camel milk at different blending levels

- To analysis chemical and microbial quality of butter made from camel milk blended with other milk types

2. Materials and methods

2.1. Description of the study area

The study was conducted in Borana zone at Yabello Pastoral and Dryland Agriculture Research Center. The center is located about 568 km South of Addis Ababa on Addis-Moyale road. The Borana plateau, the portion of the Southern Ethiopia rangelands, comprises an area of about 95,000 km² which is about 14.6% of the lowland areas or about 8.5% of the country's total area. The climate is generally semi-arid with annual rainfall range of 500 mm in the South and 700 mm in the North. Its altitude ranges from 1000m in the South to 1500m in the Northwest and its annual mean daily temperature varies from 19 to 24°C. The area receives a bimodal rainfall with an erratic distribution. Fifty –nine percent of annual precipitation occurs from March to May and 27% from September to November. Four major seasons are identified in Borana plateau. These are: (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).

2.2. Methods of data collection

2.2.1. Milk sample collection

Before collection of camel, cow, goat and sheep milk samples, arrangements were made with local pastoral people to identify the areas with surplus camel milk production. Camel milk samples were collected from households owning camels. The camels used in the experiment were from two to three parities at their second stage of lactation. On the other hand, cows were at their second stage of lactation and of two to fourth parities. In addition, goat and sheep milk samples were collected from 35 and 45 goats and sheep, respectively on their mid to end lactation (5th to the 16th week) stages. After collection, the milk samples were brought to Yabello Pastoral and Dryland Agricultural Research Center laboratory. For fermentation and churning the milks, plastic Jerican/containers of 5 L and 3 L capacity were used for camel-cow and camel-shoat milk blended, respectively. Either the pure camel milk, cow milk, shoats milk or camel milk blended with either the cow or the shoat milk at different proportions were poured into these containers and the milk samples were kept in laboratory at room temperature until the required level of churning time was attained. Churning was done traditionally as well as manually by agitating the milk with up and downward movements. A total of 66 L of camel milk, 331 L of cow and 33 L of shoat milk were collected from the areas mentioned above for butter making.

2.2.2. Experimental layouts and design

In this experiment, three separate experiments each with three treatments consisting of three levels of blending (25, 50 and 75%) camel milk with each of the cow, goat and sheep milk and two other treatments comprising pure milk of respective types were studied in a completely randomized design (CRD). These arrangements are shown in Table 1.

Table 1. Different experimental layouts

Experiment 1 (camel and cow milk)		Experiment 2 (camel and goat milk)		Experiment 3 (camel and sheep milk)	
T1	100% camel milk	T1	100% camel milk	T1	100% camel milk
T2	75% camel 25% cow	T2	75% camel 25% goat	T2	75% camel 25% sheep
T3	50% camel 50% cow	T3	50% camel 50% goat	T3	50% camel 50% sheep
T4	25% camel 75% cow	T4	25% camel 75% goat	T4	25% camel 75% sheep
T5	100% cow	T5	100% goat	T5	100% sheep

2.2.3. Physicochemical analysis of raw milk

Physicochemical properties (pH, fat, protein, lactose, total solids, density, freezing points and ash) of raw camel, cow, goat, and sheep milk were analyzed using Lacto-scan model (Lactoscan SA, 8900 Nova Zagora Bulgaria).

2.2.4. Analysis of physicochemical properties of butter

Some physicochemical properties like moisture content, pH, melting range and yield recovery efficiency of butter samples were determined based on the AOAC (1995) procedure. Moisture content of butter was determined first by placing 5g of butter sample in crucible and immersing it into hot water at 390C and shacking well until creamy consistency is obtained. Then it was dried in atmospheric oven (EDSC, 96H203: England) at 100±5⁰C for 24h until constant weight was obtained. After drying, the butter sample and the crucible were weighed. Then IUPAC (1979) method was used to determine the moisture content of butter.

$$\text{Moisture content of butter} = (W1-W2/W1) \times 100$$

Where, W2 = weight of butter sample after drying (final weight)

W1 = weight of butter sample before drying (original weight)

pH of butter was determined according to the method of Weckel (1932). , Thirty five grams of a representative and well mixed samples of butter were taken and warmed in a water bath at 55-60⁰C. The flask was whirled in a centrifuge at a speed of 60 revolutions per minute, for one minute. The serum of the butter was separated out and placed in a flask. Then the pH of the serum was measured using digital pH meter (Crison Basic 20, Barcelona).

Melting range of butter samples was determined by holding a test tube containing 2g of butter into a half-filled beaker with water. The water in the beaker was heated slowly in a water bath while the tube was gently shaken. As soon as the butter was completely melted, the tube was removed from the water and the temperature of the melting range of the butter sample was measured using a thermometer.

2.2.5 Micro-biological analysis of raw milk and butter

Microbiological counts of milk samples considered were total plate count, coliform count (CC) and yeast and mould count (YMC). Proteolytic bacteria, total plate count, coliform count (CC) and yeast and mould count (YMC) were determined also for butter samples. For determination of these counts, peptone water was sterilized by autoclaving at 121⁰C for 15 minutes. The total plate count agar used for determination of total viable organisms, skim milk agar used for determination of proteolytic bacteria and potato dextrose agar used for determination of yeast and mould count were sterilized by autoclaving at 121⁰C for 15 minutes, while the violet red bile agar used for determination of CC was sterilized by boiling (Richardson, 1985). The Medias used for all counts were prepared according to the guidelines given by the manufacturers. Each count was done in duplicate. For microbiological analysis of butter, one gram of the prepared butter samples were transferred into a sterile test tube having 9ml of warm sterile peptone water (39±1⁰C) to prepare a dilution of 10⁻¹ from which decimal dilutions up to 10⁻⁵. A Standard Plate Count Agar, Violet Red Bile Agar, Skim Milk Agar and Potato Dextrose Agar were used for determination of total bacterial count, coli form count, proteolytic bacteria and yeast and mould count, respectively. All dilutions and counts of butter were analyzed according to what Richardson (1985) used for milk.

Total viable bacteria count (TBC) was made by adding 1ml of milk sample into sterile test tube having 9ml of peptone water. After thorough mixing, the sample was serially diluted up to 1:10⁻⁵ dilution level and duplicate samples (1ml) were poured plated using 15 - 20ml of molten standard plate count agar. The plated samples were allowed to solidify and then incubated at 32⁰C for 48 hours. Colony count was made using colony counter (Marth, 1978).

Coli form count was done by adding one ml of milk sample into sterile test tube having 9 ml of peptone water. After mixing, the sample was serially diluted up to 1: 10⁻³ dilution level and duplicate samples (1ml) were poured on plated agar using 15 - 20ml of molten Violet Red Bile agar. After thorough mixing, the plated sample was allowed to solidify and then incubated at 30⁰C for 24 hours. Finally, colony count was made using colony counter (Abdel-Rahman *et al.*, 2009). Typical dark red colonies were considered as coli forms.

Yeast and mould count was done by adding one ml of milk sample into sterile test tube having 9 ml of peptone water. After mixing, the sample was serially diluted up to 1: 10⁻³ dilution level samples (1ml) were pour plated using 15 - 20ml of molten potato dextrose agar. After thorough mixing, the plated sample was allowed to solidify and then incubated at 25⁰C for 5 days. Finally, colony count was made using colony counter (Marth, 1978).

Proteolytic bacteria count was also done by adding one g of butter sample into sterile test tube having 9 ml of peptone water. After mixing, the sample was serially diluted up to 1: 10⁻³ dilution level samples (1g) were poured plated using 15 - 20ml of molten skim milk agar. Proteolytic bacteria were counted using skim milk agar. The plates were incubated at 28⁰C for 7 days. Colonies with clear zones of hydrolysis were counted as proteolytic bacteria (Marth, 1978).

2.3. Data management and analysis

Analysis of variance (ANOVA) was used for analyzing of milk composition, microbial quality, the amount of butter yield, by using SAS (2004). Significant differences were confirmed at 5% significance level. The model for the treatment was:

$$Y_{ij} = \mu + t_i + \epsilon_{ij}$$

Where, Y_{ij} = the j^{th} observation of the i^{th} treatment

μ = overall mean

t_i = the treatment effect (blend level) of the i^{th} treatment

ϵ_{ij} = random error

3. Results and Discussions

Experiment 1: Analyzing the Effects of Blending Camel Milk with Cow Milk on Butter Yield and Quality

3.1.1. Physicochemical properties of camel and cow milk

Mean composition of camel and cow raw milk samples used for butter preparation is shown in Table 2. It was reported that camel milk contains all the essential nutrients present in cow's milk and its composition is similar to that of cow's milk (Yagil, 1982; Morton, 1984). Results presented in Table 2 show that the values for the physical and chemical properties of camel milk and cow milk are similar except pH, titratable acidity (expressed as lactic acid %), fat and total solids contents. In this experiment lower pH, fat and total solids were recorded for camel milk than for cow milk. On the other hand, higher titratable acidity was recorded for camel milk compared to the cow milk.

Khaskheli *et al.* (2005) found higher pH (6.77) and same titratable acidity (0.18%) for camel milk. The variations in pH could be attributed to the difference in hygiene of the actual milking and the total microbial count of the milk (Al and Kanhal, 2010). The total solids and Solid Not Fat (SNF) values of camel milk observed in this study are higher than the values of 11.39 and 7.60%, respectively reported by El-Zubeir (2016) for camel milk. The protein values of camel milk recorded is in line with the value of 3.20% reported by Mehaia (2006) for camel milk. However, Mehaia (2006) reported higher values of fat and ash (3.60 and 0.8%, respectively) for camel milk than the values obtained in the present study. The fat content of camel milk in the current study is different from the earlier report of El-Zubeir (2016), who reported fat content of 2.66%.

Table 2. Physicochemical properties of camel and cow milk

Variables	Camel milk	Cow milk	P-value
pH	6.67±0.01	6.76±0.02	0.0022
TA (%)	0.18±0.01	0.16±0.01	0.0249
Fat (%)	3.37±0.14	4.12±0.16	0.0035
Protein (%)	3.20±0.09	3.03±0.08	0.0776
SNF	8.67±0.09	8.45±0.16	0.1072
Lactose (%)	4.49±0.21	4.65±0.09	0.3167
Ash (%)	0.72±0.01	0.78±0.04	0.0603
TS (%)	11.79±0.04	12.58±0.19	0.0022
Density (g/cm ³)	1.025±0.01	1.026±0.02	0.2051
Freezing point (°C)	-0.584±0.02	-0.559±0.19	0.1743

3.1.2. Microbial quality of camel and cow milk

Microbial count of milk samples used for butter making is indicated in Table 3. Higher total viable bacteria count (TVBC) was observed in fresh camel milk than in fresh cow milk (Table 3). This may be attributed to poor hygienic practices in handling camel milk by pastoral areas as compared to the condition in the Dairy Farm of YPDARC. On the other hand, there were no significant differences observed in coli form and yeast and mold counts between camel and cow milk samples. The total coliform count of camel milk observed in the present study is lower than the values of 5.7 Log₁₀ cfu/ml and 6.8 Log₁₀ cfu/ml reported by Al-Mohizea (1994) and Benkerroum *et al.* (2003), respectively. On the other hand, the count of yeast and mould of milk in the present study is in line with the value of 4.6 Log₁₀cfu/ml reported for yeast count by Benkerroum *et al.* (2003). Higher microbial counts of a milk samples indicate poor hygienic practices during production and processing.

The TVBC of cow milk observed in the present study is slightly less than the TVBC values of 7.58 and 7.6 Log₁₀cfu/ml reported by Asaminew and Eyassu (2010) and Alganesh *et al.* (2007), respectively. On the other hand, the TVBC of cow milk observed in the present study is higher than the acceptable standard of TVBC (5×10⁵cfu/ml) (O'Connor, 1995). The total coliform count of cow milk observed in the present study is higher than the values of 4.5 and 4.49 log₁₀cfu/ml reported by Alganesh, *et al.* (2007) and Asaminew and Eyassu (2010), respectively.

Table 3. Microbial count (mean log₁₀cfu/ml) of camel and cow milk samples used for butter making (mean±SD)

Milk type	TVBC	Coliform	Yeast and mold
Camel	7.32±0.02	5.17±0.06	4.64±0.05
Cow	7.25±0.03	5.07±0.15	4.57±0.13
P-value	0.0366	0.3358	0.3731

3.1.3. Acidification process of camel milk, cow milk and their blends

Figure 1 indicates the acidification process of pure camel milk, cow milk and their blends to attain churning pH ≈ 4.25 during fermentation for butter production. This acidification process affects a number of aspects of the milk components and finally butter properties. Results showed that pure camel milk registered slower acidification time than the other treatments. With increased proportions of cows' milk in the blend, the acidification time for attaining churning pH decreased. The variation of this acidification time may be due to the distinctive structural and functional properties of the composition of camel milk. It was reported that the main reason for long time fermentation of raw camel milk and difficulty of producing products from camel milk is owing to the unique structural and functional properties of the milk components (Berheet *al.*, 2017).

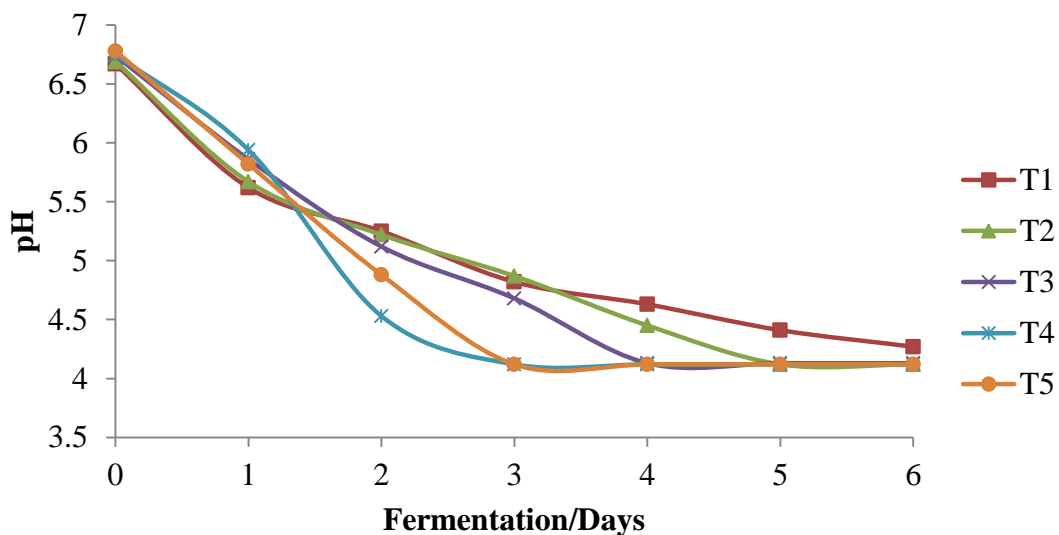


Figure 1. Acidification process of camel and cow milk: T1 = 100% camel milk; T2 = 75% camel milk and 25% cow milk; T3 = 50% camel milk and 50% cow milk; T4 = 25% camel milk and 75% cow milk; T5 = 100% cow milk.

3.1.4. Physicochemical properties of butter made from camel milk, cow milk and their blends

Statistical analysis showed that there were significant differences ($p < 0.05$) in butter physicochemical properties, except in churning temperature, among treatments (Table 4). The fermentation time was longer (6 days) for pure camel milk shorter (3 days) for pure cow milk. Higher pH value of fermented milk was recorded for T1 and T4, whereas lower and almost similar values were recorded for treatments T2, T3 and T5. Butter yield obtained from cow milk (T5) was significantly higher ($p < 0.05$) than butter yield obtained from all other treatments. The lower butter yield was obtained from pure camel milk (T1) whereas the butter yield obtained from T2, T3 and T4 had intermediate values ranging from 5.43-5.73%, with no significant difference among these treatments. Results this experiment showed that blending camel milk with cow milk at any level resulted in significantly higher butter yield as compared to better butter yield obtained from pure camel milk (T1).

Significant differences in butter pH were observed among treatments during butter collection (Table 4). Higher pH value of butter was recorded in T1 than in the other treatments, whereas lower butter pH was recorded in T5. Significant differences ($p < 0.01$) were observed in total solids (TS) content of butter among treatments (Table 4). The total solids content of T1 was significantly ($p < 0.01$) lower than the total solid contents of the other treatments. On the other hand, significantly higher total solids were recorded for T5 than for the other four treatments. There were also significant differences ($p < 0.05$) among treatments in butter yield recovery (Table 4). The butter yield recovery percentage of pure cow milk (T5) was significantly higher ($p < 0.01$) than that of in the other treatments. There were significant differences ($p < 0.001$) in melting point of butter values observed among treatments (Table 4). The melting point of butter samples obtained from pure camel milk (T1) was significantly higher than the values of butter obtained from T2, T3, T4 and T5. This is in agreement with the values (41.4-44.1°C) reported by Park and Haenlein (2006) for butter from camel milk. The lowest melting point was observed in T5 (pure cow milk butter).

Table 4. Physicochemical properties of butter made from camel milk, cow milk and their blends

Variable (mean±SD)	Treatment				
	T1	T2	T3	T4	T5
F (days)	6.00 ^a ±0.00	5.00 ^b ±0.00	4.00 ^c ±0.00	4.00 ^c ±0.00	3.00 ^d ±0.00
FMpH	4.27 ^a ±0.04	4.12 ^b ±0.01	4.15 ^b ±0.02	4.25 ^a ±0.05	4.16 ^b ±0.01
Chti (min)	115.00 ^a ±5.00	97.67 ^b ±2.52	80.67 ^c ±0.15	69.33 ^d ±1.15	57.67 ^e ±2.52
Chte (°C)	23.5±0.50	23.17±0.76	23.5±0.50	23±0.5	22.67±0.76
Yield (g/4lit)	160.00 ^c ±10.00	217.33 ^b ±14.19	217.37 ^b ±14.79	229.17 ^b ±3.62	254.05 ^a ±7.44
BpH	4.82 ^a ±0.04	4.75 ^b ±0.03	4.65 ^c ±0.03	4.44 ^d ±0.02	4.28 ^e ±0.03
TS (%)	67.57 ^c ±0.60	70.94 ^d ±0.18	73.57 ^c ±1.27	76.19 ^b ±0.32	80.01 ^a ±0.36
BYRE (%)	4.00 ^c ±0.25	5.43 ^b ±0.35	5.44 ^b ±0.37	5.73 ^b ±0.09	6.35 ^a ±0.19
MR (°C)	43.00 ^a ±0.20	42.23 ^b ±0.25	41.11 ^c ±0.10	40.30 ^d ±0.04	38.55 ^e ±0.29

a, b,c,d,e = means with the same superscript letters in a row are not significantly different from each other ($P>0.05$: F= fermentation; FMpH = fermentation milk pH; Chti= churning time; Chte=churning temperature; BpH= butter pH; BYRE butter yield recovery efficiency; MR= melting point; T1 = 100% camel milk; T2 = 75% camel milk and 25% cow milk; T3 = 50% camel milk and 50% cow milk; T4 = 25% camel milk and 75% cow milk; T5 = 100% cow milk.

3.1.5. Microbial quality of Butter made from camel milk, cow milk and their blends

Microbial quality parameters for butter made from camel milk, cow milk and their blends is given in Table 5. Except in yeast and mold counts, statistically significant differences ($P< 0.05$) were observed in total viable bacteria, proteolytic bacteria and coliform bacteria counts among treatments (Table 5). Among the treatments, higher total viable bacteria count was recorded for butter made from milk in T3 compared with butter made from milk in T2 and T4. The TVBC observed in butter samples made from pure camel milk (T1) in the present study is higher than the finding of Mourad and Nour-Eddine (2006) who reported $3.54 \pm 0.19 \log_{10}$ cfu/g for total bacteria count in traditional butter (*Shmen*) made from camel milk in Saidaregions of Algeria. On the other hand, TVBC observed in T1 is slightly in line with the finding of Asresie *et al.* (2013) who stated $4.35\pm 0.01 \log_{10}$ cfu/g in total bacteria count of butter made from pure camel milk. The high count of total bacteria observed in the present study may be attributed to the use of raw milk samples for butter-making.

Statistically, higher proteolytic bacteria counts were obtained in butter made from T1 and T2 milk as compared with butter made from T3, T4 and T5 milk. The lipolytic bacteria count observed in butter sample made from T1 in the present study is higher than the result reported earlier by Asresie *et al.* (2013) who reported a lipolytic count of $3.64\pm 0.02 \log_{10}$ cfu/ml. Lipolytic bacteria are one of the spoilage microorganisms in butter and used to predicate the shelf life and the keeping quality of butter (Lejko *et al.*, 2009). Higher coliform bacteria count was observed in butter made from T3 and T4 milk than butter made from T1 and T5 milk. The coliform count observed in butter samples made from pure camel milk (T1) in the present study is higher than the results reported earlier by Asresie *et al.* (2013) who reported a coliform count of $3.07\pm 0.00 \log_{10}$ cfu/ml. The high coliforms counts of camel milk butter may be a consequence of the low level of hygiene during milking and handling of the milk prior to butter-making. Significant difference ($P>0.05$) was not observed in this experiment among treatments in yeast and mold counts. In the present study, butter made from T1 is in line with previous finding of Asresie *et al.* (2013)

who stated $5.27 \pm 0.004 \log_{10}$ cfu/g in yeast and mold count of butter made from pure camel milk. The presence of yeasts in butter might be attributed to contamination from air or to the lack of proper hygiene during butter-making process, while the presence of molds indicates contamination of the product from air during the preparation of butter (Walstra *et al.*, 2006).

Table 5. Microbial count (\log_{10} cfu/g) of the butter samples made from camel milk, cow milk and their blends (mean \pm SD)

Treatment	TVBC	Proteolytic	Coli form	Yeast and mold
T1	4.76 ^{ab} \pm 0.09	5.11 ^a \pm 0.05	4.56 ^{bc} \pm 0.06	5.30 \pm 0.01
T2	4.67 ^b \pm 0.06	5.06 ^a \pm 0.05	4.78 ^{ab} \pm 0.10	5.29 \pm 0.02
T3	4.91 ^a \pm 0.02	4.97 ^b \pm 0.03	4.90 ^a \pm 0.09	5.26 \pm 0.01
T4	4.74 ^b \pm 0.06	4.94 ^{bc} \pm 0.02	4.88 ^a \pm 0.22	5.14 \pm 0.14
T5	4.79 ^{ab} \pm 0.14	4.90 ^c \pm 0.01	4.52 ^c \pm 0.12	5.24 \pm 0.02

^{a, b, c} means with the same superscript letters in a column are not significantly different from each other ($P > 0.05$). T1 = 100% camel milk; T2 = 75% camel milk and 25% cow milk; T3 = 50% camel milk and 50% cow milk; T4 = 25% camel milk and 75% cow milk; T5 = 100% cow milk.

Experiment 2: Analyzing the Effects of Blending Camel Milk with Goat Milk on Butter Yield and Quality

2.1 Physicochemical properties of camel and goat milk

The mean composition of camel and goat raw milk samples used for butter preparation is shown in Table 6. It was observed from results that statically higher pH, fat, SNF, lactose, ash and total solids were recorded for goat milk than for camel milk. The titratable acidity values of 0.14% obtained for goat milk in present study was less than the earlier report of Legesse *et al.* (2017), whereas the pH value of 6.82 was higher than mentioned report. The fat content of goat milk in the present study is in agreement with values of 3.16 - 4.73% reported by Mahmood and Usman (2010). The protein content of goat milk in the present study is also in line with the protein contents 2.05-6.00% reported by Legesse *et al.* (2017). In the present study, higher lactose and total solids contents of goat milk was recorded than the values of lactose and total solids of 4.44 and 12.62%, respectively reported by Mayer and Fiechter (2012). The value of ash obtained for goat milk in the present study is in agreement with the ash content of 0.56-0.80% illustrated in the finding of Legesse *et al.* (2017).

Table 6. Physicochemical properties of camel and goat milk

Variables	Camel milk	Goat milk	P-value
pH	6.68 \pm 0.01	6.82 \pm 0.03	0.0010
TA (%)	0.15 \pm 0.01	0.14 \pm 0.01	0.0705
Fat (%)	3.35 \pm 0.14	4.12 \pm 0.19	0.0055
Protein (%)	3.22 \pm 0.09	3.26 \pm 0.09	0.4583
SNF	8.42 \pm 0.09	9.10 \pm 0.13	0.0091
Lactose (%)	4.46 \pm 0.21	5.06 \pm 0.08	0.0121
Ash (%)	0.74 \pm 0.01	0.76 \pm 0.02	0.0241
TS (%)	11.77 \pm 0.04	13.18 \pm 0.29	0.0012
Density (g/cm ³)	1.025 \pm 0.00	1.027 \pm 0.01	0.0572
Freezing point (⁰ C)	-0.5840.02	-0.613 \pm 0.00	0.0920

3.2.2. Microbial quality of camel and goat milk

Microbial counts of camel and goat milk samples used for butter making are indicated in Table 7. Higher coli form bacteria and yeast and mold count of fresh camel milk were recorded in the present study than the respective counts in fresh goat milk (Table 7). However, there was no significant difference observed in total viable bacteria count between camel and goat milk samples. The total viable bacteria, coliform bacteria and yeast and mold counts of goat milk observed in the present study are higher than the values (4.57 log₁₀ cfu/ml, 4.22 log₁₀cfu/ml and 3.09 log₁₀ cfu/ml) of goat milk reported by Asresie *et al.* (2013).

Table 7. Microbial count (mean log₁₀cfu/ml) of camel and goat milk samples used for butter making (mean±SD)

Milk type	TVBC	Coli form	Yeast and mold
Camel milk	7.32±0.02	5.17±0.06	4.59±0.03
Goat milk	7.33±0.03	4.94±0.01	4.13±0.05
P-value	0.5665	0.0035	0.0002

3.2.3 Acidification process of camel milk, goat milk and their blends

Acidification process of pure camel and goat milk as well as their blends to attain churning pH ≈ 4.25 during fermentation for butter production is indicated in Figure 1. Results showed that T1 (pure camel milk) resulted in slower acidification time than all the other treatments. With increased proportions of goat's milk in the blend, the acidification time for attaining churning pH decreased. The variation of these acidification time and pH values may be due to the unique structural and functional properties of both camel and goat milk components.

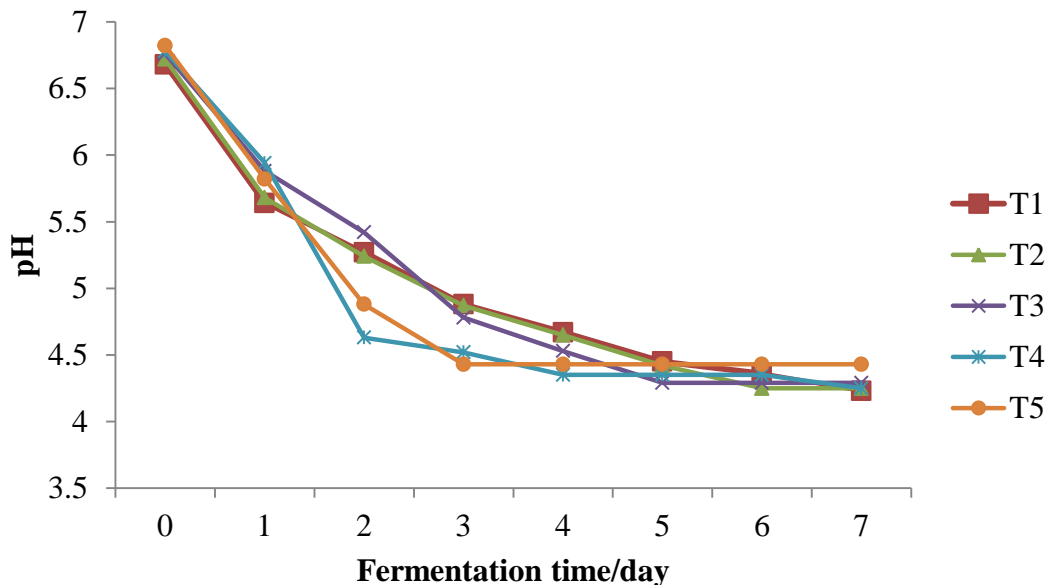


Figure 2. Acidification process of camel and goat milk blended: T1 = 100% camel milk; T2 = 75% camel milk and 25% goat milk; T3 = 50% camel milk and 50% goat milk; T4 = 25% camel milk and 75% goat milk; T5 = 100% goat milk.

3.2.4. Physicochemical properties of butter made from camel milk, goat milk and their blends

Physicochemical properties of butter made from camel milk, goat milk and their blends are shown in Table 8. Statistical analysis showed that there were significant differences ($P<0.05$) in butter physicochemical properties among treatments. Long fermentation time (7 days) was observed for pure camel milk while shorter (3 days) was observed for pure goat milk. Higher pH value of fermented milk for churning was recorded in T5 than the churning pH of fermented milk in T1, T2, T3 and T4. Pure camel milk (T1) required significantly higher ($P<0.001$) temperature for churning as compared to the other milk samples, whereas the churning temperature of pure goat milk (T5) was significantly lower ($P<0.001$) than that of the other milk samples (Table 8). With increased proportions of goats' milk in the blend, the churning temperature decreased. The churning temperature recorded for pure camel milk in the present study is in agreement with the finding of (Farah *et al.*, 1989) who reported that butter can be made from camel milk by churning the fermented milk at 20-25⁰C.

The churning time of pure camel milk (T1) was significantly ($P<0.001$) longer than that of the other milk samples, while the churning time of pure goat milk was significantly ($P<0.001$) shorter than the others. With increased proportions of goats' milk in the blend, the churning time kept on decreasing (Table 8). The reason for the different churning behavior of fat from camel milk in comparison with the fat from goat milk can partly be attributed to the high melting point of fat from camel milk. This seems to shift the ideal ratio of solid to liquid fat in the fat globules at a given temperature towards a point higher than that of goat milk fat. The different churn ability observed could also be attributed to the reported small size of fat globules of camel milk (Yagil, 1982). Butter yield from camel and goat milk blends (T4) was significantly higher ($p<0.05$) than butter yield from T1, T2, T3 and T5. Low butter yield was obtained from pure camel milk compared with butter yield obtained from milk samples T2, T3, T4 and T5. Result of this experiment showed that blending of camel milk with goat milk at any level had significant effect on butter yield in such a way that more butter is obtained from the blend as compared to from pure camel milk (Table 8).

The total solids content of butter made from pure goat milk (T5) was significantly ($P<0.001$) higher than those made from milk samples of other treatments (Table 8). Butter made from pure camel milk had the lowest total solids content as compared to the butter made from other milk samples. With increased proportion of goats' milk in the blend, an increase in total solids content of butter was observed (Table 8). The butter yield recovery from milk samples of T4 was significantly higher ($p<0.01$) than the butter yield recovery from other milk samples. Significant differences ($p<0.001$) in melting point of butter was observed among treatments (Table 8). The melting point of butter samples made from pure camel milk (T1) was significantly higher than the values observed for butter made from milk samples in T2, T3, T4 and T4 (Table 8).

Table 8. Physicochemical properties of butter made from camel milk, goat milk and their blends

Variable	Treatment				
	T1	T2	T3	T4	T5
F (days)	7.33 ^a ±0.58	6.00 ^b ±0.00	5.00 ^c ±0.00	4.00 ^d ±0.00	3.00 ^e ±0.05
FMpH	4.23 ^c ±0.02	4.25 ^{bc} ±0.068	4.29 ^{bc} ±0.02	4.35 ^c ±0.04	4.43 ^a ±0.06
Chti (min)	117.33 ^a ±4.04	92.33 ^b ±2.52	41.67 ^c ±1.53	25.33 ^d ±1.52	14.67 ^e ±0.58
Chte (°C)	27.67 ^a ±0.29	21.50 ^b ±0.50	17.67 ^c ±0.27	15.33 ^d ±0.28	12.16 ^e ±0.29
Yield (4lit)	99.33 ^e ±10.06	137.91 ^d ±2.45	205.40 ^b ±4.22	250.38 ^a ±5.51	190.2 ^c ±9.36
BpH	4.74 ^a ±0.02	4.71 ^b ±0.01	4.65 ^c ±0.02	4.45 ^d ±0.02	4.46 ^d ±0.01
TS (%)	72.61 ^d ±0.99	65.56 ^c ±1.52	73.58 ^c ±2.99	78.20 ^b ±0.30	82.19 ^a ±0.44
BYRE (%)	4.97 ^e ±0.50	6.89 ^d ±0.13	10.27 ^b ±0.21	12.52 ^a ±0.28	9.51 ^c ±0.47
MR (°C)	42.50 ^a ±0.03	34.00 ^b ±0.50	24.83 ^c ±1.04	18.5 ^d ±0.50	14.83 ^e ±0.28

^{a, b, c, d, e} = Means with the same superscript letters in a row are not significantly different from each other ($P>0.05$) F= fermentation; FMpH = fermentation milk pH; Chti= churning time; Chte=churning temperature; BpH= butter pH; BYRE butter yield recovery efficiency; MR= melting point; T1 = 100% camel milk; T2 = 75% camel milk and 25% goat milk; T3 = 50% camel milk and 50% goat milk; T4 = 25% camel milk and 75% goat milk; T5 = 100% goat milk.

3.2.5. Microbial quality of butter made from camel milk, goat milk and their blends

The microbial quality of butter made from camel milk, goat milk and their blends is shown in (Table 9). The total viable bacteria counts (TVBC) of butter made from milk samples in T1, T3 and T4 were significantly higher than the butter made from milk samples of T2 and T5. The total bacteria count observed in butter samples made from pure camel milk (T1) and from blended milk (T2, T3 and T4) in the present study are higher than the results reported earlier by Asresie *et al.* (2013) who reported the values of 4.35; 4.54, 4.51 and 4.35 Log₁₀cfu/ml for butter made from pure camel milk and the blended milk, respectively. The high count of total bacteria observed in the present study may be attributed to the use of raw milk samples for butter-making. On the other hand, lower value of TVBC was recorded in butter made from pure goat milk as compared to the value of 4.72 log₁₀cfu/ml found by Asresie *et al.* (2013) for butter made from pure goat milk.

The proteolytic count (PC) of butter samples made from pure camel milk (T1) and from blended milk in T2 were significantly higher than PC of butter made from blended milk of T3, T4 and T5 (pure goat milk). The proteolytic bacteria counts observed in butter samples made from milk of all treatments in the present study are higher than the results reported earlier by Asresie *et al.* (2013). This author reported the PC values of 3.64, 3.31, 2.70, 2.19 and 4.24 Log₁₀cfu/ml for butter made from pure camel milk, from blended milk and from pure goat milk, respectively. There was no significant ($P>0.05$) difference in coliform count among the treatments. Statistically higher yeast and mold values were recorded in butter made from milk of T1 and T2 than butter made from milk of T4 and T5. The yeast and mold count observed in butter samples made from pure camel milk (T1) in the present study is in line with the value of 5.27 log₁₀cfu/ml reported by Asresie *et al.* (2013) for butter made from pure camel milk.

Table 9. Microbial count (\log_{10} cfu/g) of the butter samples made from camel milk, goat milk and their blends (mean \pm SD)

Treatment	TVBC	Proteolytic	Coli form	Yeast and mold
T1	4.84 ^a \pm 0.02	5.04 ^a \pm 0.02	4.67 \pm 0.05	5.28 ^a \pm 0.01
T2	4.76 ^b \pm 0.01	5.04 ^a \pm 0.03	4.67 \pm 0.11	5.24 ^a \pm 0.01
T3	4.86 ^a \pm 0.06	4.93 ^b \pm 0.06	4.78 \pm 0.15	5.17 ^{ab} \pm 0.13
T4	4.85 ^a \pm 0.02	4.94 ^b \pm 0.02	4.81 \pm 0.04	5.11 ^b \pm 0.03
T5	4.46 ^c \pm 0.06	4.82 ^c \pm 0.01	4.62 \pm 0.04	4.98 ^c \pm 0.04

^{a, b, c,} = Means with the same superscript letters in a column are not significantly different from each other ($P>0.05$). T1 = 100% camel milk; T2 = 75% camel milk and 25% goat milk; T3 = 50% camel milk and 50% goat milk; T4 = 25% camel milk and 75% goat milk; T5 = 100% goat milk.

Experiment 3: Analyzing the Effects of Blending Camel Milk with Sheep Milk on Butter Yield and Quality.

3.1. Physicochemical properties of camel and sheep milk

The mean composition of camel and sheep raw milk samples used for butter preparation is shown in Table 10. Statically higher composition of titratable acidity fat, protein, SNF, ash and total solids were recorded for sheep milk than for camel milk. Similar pH and lactose contents were obtained for both camel and sheep milk. The value of pH recorded for sheep milk in the present study is in agreement with the values of 6.51–6.85 reported by Mohapatra *et al.* (2019) for sheep milk whereas the titratable acidity was lower than the values of 0.22–0.25% reported by this same author.. The fat content of sheep milk in the present study is also in agreement with results of Mohapatra *et al.* (2019) who reported a fat content of 6.99% for sheep milk. On the other hand, milk in the present study produced higher fat percentage than the fat percentage of 5.90% previous reported by Balthazar *et al.* (2017) for sheep milk.

The protein content of sheep milk in the present study is within the range of 4.50–6.60 % protein reported by Mohapatra *et al.* (2019), whereas it was lower than the protein content of 5.50% reported by Balthazar *et al.* (2017). The lactose content observed in the present study was also in line with the lactose contents of 3.90–4.90% reported by Mohapatra *et al.* (2019), for sheep milk, whereas it was lower than the value of 4.80% reported by Balthazar *et al.* (2017). The value of ash obtained for sheep milk in the present study is slightly higher than the value of 0.9% illustrated in the finding of Balthazar *et al.* (2017). The total solid content of sheep milk in the present study is less than the total solid content of 18.50% reported by Mohapatra *et al.* (2019), whereas it was slightly greater than the total solids content of 17.1% indicated in the report of Balthazar *et al.* (2017). The chemical composition of fresh sheep milk varies over time and depends on several factors, such as the stage of lactation, parity, season, environmental temperature, lactation efficiency, animal age and nutrition, genetic factors (species and breed), and diseases of the udder (Tamime *et al.*, 2011; Claeys *et al.*, 2014).

Table 10. Physicochemical properties of camel and sheep milk

Variables	Camel milk	Sheep milk	P-value
pH	6.47±0.24	6.79±0.01	0.0832
TA (%)	0.16±0.02	0.13±0.01	0.0502
Fat (%)	3.21±0.18	6.99±0.21	0.0001
Protein (%)	3.12±0.18	4.91±0.12	0.0001
SNF	8.34±0.10	10.49±0.12	0.0001
Lactose (%)	4.49±0.21	4.62±0.06	0.3761
Ash (%)	0.72±0.01	0.96±0.01	0.0015
TS (%)	11.55±0.16	17.48±0.33	0.0001

3.3.2. Microbial quality of camel and sheep milk

Microbial counts of camel and sheep milk samples used for butter making are indicated in Table 11. Higher ($P<0.05$) total viable bacteria, coli form bacteria and yeast and mold counts were recorded for fresh sheep milk than for fresh camel milk (Table 11). Milk synthesized in a healthy ovine mammary gland contains relatively low numbers of microorganisms. However, during and after milking, it may become colonized by a variety of microbes from the teat surface, air, feed, water, milking equipment, and other environmental sources. Andualem *et al.* (2020) also stated that the microbiological quality of sheep milk is largely influenced by the method of milking, breed, housing, season, stage of lactation, and farm hygiene.

Table 11. Microbial quality of camel and sheep milk

Milk type	TVBC	Coli form	Yeast and mold
Camel milk	7.27±0.02	5.13±0.03	4.50±0.05
Sheep milk	7.42±0.03	5.27±0.01	4.62±0.05
P-value	0.0017	0.0016	0.0439

3.3.3. Acidification process of camel and sheep milk blended

Figure 3 shows the acidification process of pure camel and sheep milk as well as their blends to attain churning $\text{pH} \approx 4.25$ during fermentation for butter production. Results showed that pure camel milk resulted in slower acidification time than milk in the other treatments. With increased proportions of sheep milk in the blend, the acidification time decreases. The variation of these acidification time and pH values may be due to the unique structural and functional properties of both camel and sheep milk components.

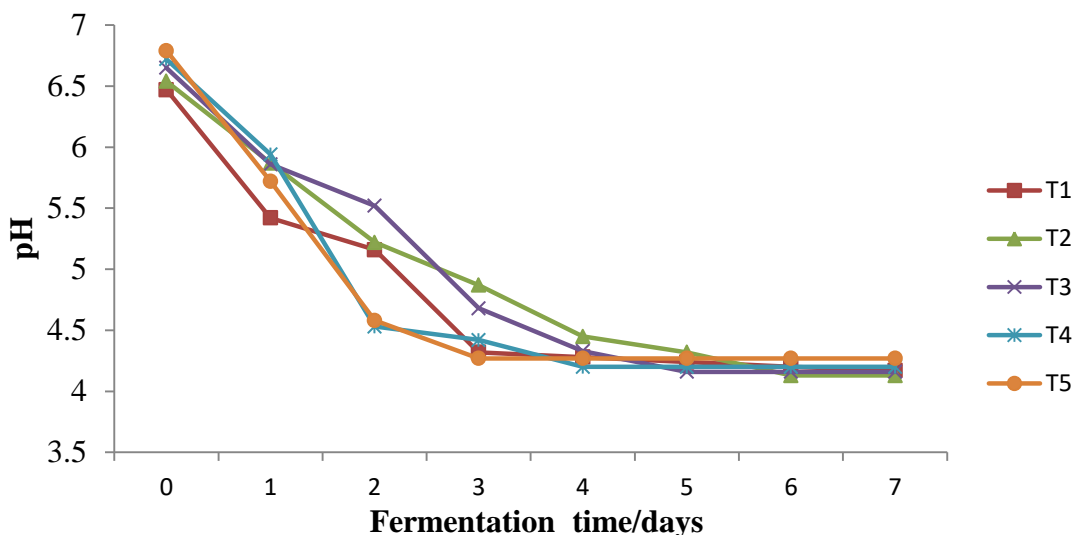


Figure 3. Acidification process of camel milk, sheep milk and their blends: T1 = 100% camel milk; T2 = 75% camel milk and 25% sheep milk; T3 = 50% camel milk and 50% sheep milk; T4 = 25% camel milk and 75% sheep milk; T5 = 100% sheep milk.

3.3.4. Physicochemical properties of butter made from camel milk, sheep milk and their blends

Physicochemical properties of butter made from camel milk, sheep milk and their blends are shown in Table 12. There were significant differences ($P < 0.05$) among treatments physicochemical properties of butter made from camel milk, sheep milk and their blends. Significant difference ($P < 0.001$) was observed in fermentation time for attaining churning pH among treatments. The fermentation time was long (7 days) for pure camel milk while it was shorter (3 days) for pure sheep milk. Higher pH value of fermented milk for churning was recorded in pure sheep milk (T5) than in milk of all other treatments. The churning time of pure camel milk (T1) was significantly ($P < 0.001$) longer than the other milk samples while the churning time of pure sheep milk was significantly ($P < 0.001$) shorter than the other milk samples. With increased proportions of sheep milk in the blend, the churning time kept on decreasing (Table 12). The reason for the different churning behavior of camel milk fat in comparison with sheep milk fat can partly be attributed to the high melting point of camel milk fat. This seems to shift the ideal ratio of solid to liquid fat in the fat globules at a given temperature towards a point higher than that of sheep milk fat.

Pure camel milk (T1) required significantly higher ($P < 0.001$) temperature for churning as compared to camel milk blended (T2, T3, and T4) and pure sheep milk butter (T4) samples, whereas the churning temperature of pure sheep milk (T5) was significantly lower ($P < 0.001$) than the other milk samples (Table 12). With increased proportions of sheep milk in the blend, the churning temperature kept on decreasing. The churning temperature observed for pure camel milk in the present study is in agreement with the finding of (Farah *et al.*, 1989) who reported the churning temperature of 20-25°C.

Butter yield from pure sheep milk (T5) was significantly higher ($p < 0.05$) than the butter yield obtained from pure camel milk and the blended milk (T1, T2, T3 and T4). Low butter yield was obtained from pure camel milk (T1) than from milk of T2, T3, T4 and T5. Results of this experiment showed that blending of

camel milk with sheep milk at any level had significant effects on butter yield in that blending gave better butter yield than pure camel milk (Table 12).

The total solids content of butter made from pure sheep milk (T5) was significantly ($P<0.001$) higher than the total solids content of butter from other milk samples (Table 12). Butter made from pure camel milk had the lowest total solids content as compared to butter made from other milk samples. With increased proportion of sheep' milk in the blend, increase in total solids content of butter was observed. The butter yield recovery from pure sheep milk (T5) was significantly higher ($p<0.001$) than recovery from other milk samples. There were significant difference ($p<0.001$) in melting point of butter among treatments. The melting point of butter samples made from pure camel milk (T1) was significantly higher than the values for butter made from milk of T3, T4 and T5. Lower melting point was recorded for butter made from pure sheep milk (T5). The melting point of butter (T5) made from pure sheep milk in the present experiment is in agreement with the finding of Morteza *et al.* (2017) who reported the values of 31.24^oC for butter from sheep milk.

Table 12. Physicochemical properties of butter made from camel milk, sheep milk and their blends

Variable	Treatment				
	T1	T2	T3	T4	T5
F (days)	7 ^a ±0.00	6 ^b ±0.00	5 ^c ±0.00	4 ^d ±0.00	3 ^e ±0.00
FMpH	4.17 ^{cb} ±0.01	4.13 ^d ±0.01	4.16 ^c ±0.02	4.20 ^b ±0.01	4.27 ^a ±0.03
Chti (min)	220 ^a ±5.00	171.67 ^b ±2.89	135.00 ^c ±5.00	58.33 ^d ±2.89	47.33 ^e ±2.52
Ch _{te} (°C)	24.50 ^a ±0.50	23.33 ^b ±0.27	22.33 ^c ±2.29	20.16 ^d ±2.28	18.16 ^e ±
Yield (4lit)	100.31 ^e ±2.34	144.31 ^d ±2.63	154.35 ^c ±2.17	179.56 ^b ±4.92	221.12 ^a ±2.70
BpH	4.85 ^a ±0.02	4.82 ^b ±0.01	4.76 ^c ±0.02	4.55 ^d ±0.02	4.57 ^d ±0.02
TS (%)	69.26 ^e ±0.40	72.31 ^d ±0.20	75.60 ^c ±1.27	78.22 ^b ±0.32	83.08 ^a ±0.41
BYRE (%)	5.02 ^e ±0.12	7.71 ^d ±0.13	7.72 ^c ±0.11	8.98 ^b ±0.24	11.05 ^a ±0.14
MR (°C)	42.67 ^a ±0.76	41.23 ^{ab} ±0.25	39.17 ^b ±0.29	37.33 ^c ±0.29	32.53 ^d ±1.86

a, b, c, d, e =Means with the same superscript letters in a row are not significantly different from each other ($P>0.05$) F= fermentation; FMpH = fermentation milk pH; Chti= churning time; Ch_{te}=churning temperature; BpH= butter pH; BYRE butter yield recovery efficiency; MR= melting point; T1 = 100% camel milk; T2 = 75% camel milk and 25% sheep milk; T3 = 50% camel milk and 50% sheep milk; T4 = 25% camel milk and 75% sheep milk; T5 = 100% sheep milk.

3.3.5. Microbial quality of butter made from camel milk, sheep milk and their blends

Mean microbial counts of butter made from camel milk, sheep milk and their blends are indicated in the Table 13. Statistically significant differences were observed in microbial quality of butter among treatments. The total viable bacteria count (TVBC) of butter made from milk of T3 was significantly higher than that of butter made from milk of T1, T2, T4 and T5. The proteolytic count (PC) of butter samples made from pure camel milk (T1) was significantly higher than PC of butter made from milk of T3, T4 T5. There was no significant ($P>0.05$) difference in PC among butter made from milk samples of T3, T4 and T5.. Statistically higher yeast and mold values were recorded in butter made from milk of T1, T2 and T3 than those butter made from milk of T4 and T5.

Table 13. Microbial count (\log_{10} cfu/g) of the butter samples made from camel milk, sheep milk and their blends (mean \pm SD)

Treatment	TVBC	Proteolytic	Coliform	Yeast and mold
T1	4.77 ^b \pm 0.09	5.15 ^a \pm 0.03	4.60 ^c \pm 0.07	5.27 ^a \pm 0.02
T2	4.81 ^b \pm 0.03	5.04 ^{ab} \pm 0.04	4.66 ^{bc} \pm 0.09	5.30 ^a \pm 0.01
T3	4.93 ^a \pm 0.02	4.96 ^{bc} \pm 0.01	4.89 ^a \pm 0.04	5.26 ^{ab} \pm 0.01
T4	4.74 ^b \pm 0.02	4.48 ^c \pm 0.76	4.80 ^{ab} \pm 0.18	5.10 ^c \pm 0.07
T5	4.76 ^b \pm 0.11	4.89 ^c \pm 0.04	4.56 ^c \pm 0.02	5.20 ^b \pm 0.01

^{a, b, c} = Means with the same superscript letters in a column are not significantly different from each other ($P > 0.05$). T1 = 100% camel milk; T2 = 75% camel milk and 25% sheep milk; T3 = 50% camel milk and 50% sheep milk; T4 = 25% camel milk and 75% sheep milk; T5 = 100% sheep milk.

4. Conclusion and Recommendations

Three experiments were conducted to analyze the effects of blending camel milk with cow, goat and sheep milk on yield and quality of butter. Camel milk contains all the essential nutrients present in cow's milk and its composition is similar to that of cow's milk except in fat and total solids. Besides, higher pH, fat, SNF, lactose, ash and total solids were recorded for goat milk than for camel milk. Similar titratable acidity and protein contents were observed for both goat and camel milk. In addition, higher titratable acidity, fat, SNF, ash and total solids were recorded for sheep milk than for camel milk. Similar pH and lactose contents were obtained from both camel and sheep milk.

Higher total viable bacteria count (TVBC) was registered for fresh camel milk than for raw cow milk. Moreover, higher coliform bacteria and yeast and mold counts were recorded for fresh camel milk than for goat raw milk. On the other hand, higher total viable bacteria, coli form bacteria and yeast and mold counts were recorded for fresh sheep milk than for fresh camel milk. In all experiments there were significant differences ($P < 0.05$) in butter yield recovery among treatments in which camel milk was blended with cow, goat and sheep milk. Results from all experiments showed that butter made from camel milk blended with cow, goat and sheep milk as well as from pure cow, goat and sheep milk gave higher butter yields than butter made from pure camel milk. The present study discovered the possibility of making butter from pure camel milk by controlling the parameters such as churning temperature and pH of milk. Blending of camel milk with cow, goat and sheep milk at any level and pure cow, goat and sheep milk gave better butter yield than pure camel milk. The shortcoming of making butter from pure camel milk was that it took very long time to churn the milk and it gave lower butter yield. Thus, research is needed to decrease the churning time of camel milk and to increase butter yield from the milk.

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Evaluation of the Effects of Formulated Concentrate Feeds on Feed Intake and Milk Yield of Lactating Upgraded Dairy Cows in Urban and Peri urban Area of Nekemte and Ijaji Towns of Western Oromia, Ethiopia

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Abstract

A study was carried out at urban and peri-urban areas of Nekemte and Ijaji towns to evaluate the effects of formulated concentrate feeds on feed intake and milk yield of upgraded dairy cows kept by smallholder dairy keepers. Twenty four lactating dairy cows within their 2nd months after calving and with similar status were selected from smallholder dairy keepers under zero grazing system whereby feed intake, BCS and milk yield data were recorded for a period of 90 days. The cows were randomly allocated to four feeding groups (T1, T2, T3 and T4) in a completely randomized design (CRD). Cows in T1, T2, T3 and T4 were fed commercial dairy feed, recommended concentrate feed of ATRC, recommended concentrate feed of HARC and recommended concentrate feed of BARC, respectively. Basal diet for all dietary treatments were natural grass hay ad libitum. The average CP (%) and IVDMD % of the natural grass hay and recommended concentrates of BARC were (8.2, 7.95) and (25.88, 10.59), respectively. The daily concentrate DM) and CP intakes were significantly different ($p < 0.001$) among the dietary treatments with highest values being registered for T4 (8.2 and 2.12 kg/d, respectively) and for T1 7.55 and 1.77kg/d respectively while the lowest values being registered for T2 (6.28 and 1.3kg/d, respectively) and for T3 (5.83 and 1.12kg/d, respectively). The daily mean milk yields were higher ($P < 0.001$) for cows in T4 (16.42 liter/day) and in T1 (15.10 liter/day) than those for those in T2 (12.55 liter/day) and in T3 (11.66 liter/day). Milk yield was also affected by location ($P < 0.001$) with the highest milk yield being obtained at Nekemte town (15.15 liter/day) and the lowest at Ijaji town (11.49 liter/day). The largest change noted in variable costs was birr 120.79 per day and the change observed in net income was birr 361.24 per day, resulting in a marginal rate of return of 76.69% for T4. Among the concentrates recommended different research centers and the commercial concentrate, concentrate recommended at BARC and the commercial ones increased milk production and profitability of the dairy enterprise. Therefore, Feeding these concentrate types for upgraded lactating cows under smallholder dairy keepers is profitable both biologically and economically

Keywords: concentrate, dairy, feed intake, and milk yield

Introduction

The livestock sector has been contributing a considerable portion to the economy of Ethiopia, and still promising to rally around the economic development of the country. Dairy farming is expanding with high yielding crossbred cows flourishing in urban and peri-urban areas of Ethiopia. Urban and peri-urban dairy production systems are becoming important suppliers of milk and milk products to urban centers (Azage et al 2013) and contributing immensely towards filling the large demand-supply gap for milk and

milk products in urban centers, where consumption of dairy products is remarkably high and they are the main suppliers of raw milk to processors of different scales (Zelalem et al 2011). However, this dairying is constrained by feed scarcity, both in terms of quantity and quality (Belay et al 2012). Reports have shown that breed improvement will lead to an improvement in milk productivity of cattle ranging from 60 to 300% if accompanied by better feeding regimes (McDermott et al 2010).

Home-mixed concentrate mixtures are blended using locally available feed ingredients in various proportions with no awareness of their quality and impact of nutrient imbalances to productive and reproductive performances of crossbred dairy cows. Nutrient concentrations in feeds vary considerably, and not all nutrients in feeds are equally available to the animal (Adugna 2008). The usual practice by dairy producers to judge quality is mainly based on visual perceptions of mixed rations without laboratory based compositional confirmation (Land O'Lakes 2010). In urban and peri-urban dairy production systems, the success of dairy production in general and crossbreeding programs in particular needs to be monitored regularly by assessing the productive and reproductive performances under the existing management, feed formulation and feeding of dairy cows.

The classical approach of increasing dairy production is through genetic means by crossing with improved breeds. Unless feeding management is improved, these animals may be limited to fully express their potential genetic superiority. It is fundamental approach to provide good quality diets to dairy cattle in sufficient amounts to maximize production. But in Ethiopia there is critical shortage of feed both in quantity and quality. The traditional feeding system for dairy cattle is based on the use of crop residues, and natural grazing supplemented with a little or no concentrates. According to Gillah *et al.* (2012), dairy farmers rarely feed concentrates at recommended levels and required quality. Thus, effective utilization of the available feed resources (agricultural and agro-industrial byproducts, natural pastures and browse) and appropriate supplementation of poor quality natural pasture and crop residue based diets appear to be the necessary step to alleviate the nutritional problems of dairy animals. Different supplementation strategies could be applied depending upon the type, accessibility and price of supplementary feeds in a given area.

Since feeding of these low protein roughages hardly support the maintenance requirements and leads to low production and reproduction of the ruminant livestock, various options to alleviate these constraints have been carried out both locally and globally among which up-grading them through supplementation with escape protein is the remarkable one. A supplement of bypass protein is the most important. Most of the oil seed plants such as noug (*Guizotia abyssinica*), linseed, groundnuts, rapeseed, sesame, cottonseed and sunflower are widely grown in Ethiopia. The cakes of these crops, after the oil is extracted are used as a protein supplement to low quality crop residues and hays. Thus, this study was initiated with the aim of evaluating the effects of formulated concentrate feeds on feed intakes and milk yield of upgraded dairy cows kept by smallholders in urban and peri-urban areas under zero grazing system.

Materials and methods

The study area

An on-farm experiment was conducted on lactating upgraded dairy cows kept by dairy keepers in urban and peri-urban areas under zero grazing system during the dry season (January 2018 to march 2018) at Nekemte (East Wellega zone) and Ijaji (West Shoa zone).

Preparation of hay and concentrate feed

Natural grass hay purchased from the local farmers around the town was used for the experiment. A concentrate mix sufficient for the entire experimental period was formulated on-station using milled Maize grain, wheat bran and Noug seed cake. This ration was formulated at BARC to fully meet the requirement for major nutrients of lactating crossbred cows with milk yield, butter fat and milk protein content as described in (NRC, 2001). Representative samples were taken for laboratory analysis and DM and nutrient content of the diets were analyzed.

Experimental animals

Twenty four farmers, having lactating upgraded cows in mid lactation (2 months after calving) were selected for the on-farm feeding trial based on their willingness to participate, commitment and presence of physical structure for monitoring feed intake. Milk yield of the cows ranged from 9.0 to 14.42kg/cow/day with an average of 11.4 ± 0.41 kg/cow/day. Based on the level of milk yield, body condition and parity, the animals were divided into four equal groups (6 cows in each group):

Experimental diets, feeding management and measurements

The four groups were fed with, dietary treatment (1) composed of Natural grass hay *ad libitum* + Commercial dairy Ration, dietary treatment (2) composed of Natural grass hay *ad libitum* + 50% wheat bran + 48% noug cake + 2% salt), dietary treatment (3) composed of Natural grass hay *ad libitum* + 74% wheat bran + 25% noug cake + 1% salt and dietary treatment (4) composed of Natural grass hay *ad libitum* + 49.5% maize grain + 49.5% noug cake + 1% salt. The supplement diets were fed at the levels required to fulfill nutrient requirements of the cows based on NRC (2001). The amounts of supplement fed to each cow in all dietary treatment during the entire experiment were at the rate of 0.5 kg per kg of milk production per day as per the previous recommendations. Adjustment of the concentrate supplement was made weekly based on the milk yield of each cow. The daily supplement allowance of each cow was divided into two equal parts and offered twice per day, in the morning and the evening milking times. . The natural grass hay was offered *ad libitum* 3 times a day by weighing the daily allowance to ensure some amount of refusals (10-15% of hay offered) next morning. Adjustment of roughage offered was made weekly based on the amount of refusal recorded every morning. All the cows were hand milked twice a day, in the morning and in the evening. Milk yield was measured daily and recorded right at milking. The selected animals were dewormed before the commencement of the experiment. The animals had free access to water throughout the experimental period.

Duration, monitoring and data recording

The data were recorded over a period of 90 days after an adaptation period of 15 days. Field visits were carried out every two weeks to monitor the feed intake and milk yield of the animals. The body condition of each cow was scored at the beginning and end of the experiment on a scale of 1 through 5. Condition score 1 indicates severe under-condition and 5 indicates severe over-condition as described by Wildman et al (1982). Cows were scored on appearance and palpation of back and hind quarters only. The

enumerators daily recorded the intake of roughage and concentrate and milk yield on pre-designed data recording sheet. These sheets were checked at each visit for accuracy and consistency. Feed samples were collected at monthly intervals and brought to the laboratory for further analysis. Perceptions of the participating farmers at the end of the experiment regarding the feasibility of different feed supplementation were assessed.

Sampling and analytical techniques

Samples of the feeds obtained during the experiment were bulked, ground and analyzed for dry matter (DM), organic matter (OM) and crude protein (CP) according to the standard procedures of AOAC (1995). Neutral detergent fiber (NDF), acid detergent fiber (ADF) and lignin were determined by the method of Van Soest (1994). The two stage *in vitro* technique developed by Tilley and Terry (1963) was used to determine *In vitro* Organic Matter Digestibility (IVOMD) of the feeds.

Partial Budget Analysis

Economic analysis was based on calculations of the total cost of production and the income from milk sales. The prices of feeds and milk were obtained from the prevailing market price in the area during the experimental period. The net profit/cow/day was calculated for the whole experimental period as a difference between the cost of production and the income generated from milk sales.

Statistical analysis

Voluntary DM and nutrient intakes, BCS and milk yield were subjected to GLM procedure for CRD using Statistical Analysis System (SAS, 2002). Treatment means were separated using Least Significant difference (LSD). The model used for the analysis of data was:

$$Y_{ij} = \mu + L_i + T_j + E_{ij}$$

Where; μ = Overall mean

C_i = Location effect (place)

T_j = Treatment effect

E_{ijk} = Experimental error

Results and Discussions

Chemical composition of feeds

The chemical compositions of feeds are shown in Table 1. The CP content of hay offered to the experimental animals in the current study was higher than the 7.02% CP reported by Abebaw (2007) and almost similar with the findings of Abuye et al., (2018) who reported 8.4% CP content. Feeds that contain lower proportion of ADF have better availability of nutrients due to ADF being negatively correlated with feed digestibility (McDonald *et al.*, 2002). The ADF value observed in the hay used in the current study was relatively comparable to the 48.3% values reported by Fentie (2007) and but higher than the 37.59% reported by Abuye et al., (2018).

The supplemental concentrates had higher CP and lower NDF concentrations relative to the basal diet. The NDF values of the supplemental feeds are lower than the 55% reported by Van Soest (1965) to limit appetite and digestibility. According to Singh and Oosting (1992), roughages with NDF content of 45-65% are generally categorized as medium quality feeds, while feeds with NDF below 45% are grouped as high quality feeds. The concentrate mix used in the present study with NDF values ranging from **39.22 - 42.53%** fall in the category of high quality feeds, The IVOMD of NGH used in the present study was less than the 59.3% IVOMD reported for native hay at BARC, and the 61.5% reported for hay harvested from Bako area (Diriba *et al.*, 2013). The difference in chemical composition might have occurred as a result of the stage of harvest.

Table 1. Chemical composition of feeds used for the experiment.

Parameter	DM (%)	Ash (%)	CP (%)	NDF (%)	ADF (%)	ADL (%)	IVDM D (%)
ATARC(Concentrate feed recommendation)	91.88	11.41	23.5	41.47	16.16	4.35	66.44
Commercial dairy ration	91.35	8.74	20.81	41.98	18.05	4.42	68.01
HARC(Concentrate feed recommendation)	90.96	8.64	19.19	42.53	14.49	2.66	69.5
BARC (Concentrate feed recommendation)	92.49	11.62	25.88	39.22	18.78	6.56	66.21
Natural grass Hay	92.61	11.26	8.2	72.6	48.9	10.87	49.7

ATARC: Adami Tulu Agricultural Research Center, HARC: Holetta Agricultural Research Center and BARC: Bako Agricultural Research Center

Dry matter and nutrients intakes

The daily DM and nutrient intake of lactating Upgraded Dairy Cows fed hay supplemented with different concentrate mix are presented in Table 2. The difference in daily DM intake was highly significant ($P < 0.001$) among treatments. The highest daily dry matter intake was observed when cows were fed dietary treatment T4 and T1. The difference could be attributed to high energy and protein concentration in these treatments, which might have enhanced the efficiency of rumen microorganisms that increased fiber degradability and digestibility thereby improving feed intake (McDonald *et al.*, 2002). Animals consume more of the feeds containing better protein as compared to those containing less protein (Steinshamn, 2010).

Intake of feed by ruminant can be improved through concentrate supplementation (Ba. *et al.*, 2015). Addition of CP supplement may stimulate efficient rumen fermentation, more passage rate and intake (Wanapat *et al.*, 2013). This implies the presence of direct relationship between CP content of feeds and feed intake. Earlier report (Ba. *et al.*, 2015) showed improvement in the daily total DM intake due to supplementation. This may be attributed to the ability of the supplements to provide nitrogen and energy for the cellulolytic microbes upon degradation in the rumen (Wambui *et al.*, 2006) and increases the nitrogen content of the total diet, which in turn is likely to increase feed intake and the rate of degradation of the basal diet in the rumen (De Almeida Rufino *et al.*, 2016). When the rate of breakdown of digesta increases, feed intake increases accordingly (Van Soest, 1982). Moyo *et al.* (2018) reported that if the

ingested feed is retained longer in the rumen, it is expected that the animal would consume less feed, because of the occupied space or 'gut fill'. The highest ($p < 0.001$) DM intake obtained for T4 and T1 might have arisen from the more balanced intakes of both CP and ME that have led to a more efficient utilization of the fiber in the total diet, which is in agreement with other studies (Huhtanen *et al.*, 2008).

The CP intake has shown significant difference ($P < 0.001$) among the dietary treatments with high values for dietary treatment T4 due to the relatively higher CP content of the feed. As far as protein requirements are concerned, the CP intake in all treatments of the present study was higher than the estimated daily CP requirement (866.5 g/d) of lactating cows producing 8-10 kg milk with 4.5% butter fat per day (ARC, 1990).

The intake of ADF and ADL were higher in T4 ($p < 0.001$) as compared to the intake in other treatments. This is likely to be due to the corresponding higher total DM consumed by the cows in that treatment. . IVDMD intake among the treatments was highly significant ($P < 0.001$). Higher IVDMD intake was observed in T1 and T4 compared to other treatments.

Table 2. DM and nutrient intakes and IVDMD of the Dairy Cow supplemented with different concentrate mix in kg/day

Treatments	Parameter						
	DM	ASH	CP	NDF	ADF	ADL	IVDMD
1	6.94 ^a ±0.49	0.86 ^b ±0.06	1.77 ^b ±0.12	3.13 ^a ±0.22	1.22 ^b ±0.09	0.33 ^b ±0.22	5.01 ^a ±0.36
2	5.73 ^b ±0.74	0.55 ^c ±0.07	1.30 ^c ±0.17	2.63 ^b ±0.33	1.13 ^b ±0.14	0.28 ^c ±0.04	4.27 ^b ±0.55
3	5.30 ^b ±0.38	0.50 ^c ±0.04	1.12 ^c ±0.08	2.48 ^b ±0.17	0.85 ^c ±0.06	0.16 ^d ±0.01	4.05 ^b ±0.29
4	7.60 ^a ±0.77	0.95 ^a ±0.09	2.12 ^a ±0.21	3.22 ^a ±0.33	1.54 ^a ±0.16	0.54 ^a ±0.06	5.44 ^a ±0.54
SE	0.25	0.028	0.062	0.11	0.05	0.015	0.18
SL	***	***	***	***	***	***	***

T1: Hay + Commercial dairy ration, T2: Hay + 48% Noug cake + 50% Wheat bran +2% salt (ATARC recommendation), T3: Hay + 25% Noug cake + 74% Wheat bran +1% salt (HARC recommendation) and T4: Hay + 49.5% Noug cake + 49.5% Maize grain +1% salt (BARC recommendation), SE: standard error, SL: significance level

Milk yield

The results of daily milk yield of Upgraded Dairy Cows fed concentrate mix are shown in Table 3. Daily milk yield was significantly different among treatments ($P < 0.0001$) being higher for cows in T4 and T1 as compared to those in the other treatments. The difference in milk yield among treatment groups is attributed to the differences in crude protein and energy contents of the diets (Steinshamn, 2010). Falk *et al.* (2018) indicated that supplemented cows produced significantly more milk than the un-supplemented ones.

In this experiment both the CP and ME intakes were sufficient to meet requirement for the observed milk yield. The mean daily milk yield obtained from cows in the present study was almost comparable to the values of 16.6 kg d⁻¹ reported by (Demski *et al.*, 2019). The variation between different reports might be due to the differences in IVDMD intake and intrinsic factors like level of production, parity, stage of

lactation, external factors like environmental stress, and unequal intervals between milking and changes in feeding.

Table 3. Milk yield and body condition score of the Dairy Cows supplemented with different concentrate mix in kg/day

Parameter	Treatments				SE	SL
	1	2	3	4		
MY(litre/day)	15.10 ^a ± 1.07	12.55 ^b ± 1.61	11.66 ^b ± 0.83	16.42 ^a ±1.66	0.55	***
BCS	3.33 ±0.52	3.5 ±0.55	3.33±0.82	3.5±0.55	0.25	ns
Concentrate(kg/d)	7.55 ^a ±0.54	6.28 ^b ±0.80	5.83 ^b ±0.42	8.20 ^a ±0.83	0.27	***
Hay intake (kg/d)	11.20±0.4	9.50±0.6	9.30±0.55	10.40±0.45	1.10	ns

T1: Hay + Commercial dairy ration, T2: Hay + 48% Noug cake + 50% Wheat bran +2% salt (ATARC recommendation), T3: Hay + 25% Noug cake + 74% Wheat bran +1% salt (HARC recommendation) and T4: Hay + 49.5% Noug cake + 49.5% Maize grain +1% salt (BARC recommendation), My: milk yield, BCS: body condition score, FI: feed intake, SE: standard error, SL: significance level.

The results of daily milk yield and feed intake of Upgraded Dairy Cows fed concentrate mix are shown in Table 4. Daily milk yield were significantly different between location ($P<0.0001$) with high milk yield for cows in Nekemte town and lower for cows in Ijaji. This is likely to be due to the fact that Nekemte town is highland and convenient for dairy rearing as compared to Ijaji which is somewhat hot area. The daily feed intake was also high in Nekemte town as compared to in Ijaji town ($P<0.0001$) because the cows feed intake was high in highland as compared to in midland.

Table 4. Effect of location on Feed intake, milk yield and body condition score of the Dairy Cows supplemented with different concentrate mix

Parameter	Location		SE	SL
	Nekemte	Ijaji		
MY (Litre/day)	15.15 ^a ± 1.13	11.49 ^b ± 1.70	0.47	***
BSC	3.5 ± 0.52	3.25 ± 0.71	0.18	ns
FI (kg/day)	7.58 ^a ± 0.85	5.75 ^b ± 0.56	0.23	***

MY: milk yield, BCS: body condition score, FI: feed intake, SE: standard error, SL: significance level
Table 3. Effect of concentrate feed intake on milk yield and body condition of Upgraded Dairy Cows at Nekemte and Ijaji town

Partial budget analysis

The economic feasibility of this study was analyzed using partial budget analysis. According to this analysis, T4 gave higher net benefit (Birr 240.45 per cow/day), than other treatments. The minimum rate of return acceptable by the dairy farmer was assumed to be 50% (CIMMYT, 1985). This implies that the dairy farmer expects a minimum rate of return of 50% if he is to adopt a new practice as compared to the practice he used to do. Change in cost that varies was birr 28.04 per day and the change in net income was birr 76.68 per day resulting in 273.47% marginal rate of return for T4. So, for each birr invested as input

for a cow, the farmer will recover birr 1.00 and an additional birr 2.73 at a given prices. Therefore, on the basis of MRR the technology is recommended for increasing milk productivity of cows. The result of MRR of the present study was in the profitable range of 158% and 131.85% reported by Shah et al. (2009) for milking cows and buffaloes, respectively. . Therefore, considering milk yield and economic return in this study, it can be concluded that cows fed with concentrate diet of T4 with *adlibitum* hay optimize both biological and economic benefits as compared to cows consumed with other treatment feeds (Table 5).

Table 5. Partial budget analysis for lactating Upgraded Dairy Cows fed natural grass hay as basaldiet supplemented with concentrate mix at the rate of 0.5kg/liter of milk.

Variable	Cost per unit	Treatments			
	Benefits (ETB/cow/d)	T1	T2	T3	T4
Milkyield(kg/cow/d)	-	15.10	12.55	11.66	16.42
Grossfield benefit(ETB/cow/d /cow/day)	-	332.20	276.10	256.52	361.24
Hay intake (kg/cow/day)	-	11.20	9.50	9.30	10.40
Costofhay(ETB/ kg/cow/day)	7/kg	78.40	66.50	65.10	72.80
concentratemix intake(kg/cow/day) (kg/cow/d)(ETB/kg/cow/day)	-	7.55	6.28	5.83	8.20
Cost for concentrate (ETB/ kg)	-	7.70	7.06	6.40	7.03
Noug cake intake (kg/cow/d)	-	-	3.39	1.60	3.51
Cost for Noug cake(kg/cow/d)	6.99/kg	-	23.69	11.18	24.52
Wheat bran intake (kg/cow/d)	-	-	3.53	4.74	-
Cost for wheat bran(kg/cow/d)	4.8/kg	-	16.94	22.73	-
maize grain intake (kg/cow/d)	-	-	-	-	3.51
Cost for maize grain (kg/cow/d)	5/kg	-	-	-	17.54
Salt intake (kg/cow/d)	-	-	0.14	0.06	0.07
Cost of Salt (kg/cow/d)	10/kg	-	1.4	0.6	0.7
Total cost for concentrate in take cost per	-	58.14	44.34	37.31	57.65
Costoftablet,salt andlabour (ETB /cow/day)	-	110.00	120.00	110.00	110.00
Totalvariablecost(ETB /cow/day)	-	246.54	230.84	212.41	240.45
Grossincome,ETB/head	-	332.20	276.10	256.52	361.24
Net benefit(ETB cow/day)	-	85.66	45.26	44.11	120.79
Changeinnetincome(ETB)	-	41.55	1.15	0	76.68
Changeintotalvariablecost(ETB	-	34.13	18.43	0	28.04
MRR%	-	121.74	6.24	0	273.47

Ethiopian Birr; MRR = Marginal Rate of Return;

T1: Hay + Commercial dairy ration, T2: Hay + 48% Noug cake + 50% Wheat bran +2% salt (ATARC recommendation), T3: Hay + 25% Noug cake + 74% Wheat bran +1% salt (HARC recommendation) and T4: Hay + 49.5% Noug cake + 49.5% Maize grain +1% salt (BARC recommendation)

Dairy keepers' perception

Participants noticed that the benefits of feeding the intervention diet were immediately visible. Dairy keepers noticed that cows in T₁ and T₂ eat the concentrate feed more quickly as compared to cows in the other treatments. Furthermore, after feeding the intervention diet, an increase in milk production and an improvement in fertility levels (cows show more intensely the characteristic of being on heat) were noted.

Conclusion and recommendations

The results of the present study lead to acceptance of the fact that using feed of high protein and energy sources can increase milk yield of upgraded dairy cows. Upgraded lactating cows under dairy keepers at Nekemte and Ijaji towns are profitable both biologically and economically. Among the different recommended concentrate at different research centers and a commercial ration, BARC concentrate recommendation and the commercial ration increased milk production and profitability of the dairy enterprise. These feeds are then recommended as supplemental feeds for crossbred dairy cows.

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Comparative Evaluation of Urea and Effective Microbes Treated Finger Millet Straw on Feed Intake, Milk Yield and Composition of Lactating Crossbred Dairy Cows

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Abstract

The experiment was conducted at Bako agricultural research center to evaluate the effect of EM2 and urea treated finger millet straw supplemented with concentrate mix on feed intake, milk yield and composition of crossbred dairy cows. Four cows of same milk yield, body weight, stage of lactation, but differing parities were arranged in 4x4 Latin square design. The animals were provided with natural grass hay (T1), untreated finger millet straw (T2), EM2 treated finger millet straw (T3) and urea treated finger millet straw (T4) diet ad libitum and all treatments were supplemented with concentrate mix. Results of chemical analysis of the treated finger millet straw showed that the treated straw had good nutritive value. The daily dry matter (DM) and crude protein (CP) intakes were significantly ($P < 0.001$) different among the treatments with the highest intake observed for cows fed EM2 and urea treated finger millet straw (T3 and T4). Milk yields varied significantly among the dietary treatments with the lower mean milk yield recorded for cows in T1 and T2 as compared to those in T3 and T4. This study indicated that EM2 and urea treated finger millet straw diet increased the net return. Feeding EM2 and urea treated finger millet straw with concentrate mix was found to be an effective approach to maximize the utilization of locally available feed resources for relatively high animal productivity during the dry season for small scale dairy keepers in rural areas. Therefore, the result demonstrated that EM2 and urea treated finger millet straw had better feeding value as compared to untreated finger millet straw and natural grass hay for lactating crossbred dairy cows.

Keywords: Crossbred, Effective microbe, Finger millet straw, Natural grass hay and Urea

Introduction

Feed represents the largest single expense among the inputs for livestock production. Livestock farmers search for inexpensive feed alternatives, especially when conventional feeds are expensive. Many of these alternative feeds are by-products and waste products from the processing of various food and fiber crops, or crop residues, tree leaves, farm animal wastes etc. There is a need to explore the possibility of utilizing novel feed stuffs, agricultural crop residues, and agro-industrial by-products as complete feed allowance in comprehensive feeding scheme to reduce the feed deficiency and to economize the production (Sudheer Babu *et al.* 2013). One of such usable crop residues as ruminant animal feed is finger millet straw.

Finger millet straw (FMS) consists of dry stems and leaves. FMS is the by-product obtained after harvesting the crop and can be used as ruminant feed as source of roughage (Malisetty *et al.* 2013).

The straw is available after harvesting, threshing and collecting the grains for human consumption. FMS is considered to have a nutritive value better than other cereals straw (Subba Rao *et al.* 1995). However, still it needs treatment to be available nutrient for better nutritive value therefore it must be supplemented with nitrogen and energy sources to meet maintenance and production requirements (Heuze and Trans, 2013). Finger millet straw is readily available especially during the dry season, after the year's harvest. It is cheaper to cure and preserved. Consequently, it could be fed to ruminant animals as a basal feed.

Nutrient deficiencies affect microbial growth, microbial protein synthesis and overall fermentation in the rumen that further result in low voluntary intake and fiber fermentation and digestibility (Mahesh and Madhu, 2013). The barrier can be removed through biological treatment that increase digestibility by decreasing strength of bonds between lignin and polysaccharides (Russell *et al.*, 2011 and Peterson, 2014). Biological treatment of crop residues based on the use of enzyme or microbes have shown to improve palatability and degradability potential and keeping quality of the feed material (Milligan *et al.*, 1995). Feeding effective microbes (EM2) treated hay supplemented with escape protein resulted in remarkable increase in feed efficiency of stationed lactating dairy cows (Mulugeta, 2015). Biological treatment of maize stover based on the use of enzyme or microbes have shown to improve the quality of feed materials by increasing their palatability and degradability (Milligan *et al.*, 1995). Therefore, this study was aimed to evaluate the effect of feeding EM2 and urea treated finger millet straw supplemented with concentrate mix on feed intake, milk yield and composition of crossbred dairy cattle.

Materials and Methods

Study area

The experiment was carried out at Bako Agricultural Research Center, Ethiopia. The center is located at longitude 37° 09' E, latitude 09° 06' N, an altitude of 1650 m above sea level and at 260 km west of the capital city, Addis Ababa.

Experimental animals

Experimental cows with similar lactation performance, same stage of lactation, body weight, but different parity were selected from the total dairy herd available in Bako Agricultural Research Center. All experimental cows were weighed and de-wormed before starting the experiment.

Preparation of experimental feeds

Rhodes grass hay and finger millet straw were used as a basal diet throughout the experimental period. The supplemental concentrate mixture was composed of 49.5% maize grain + 49.5% noug seed cake + 1% salt. The feed supplement was weighed and offered in a separate feed trough twice a day at 8:00 AM and 6:00 PM. The basal diet was fed ad libitum and adjusted up to 20% refusal level. Feed refusals were collected and weighed before the next feeding. Samples of feed offer and refusals were separately taken and bulked over 30 days for feed intake analysis. Feed intake was calculated as difference of the quantity of feed offered and feed refused. The animals had free access to clean water throughout the experimental periods.

The experimental feeds ingredients used in this study were finger millet straw, natural grass hay, maize grain, Noug seed cake, effective micro-organism (EM2), urea and salt. The Stock EM solution (EM1) used for this study was purchased from the recognized distributor (Woljejjii PLC, Debrie-Zeit, Ethiopia). Finger millet straw was used as basal feed both in treated and untreated forms and natural grass hay was also used as untreated form. The straw was manually collected from farms and stored under shade. A 20 liter of EM2 solution was prepared by mixing 1 liter EM1 (stock solution) + 1 liter molasses + 18 liter water. Then the mixture was stored in a closed plastic container of 25 liter's capacity for 30 days to activate the stock solution. After 30 days a liter of activated EM2 solution was extra diluted by 20 liters of water and sprayed thoroughly on chopped finger millet straw of 20kg (on dry matter basis) put in a polyethylene sheet of 100m² (10m*10m). This EM2 treated finger millet straw was ensiled for 30 days after which it was used for the lactating crossbred dairy cattle.

Regarding the urea treatment, as in the case of EM2 treatment, the finger millet straw was chopped into the size of 2-3 cm prior to the ensiling process. Then 5 kg of urea was dissolved in 100 liters of water and sprinkled uniformly over the 100 kg of finger millet straw by using sprinkler and buckets. The treated finger millet straw was mixed by using a fork. All mixtures were firmly packed in a silo by trampling to remove air and finally the silo was sealed. The treated finger millet straw was ensiled for 30 days after which it was used for the experimental animals.

The amount of concentrate mix offered daily was at the rate of 0.5 kg/l of milk produced by each cow and it was offered with equal portions at 05:00 and 17:00 hours during the morning and evening milking. Adjustments for concentrate offer was made at the end of each period and for each treatment based on the actual milk produced. Adjustment of the basal feed was made weekly based on the amount of refusal weighed every morning, voluntary intake and milk yield of each cow.

Experimental design

Four lactating crossbred cows were randomly assigned in a switch over 4X4 Latin square design. There were four periods each consisting of 30 days. During the first 15 days of each period, animals were acclimated to the experimental diet and the remaining 15 days were used to collect data. Hence, the experiment took a total of 120 days.

Experimental treatments were

- T1: Natural grass hay + Concentrate mix (0.5 kg/l of milk)
- T2: Untreated finger millet straw+ Concentrate mix (0.5 kg/l of milk)
- T3: EM2 treated finger millet straw+ Concentrate mix (0.5 kg/l of milk)
- T4: Urea treated finger millet straw + Concentrate mix (0.5 kg/l of milk)

Measurements

The daily milk yield data of individual cows was taken using a Salter balance. About 100 ml milk sample in the morning and afternoon was taken twice every week during the experiment from each cow into a glass measuring cylinder (100ml capacity) after the milk was thoroughly and gently mixed. Body weight of the animals in each treatment was recorded for two consecutive days at the beginning and end of each experimental period to monitor body weight change that may occur as a

result of dietary treatments. All samples of feed offered and refusals and faeces were analyzed for DM, ash, N (Kjeldahl-N) according to AOAC (1990). Organic matter (OM) was determined as 100-ash. Crude fiber (CF) and ether extract (EE) was determined by proximate analysis. Neutral detergent fiber (NDF), acid detergent fiber (ADF), and acid detergent lignin (ADL) were determined by the methods of Van Soest and Robertson (1985). *In vitro* organic matter digestibility of feed offered and refusal was determined using the procedures outlined by Tilley and Terry (1963). The milk samples were used to determine percentage fat, protein and solid not fat (SNF) by Ultrasonic Ekomilk Analyzer (30 w Bulteh 2000, Bulgaria), which have the capacity to measure 20 – 25 samples per hour. Total milk solids (TS) were calculated as TS = SNF+Fat. Calcium and phosphorous content of the offered feeds were analyzed by atomic absorption spectrophotometry and calorimetry (AOAC, 1995) respectively.

Statistical analysis

Voluntary DM and nutrient intakes, live weight change, milk yield and compositions were subjected to GLM procedure for Latin Square Design using Statistical Analysis System (SAS, 2009). Treatment means were separated using Least Significant difference (LSD). The models used for the analysis of data were:

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

Where; μ = Overall mean, C_i = Cow effect (parity), P_j = Period effect, T_k = Treatment effect and E_{ijk} = Experimental error

Results and Discussions

Chemical composition of experimental feeds

The chemical composition of natural grass hay, untreated finger millet straw, treated finger millet straw and concentrate mixture are presented in the Table (1). The NDF, ADF, and ADL contents of natural grass hay (NGH) used in this study were higher than that of untreated finger millet straw (UFMS), Urea treated finger millet straw (UTFMS) and Effective microbes treated finger millet straw (EMTFMS). The CP contents were high in UTFMS than EMTFMS and, UFMS. The CP content of concentrate was higher than that of NGH, UTFMS, EMTFMS and UFMS.

The CP content of hay offered to the experimental animals in the current study was comparable 5.1 and 5.6% of CP reported by Ewnetu (1999) and Getachew (2005), respectively while the CP reported by Abebaw (2007) was 7.02 which is above the current study. It has been stated that CP value ranging from 7-7.5% is required to satisfy maintenance requirement of ruminant animals (Van Soest, 1982). Hence, the observed CP content of NGH in the current study was below demanded for maintenance requirements of dairy cattle. The CP content of finger millet straw in the current study was higher than the value reported by Umashankar B. C. (2011), 3.50 % CP and the NDF and ADF contents were 67.05 and 44.00%, respectively which is comparable with the current study.

Table 1. Chemical composition of experimental feeds offered to lactating crossbred dairy cows

Parameter	DM	ASH	OM	CP	NDF	ADF	ADL	IVOMD	ME	Ca	p
UFMS	90.30	10.50	89.50	4.90	69.20	43.30	18.60	52.10	7.10	1.23	0.18
UTFMS	73.50	9.90	90.10	10.10	67.80	41.00	17.20	59.50	7.60	1.28	0.19
EMTFMS	74.20	9.40	90.60	6.30	66.50	40.00	16.80	58.30	7.90	1.31	0.22
NGH	92.50	11.00	89.00	5.50	71.10	46.80	6.33	48.60	6.90	1.13	0.14
concentrate	89.70	6.20	93.80	21.50	33.30	16.20	3.10	71.20	12.90	0.17	0.96

UFMS: Untreated finger millet straw, UTFMS: Urea treated finger millet straw, EMTFMS: Effective microbes treated finger millet straw, NGH: Natural grass hay, ADF: Acid detergent fiber, CP: Crude protein, DM: Dry matter, NDF: Neutral detergent fiber, OM: Organic matter, ADL: Acid detergent lignin, IVOMD: Invitro organic matter digestibility, ME: Metabolizable energy

Feed intake

The mean daily DM, CP, OM, NDF ADF and ADL intake of lactating crossbred dairy cows fed EM2 and urea treated Finger millet straw and supplemented with concentrate mix are presented in Table 2. The daily DM, CP, OM, NDF, ADF and ADL intake were significant ($P<0.001$) among treatments. The highest daily dry matter intake was observed for cows fed with EM2 and urea treated finger millet straw (T3 and T4) as a basal diet. The difference could be attributed to the high rumen degradable protein content of the EM2 and urea treated finger millet compared to the untreated one and the natural grass hay. Such treatment of straws enhances the efficiency of rumen microorganisms to increase fiber degradability and digestibility thereby improving feed intake (McDonald *et al.*, 2002). The low CP and high fiber contents of the untreated finger millet straw and natural grass hay are likely to depress both feed intake and digestibility. NDF is negatively correlated with feed intake and its content above 55% can limit DM intake (Arelovich *et al.*, 2008). The intake of feeds by ruminants can be improved through concentrate supplementation (Gatenby, 2002). Protein meal supplements stimulate intake of low quality roughage diets by providing rumen degradable protein that are deficient in the roughage (Mohini, 2013). Earlier report of Mulu (2005) showed improvement in the daily total DM intake due to supplementation. This may be attributed to the ability of the supplements to provide nitrogen and energy for the cellulolytic microbes upon degradation in the rumen (Wambui *et al.*, 2006).

Table 2. Nutrient intake of experimental treatment of lactating crossbred dairy cows in kg/day.

Parameter	Treatments					
	1	2	3	4	Se	Sl
TDM	8.23 ^b	8.09 ^b	9.50 ^a	10.15 ^a	0.233	***
OM	7.39 ^b	7.21 ^b	8.60 ^a	9.14 ^a	0.209	***
CP	1.01 ^{bc}	0.87 ^c	1.16 ^b	1.52 ^a	0.049	***
NDF	4.69 ^c	4.59 ^c	5.16 ^b	5.52 ^a	0.088	***
ADF	2.93 ^b	2.72 ^c	3.01 ^{ab}	3.18 ^a	0.049	***
ADL	0.60 ^c	1.51 ^b	1.60 ^b	1.76 ^a	0.041	***

T1: Natural grass hay + Concentrate mix (0.5 kg/l of milk), **T2:** Untreated finger millet straw + Concentrate mix (0.5 kg/l of milk), **T3:** EM2 treated finger millet straw + Concentrate mix (0.5 kg/l of milk), **T4:** Urea treated finger millet straw + Concentrate mix (0.5 kg/l of milk), *Se:* standard error; **ns:**

non significance , **SL**: significance level, **ADF**: Acid detergent fiber, **CP**: Crude protein, **TDM**: Total dry matter, **NDF**: Neutral detergent fiber, **OM**: Organic matter and **ADL**: Acid detergent lignin.

Milk yield and composition

Milk yield and composition of the cows fed experimental feeds are given in Table 3. Daily milk yield was significantly different ($P < 0.05$) among treatments with higher values for those cows in T3 and T4 as compared to those in T1 and T2. The difference in milk yield among the treatment groups is attributed to the differences in crude protein and energy contents of the diets

(Steinshamn, 2010); Getu (2006) indicated that lactating crossbred dairy cows fed urea treated wheat straw basal diet produced significantly higher milk yield when supplemented with 50% vetch (*Vicia dasycarpa*) diet than the non-supplemented ones because of better nutrient supply. Milk protein, milk fat, solid not fat and total solid contents were non-significant ($P > 0.05$) among dietary treatments. These findings are similar to work done by Gusha *et al.* (2014) where urea and protein supplementation did not alter milk composition. The variation between different reports might be due to the differences in metabolizable energy intake and intrinsic factors like level of production, parity, stage of lactation, external factors like environmental stress, and due to unequal intervals between milking and changes in feeding.

Table 3. Effect of EM2 and urea treated finger millet straw on milk yield, composition and body weight change of lactating crossbred dairy cows.

Parameters	Treatments				Se	SL
	1	2	3	4		
Milk yield (littre/day)	6.58 ^b	6.38 ^b	8.10 ^a	7.90 ^a	0.38	*
Milk fat (%)	4.08	4.10	4.13	4.40	0.10	ns
Milk Protein (%)	3.48	3.45	3.63	3.63	0.08	ns
Solid not fat (%)	8.28	8.33	8.44	8.55	0.13	ns
Total solid (%)	12.30	11.90	13.10	12.93	0.37	ns
Ash	0.78	0.78	0.79	0.78	0.03	ns
Body weight Change (g/day)	313.90	416.58	229.15	229.15	75,52	ns

T1: Natural grass hay + Concentrate mix (0.5 kg/l of milk), **T2**: Untreated finger millet straw + Concentrate mix (0.5 kg/l of milk), **T3**: EM2 treated finger millet straw + Concentrate mix (0.5 kg/l of milk), **T4**: Urea treated finger millet straw + Concentrate mix (0.5kg/l of milk), **Se**: standard error, **ns**: non significance and **SL**: significance level

Daily body weight change

The daily mean live weight changes of crossbred dairy cows fed EM and urea treated finger millet straw are shown in Table 3. The mean daily live weight gain was non-significant ($P > 0.05$) among the dietary treatments. The presence of marked differences in nutrient intake among the dietary treatments did not bring a significant effect in weight change of the cows, which may be due to the utilization of additional nutrients consumed for milk production than for weight gain. However body weight loss of 120 g/day was reported for lactating crossbred cows by Getu (2006). Cows lost body weight after the first phase of the lactation cycle, but with a declining trend. However, improvements in body

weight condition of cows have also been observed for all dietary treatments except during the first period of the experiment. This could be probably associated with more diversion of the available nutrients to body tissue accretion.

Partial Budget Analysis

The economic feasibility of this study was analyzed using partial budget analysis (Table 5). According to this analysis, T3 and T4 gave the highest net benefit (Birr 121.15 and 115.35 per cow/day), while T1 gave the lowest net benefit (Birr 91.32 per cow/day). The minimum rate of return acceptable by a dairy farmer was assumed to be 50% (CIMMYT, 1985). This implies that the dairy farmer expects a minimum rate of return of 50% if he is to adopt a new practice as compared to the practice he used to do. The result of the present study is a little far from the report of CIMMYT (1988) which proposed a minimum rate of income of twice the cost of capital as a relevant measure for investments of capital in new technologies. Alternatively, especially for poor farmers in developing countries or for technologies requiring substantial change to a farming system, a minimum rate of income of 100% (the 2-for-1 rule) was likely to be more relevant. Farquharson (2006) has also supported this criterion.

Marginal rate of return indicates what farmers can expect to gain on the average in return from their investment when they decide to change from one practice to another. Among the treatment in this study, the largest change in cost that varies was birr 10.15 per day and the corresponding change in net benefits was birr 26.33 per day for T4 resulting in marginal rate of return of 259%.. So for each birr invested in input for a cow, the farmer would recover birr 1(one) and an additional birr 2.59 at a given prices. Therefore, on the basis of MRR the technology is highly recommended for increasing milk production of the cows, as the returns were much higher than minimum acceptable rate of return (100%) for any technology to be recommended. The result of MRR of the present study was comparable with the MRR of 158% and 131.85% reported, respectively for milking cows and buffaloes fed on urea mineral molasses blocks under an on-farm condition (Shah *et al.*, 2009).

Table 4. Partial budget analysis for lactating crossbred dairy cows fed EM2 and Urea treated Finger millet straw basal diet and supplemented with concentrate mix (0.5kg/kg milk).

Variable	Treatments			
	1	2	3	4
Milk yield (kg/cow/d)	6.58	6.38	8.10	7.9
Gross field benefit (ETB /cow/d)	157.92	153.12	194.4	189.6
Cost of NHG (ETB/ kg /cow/d)	30	-	-	-
Cost of Urea (ETB/ kg /cow/d)	-	-	5.00	-
Cost of EM2 (ETB/ kg /cow/d)	-	-	-	6.50
Cost of Finger millet straw (ETB/ kg /cow/d)	-	28.00	28.00	28.00
Cost for Concentrate mix (ETB/kg/cow/d)	16.45	15.95	20.25	19.75
Cost of Tablet, Mineral and labour (ETB /cow/d)	20.15	20.15	20.00	20.00
Total variable cost (ETB /cow/d)	66.6	64.1	73.25	74.25
Gross income, ETB/head	157.92	153.12	194.40	189.60
Net benefit (ETB cow/d)	91.32	89.02	121.15	115.35
Change in net income	2.30	-	32.13	26.33
Change in total variable cost	2.50	-	9.15	10.15
MRR, %	92	-	351	259

ETB : Ethiopian Birr, MRR : Marginal rate of return, T1: Natural grass hay + Concentrate mix (0.5 kg/l of milk), T2: Untreated treated finger millet straw + Concentrate mix (0.5 kg/l of milk), T3: EM2 treated finger millet straw + Concentrate mix (0.5 kg/l of milk) and T4: Urea treated finger millet straw + Concentrate mix (0.5 kg/l of milk)

Conclusion and Recommendation

Substantial increase in milk production per animal per day and net benefit derived from the increased milk produced indicated that the use of EM2 and urea treated finger millet straw diet is a sound technology for crossbred dairy animals under small scale farmer's condition. Thus it is possible to substantially improve the productivity of crossbred dairy cows in similar production systems by feeding EM2 and urea treated finger millet straw and supplementing concentrate mix.

A rational dairy farmer has to make a compromised decision so that he could opt for a more sustainable milk yield and reasonable profit throughout the entire lactation period, although, this study emphasizes the importance of additional observations to see the likelihood of lactation curve for all dietary treatments in the remaining part of the lactation cycle for conclusive economic decision. Generally, those cows fed basal diet of treated finger millet straw with recommended concentrate mix optimize both biological and economic benefits as compared to cows consumed other treatment rations.

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Meat Animals Research Results

Growth performance and carcass characteristics of Horro Sheep under grain-less feed regime: wheat bran as substitute for maize grain

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Abstract

A total of 27 Horro rams from Bako Agricultural Research Center were used with the main objective of evaluating growth performance and carcass characteristics of Horro rams supplemented with Wheat bran – Noug Cake concentrate. The rams were randomly assigned to three different treatments based on their initial liveweight. The three treatments were: T1= Rhodes grass hay ad lib + Concentrate (69.5% Wheat bran + 29.5% Noug cake + 1% Salt), T2= Rhodes grass hay ad lib + Concentrate (55.5% Wheat bran + 33.5% Noug cake + 10% Maize + 1% Salt), and T3= Rhodes grass hay ad lib + Concentrate (49.5% Noug cake + 49.5% Maize grain + 1% Salt). There was a significant variation in final body weight between T1 and T3, T3 showing a higher final body weight. However, the Least Square Difference of Means (LSD) showed that, there was no significant difference in final body weight between rams in T2 and T3. Similarly, there was no significant difference in final body weight between rams in T1 and T2. The results of this study have showed that total substitution of maize grain with wheat bran resulted in a lower final body weight and average daily weight gain. On the contrary, as a result of variations in price of the concentrate mixtures, the net return value was not significantly varied. So, from the control concentrate mixture which is 49.5% Noug cake + 49.5% Maize grain + 1% Salt, 40% of noug cake and the whole maize grain could be substituted with wheat bran without affecting the crude protein content. Thus, a concentrate mixture of T1 (69.5% Wheat bran + 29.5% Noug cake + 1% Salt) can be used instead of T3 (49.5% Noug cake + 49.5% Maize grain + 1% Salt).

Key words: Growth performance, grain-less, wheat bran, Horro sheep.

Introduction

In Ethiopia, sheep producers target their lambs to attain slaughter weight in a short period of time with the maximum amount of lean meat, minimum bone and an amount of fat which is desired by the consumers (Aschalew and Getachew, 2013). However, FAO (1997) reported that mean carcass weight of sheep is less than 10 kg per animal, which is the second lowest in sub-Saharan African. Gemeda et al. (2007) and Tesfa et al. (2013) suggested that, among many factors, plane of nutrition plays a major role in contributing to the variation in growth performance, carcass weight and carcass compositions in sheep. Temesgen et al. (2007) noted that sheep productivity is based on large flock number, which is not viable option due to growing human population. Instead, intensified feeding may be one way of raising production per unit of animal in a sustainable way, on the basis of utilizing available feed resources as suggested by Shapiro et al. (2004).

The major feed resources in Ethiopia are native pasture, crop residues and agro-industrial by-products. The native pasture, however, is characterized by high seasonal variation in yield and quality and animals often lose condition during the dry season. Grains are expensive and economically not suitable to use as a supplement in animal nutrition. The challenge is to develop alternative feed resources that will sustain production throughout the year.

In feeding systems where straws and hay are the basic diet for ruminants, the low intake of these roughages requires supplementation to meet the requirement for production. The rate of growth and milk production by ruminants grazing tropical pastures and depending on crop residues or grass hay alone are generally low and about 10% of the animals genetic potential (Leng, 1997). This implies that strategic supplementation of energy; protein and minerals are important means of ensuring better animal performances. The aim of supplementation for ruminants feeding system is to alleviate nutritional deficiencies in the basal diet to maintain or increase intake of the basal diet (McMeniman *et al.*, 1988). When ruminants are offered un-supplemented low quality roughage, they lose weight because of their inability to meet both energy and protein requirements (Nsahlai, 1991). Therefore, supplementation with nitrogen either as protein or non-protein nitrogen and energy has been shown to improve animal performances, mainly through increasing DM digestibility, intakes and balances of nutrients (Preston and Leng, 1987). Rates of fermentation of fibrous crop residues can often be improved by supplementing small amount of highly digestible protein and energy nutrients such as agro-industrial by-products (Leng and Preston, 1983).

Therefore, this study was intended with the general objective of evaluating growth performance and carcass characteristics of Horro rams fed grain-less supplemental feed and determining the economic feasibility of fattening with those non-grain based concentrates.

Specific objectives:

1. To evaluate growth performance and carcass traits of Horro rams supplemented with wheat bran-noug cake concentrate
2. To identify profitability of wheat bran-noug cake concentrate as compared to maize grain-noug cake concentrate

MATERIALS AND METHODS

Study site

The study was conducted in Bako Agricultural Research Centre which is located at about 250 km from Addis Ababa on the main road to Nekemte. The area is situated at an altitude of 1650 m.a.s.l. and receives mean annual rainfall of 1200 mm in a bimodal distribution, 80% of which falls from May to September. Bako area had a mean relative humidity of 60% and mean minimum and maximum temperatures of 13.5°C and 27°C, respectively.

Animals and experimental design

A total of twenty seven yearling Horro rams with mean initial body weight of 24.9 ± 0.21 kg, with a range of 20.0 ± 0.45 kg to 29.8 ± 0.32 kg have been used for this study. The experiment was laid out in completely randomized design (CRD) with three treatment groups. All the twenty seven animals were

sorted in ascending initial body weight and then stratified to three weight groups each consisting of nine animals. The randomization was done by randomly assigning the nine animals in the first strata to the three treatment groups, thus each treatment received three animals from the first strata and the same was done for the second and the third strata. Finally, the three treatment groups had nine animals each. All the rams were housed in individual pens throughout the experimental period.

The three dietary treatments were:

T1= Rhodes grass hay *ad lib* + (69.5% Wheat bran + 29.5% Noug cake + 1% Salt),

T2= Rhodes grass hay *ad lib* + (55.5% Wheat bran + 33.5% Noug cake + 10% Maize + 1% Salt),

T3= Rhodes grass hay *ad lib* + (49.5% Noug cake + 49.5% Maize grain + 1% Salt).

Each animal received 400g per day of their respective feed for 90 days with adaptation period of two weeks.

Carcass analysis

At the end of the 90 days experiment, all sheep were fasted overnight, weighed and slaughtered. Different components of the carcass were separated, weighed and recorded for each sheep.

The rib eye muscle area was traced between the twelfth and thirteenth rib and the area was measured with traced square paper. Empty BW was calculated as slaughter weight less gut content. Dressing percent was calculated as proportion of hot carcass weight to slaughter weight and/or empty BW. Percent of total edible offal (TEO) was calculated as the sum of blood, lung, trachea, heart, liver, spleen, empty gut, kidney and internal fat (mesenteric and kidney) weight to slaughter weight. Also percent of total non-edible offal (TNEO) was calculated as the sum of head, skin, genital organs, gall bladder and gut fill weight to slaughter weight

Data analysis

The GLM procedure of SAS was used to analyse all the data. Treatment means were separated by least significant difference test (LSD).

The model fitted to compute the responses was:

$$y_i = \mu + t_i + e_i$$

Where; y_i = response variable,

μ = overall mean effect

t_i = i^{th} effect of the three feed treatments

e_i = i^{th} effect of random error

Results and discussions

Body weight change

The mean final body weights for T1, T2 and T3 were 31.89 ± 2.89 , 33.17 ± 2.89 and 34.89 ± 2.89 , respectively. Higher final body weight ($P < 0.05$) was observed for rams under T3 as shown in Table 1. However, there was no significant variation among rams in T2 and T3 in final body weight. Even though

rams in T1 had showed the lowest final body weight, there was no significant ($P<0.05$) variation with rams in T2. Similarly, there were significant ($P<0.001$) variations among all rams in T1, T2 and T3 both in body weight change and average daily gains. According to this study, total substitution of maize grain with wheat bran in the concentrate mixture had significantly ($P<0.001$) affected the final body weight. However, graded substitution resulted in a gradual decline in body weight gain as compared to the control feed (T3, which is mixture of maize grain and noug seed cake). This might be due to the fact that formulation of the three rations was on the basis of CP content of the ingredients in such a way that the feeds are Iso-nitrogenous. Hence the variation in final body weight has resulted mainly from the differences in energy content of the two ingredients (maize grain and wheat bran). Moharrery *et al.* (2012) and Yagoub and Babiker (2008) also reported that daily body weight gain improved at a higher dietary energy than at a lower energy level. This study is in agreement with, supplementation of wheat bran in rice straw based feeding of entire bulls (Chowdhury, 1996), and supplementation of wheat bran in mixture with groundnut cake to pregnant does (Getnet *et al.*, 1998) in which substantial body weight gains were reported.

The above result showed that as far as the objective of this study is mainly directed towards substituting maize grain in concentrate feeds and the observed variations are not as such strong ($P<0.05$), the decision to use which feeding treatment depends on the economic value and availability of the feed ingredients at a given time.

Table 1. Body weight changes of Horro rams fed the three experimental diets.

Parameter	T1	T2	T3	SEM	SL
IBW (kg)	25.00	24.94	24.89	3.01	ns
FBW (kg)	31.89 ^b	33.17 ^{ab}	34.89 ^a	2.89	*
BWCh (kg)	6.89 ^c	8.23 ^b	10.00 ^a	1.03	***
ADG (g/day)	72.52 ^c	86.63 ^b	105.26 ^a	10.64	***

^{a,b,c} Means with the same letter in the same row are not significantly different, *= $P<0.05$, ***= $P<0.001$, IBW: initial body weight; FBW: final body weight; BWCh: body weight change; ADG: average daily gain; SEM: standard error of mean; SL: significance level.

Carcass parameters

The regression analysis of the major carcass traits had shown a strong ($P<0.001$) relationship with slaughter weight (SW) of all the rams. As a result, there were significant variations ($P<0.05$) observed in EBW, HCW, DPSW, DPEBW and REMA among rams in T1&T2 and T2&T3. , However, the variations between T1 and T2 as well as between T2 and T3 for all parameters were not significant ($P>0.05$), except for the variation in REMA between T1 and T2 which was significant. Rib-eye muscle area is mostly used as a tool to indicate the proportion of carcass lean or an expression of carcass desirability (Wolf *et al.*, 1980). In this regard, supplementation appeared to impart better carcass quality characteristics.

There was no significant variation ($P<0.05$) in DPEBW between rams fed on T1 & T2 as well as between those fed on T2 and T3. However, there was a significant variation in DPEBW between rams fed on T1 (51.68 ± 2.9) and T3 (54.44 ± 2.9). According to Devendra and Burns (1983), dressing percent helps to assess the meat production capacity of animals. In this study, the higher DPEBW was obtained as a result of higher energy content of maize grain in T3 diet. Since all the treatment feeds were subjected to iso-

nitrogen, the variation was not as a result of CP (crude protein) content. Similar results were reported by Field (1971) and Mahgoub and Lodge (1994b). The later authors reported that higher growth and carcass composition differences could be obtained when animals are fed on high energy diets.

Table 2. Carcass characteristics of Horro rams fed on the three experimental diets

Parameter	T1	T2	T3	SEM	SL
SW (kg)	31.89 ^b	33.17 ^{ab}	34.89 ^a	2.89	*
EBW (kg)	26.24 ^b	27.06 ^{ab}	29.19 ^a	2.89	**
HCW (kg)	13.56 ^b	14.38 ^{ab}	15.89 ^a	1.5	**
DPSW	42.52 ^b	43.35 ^{ab}	45.54 ^a	2.3	**
DPEBW	51.68 ^b	53.14 ^{ab}	54.44 ^a	2.9	**
REMA (cm ²)	8.97 ^b	9.99 ^a	10.37 ^a	0.63	**

^{a,b,c} Means with the same letter in the same row are not significantly different, * $P < 0.05$, ** $P < 0.01$, SW: slaughter weight; EBW: empty body weight; HCW: hot carcass weight; REMA: rib-eye muscle area; SEM: standard error of mean; SL: significance level.

Partial budget analysis

In this analysis labor cost was not considered for all the treatments as it is almost uniform for all the three treatments. Actual feed price was taken at the time the experiment was conducted. Similarly, purchase price and selling price of the rams was estimated based on the current market price at the beginning and end of the experiment, respectively. There was a considerable variation in total variable cost among the three groups. Thus T1 showed the smallest total variable cost and T3 the highest. However, due to the higher final body weight, rams in T3 resulted in a slightly higher selling price, even though the variation in net return was not significant.

Table 3. Partial budget analysis of Horro rams fed on the three experimental diets

Variables	Treatments		
	T1	T2	T3
Sheep purchase price (ETB/head)	1750.00	1753.33	1748.89
Total concentrate cost (ETB/head)	281.16	307.80	413.64
Total variable cost (ETB/head)	281.16	307.80	413.64
Sheep selling price/estimate (ETB/head)	2655.56	2683.33	2800.00
Total return (ETB/head)	905.56	930.00	1051.11
Net return (ETB/head)	624.40	622.20	637.47

T1= 69.5% Wheat bran + 29.5% Noug cake + 1% Salt, T2= 55.5% Wheat bran + 33.5% Noug cake + 10% Maize + 1% Salt, and T3= 49.5% Noug cake + 49.5% Maize grain + 1% Salt

Conclusions and recommendations

From the current study it was observed that total substitution of maize grain with wheat bran resulted in a lower final body weight and average daily weight gain. On the contrary, as a result of variations in price

of the concentrate ingredients, the net return values were not significantly varied. So, wheat bran could substitute the whole maize grain and up to 40% of noug cake from the previously recommended concentrate feed (49.5% Noug cake + 49.5% Maize grain + 1% Salt) without affecting the crude protein content of the feed.

Therefore, utilization of a concentrate mixture comprising 69.5% Wheat bran, 29.5% Noug cake and 1% Salt could be more economical for fattening of Horro rams as compared to the previously recommended concentrate feed.

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Risk factors associated with incidence rates of camel calf morbidity and mortality in Borana, Southern Ethiopia

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ABSTRACT

A longitudinal study was conducted between July 2019 and May 2020 in Yabello and Gomole districts of Borana zone, southern Ethiopia; to estimate the incidence of mortality and morbidity rates of camel calves and their vulnerability to constraints. A total of 205 camel calves from Borana (n=98) and Gabra (n=107) herds were monitored from birth to six months of age. A total of 171 (83.41%) morbidity and 34 (16.58%) mortality cases were observed, making the overall crude morbidity and mortality rates of 15 and 3 cases per 100 calf-months at risk, respectively. The probability risk of occurrence of a disease or death for calves in the cohort during the six months period of time was 60% and 16.4%, respectively. Diarrhea had the highest incidence risk of 44.6% followed by pneumonia (39.7%) and pox (31.4%). The major causes of mortality were pneumonia (23.5%), Septicemia (14.7%), camel pox (14.7%), diarrhea (11.8%), swollen lymph nodes (8.8%) and Chronic wasting disease (8.8%), sudden death (8.8%) and starvation (2.9%). Incidence of mortality was higher in households that possess more diverse species (19.70%) than those with few types of livestock species (9.50). In this study, age of calf, colostrum feeding, calf delivery statuses, season, pre-weaning calf management and GIT parasite in dry season were significantly associated risk factors ($P < 0.05$) with camel calf morbidity and mortality. The incidence of mortality was 7.38 times higher in calves that consumed small amount of colostrum than those fed adequate colostrum. The negative impact of diarrhea and pneumonia was 5.37 and 2.68 times higher in calves positive for GIT parasite in dry season than negative animals respectively. Out of 254 fecal samples examined, 158 (62.2%) and 18 (7.1%) were positive for GIT parasites and coccidia oocyst respectively. Therefore to increase camel production and productivity; a sound intervention on determinants of camel calf health and good calf management practices are needed.

Keywords: Camel calf, Cohort study, Morbidity, Mortality, Pastoral, Borana

INTRODUCTION

In Ethiopia, dromedary camels are kept under pastoral managements in arid and semi-arid areas of the country; such as in Somali, Afar, Karayua and Borana (Workneh, 2002; Simenew et al., 2013). Borana

pastoralists, who are traditionally based on cattle husbandry for milk production and wealth storage, started camel production recently as compared to Gabra pastoralists (Mirkenaet *et al.*, 2018) due to ecological changes, social conditions and extensive seasonal migration (Biffa and Chaka, 2002). Thus, Gabra and Borana have different level of indigenous knowledge in camel keeping (Megersaet *et al.*, 2008). At the moment, all communities in the Borana area are keeping camels owing to the ability of camels to tolerate drought and supply the households with milk even during the dry periods (Borueta *et al.*, 2014; Megersaet *et al.*, 2014).

Camel calves are the foundation of a camel herd establishment and the potential replacement stock. However, rearing of camel calves under traditional systems is faced with several challenges that result in high losses due to death of the calves (Megersaet *et al.*, 2014). High camel calf mortality are generally considered important problems in pastoral camel husbandry systems (Kaufmann, 2000) and seasonal calving poses heavy managerial challenge on calf health care management while camel keepers often cannot access veterinary services. Studies indicate that the average calf mortality at pre-weaning stage is as high as 20% to 46.3% (Wilson, 1986; Kaufmann, 2003; Simpkin, 1985), with female calves' mortality ranging from 20 to 30% (Kaufmann, 2000; 2003).

Camel calf morbidity and mortality have been reported to be the most important hindrances to camel production, with a mortality reported to be as high as 50% in pastoral areas. The crude mortalities reported in Ethiopia were 15- 45% (Tuffa and Baars, 1998; Getahun and Kassa, 2002; Megersaet *et al.*, 2008). Similarly, Kaufmann (2005) reported mortality rates of 22%, 25% and 27% in Gabra, Rendille and Somali camel calves of Northern Kenya, respectively. This suggests a loss of camel calf vitally affects the replacement stock in particular, and herd productivity and population growth in general. Therefore, this longitudinal prospective study was conducted to estimate the incidence rate of camel calf mortality and morbidity and associated risk factors in Borana zone, southern Ethiopia.

MATERIAL AND METHODS

Study Area

The study was conducted in Yabello and Gomole districts of Borana zone. Geographically, the areas are located between 4 and 6° N latitude, and 36 and 42° E longitude with altitude ranging from 1000 to 1700 m above sea level. Borana rangeland is characterized by a semi-arid to arid climate (Teshome *et al.*, 2018). Camel herds are kept by Borana and Gabra communities.

Study Design and sampling procedures

A mix of cross-sectional and longitudinal study design was used during the study period. Longitudinal study was carried out between July 2019 and May 2020, to determine the incidence rate of calf morbidity and mortality. Additionally, a cross-sectional questionnaire survey was administered to collect information from camel owners (respondents) at the beginning of the study. The study target populations were suckling calves of dromedary camels (*Camelus dromedaries*) up to 6 months of age, and those reared under extensive and traditional production system. The two districts: Yabello and Gomole were purposively selected taking into account the distributions of the two major camel rearing community and camel population. Borana herders predominate in Yabello whereas Gabras are the major camel herders in

Gomole district. A total of six pastoral associations (the lowest administrative unit locally known as *Ganda*), namely Dida-Yabello, Dharito and Haro-Bakke (from Yabello district); Buya, Surupa Badiya and Bakke (from Gomole district) were randomly selected. From each PAs, seven villages (pastoral encampments) having eligible camel herds (herds with five or more breeding females were considered eligible) and respective community were selected purposively considering accessibility to the villages and herds throughout the survey period. The number of breeding females per herd was determined to increase the probability of having at least one calf that born during the study period.

Sample Size Determination

The number of respondents (camel herders) for face to face interview was estimated using central limit theorem ($n = 0.25/(SE^2)$) described by Arsham (2002) taking the standard error (SE) of 0.05 with 95 % confidence level plus 10% contingency for refusal or incomplete data. From the herders that fulfilled criteria in each PA, 112 herders were purposively sampled and equally distributed by two communities. Subsequently, about half of the herds (a total of 51 herds) were recruited for the longitudinal study (monthly visit) based on willingness of herders to participate in the study and accessibility of herds throughout the study period. The study herds belonged to both Borana (n=27) and Gabra (n=24) herders.

The sample size of calves required for the cohort study was calculated using the formula described by Thrusfield (2005). For this purpose, 95% confidence level, a prevalence of 18.1% from previous study report (Megersa, 2014) and 5% desired precision were used as follow.

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

Where n = required sample size, d = desired absolute precision, P=expected prevalence (18.1%). Based on this the desired sample size was found to be 228 calves. However, the actual number of legible calves available in the study herds during cohort period was 205 calves, which were monitored over the six months cohort period.

Data collection

Questionnaire survey

During the first visit to the selected villages, semi-structured questionnaire was administered by a face to face interview to camel herders. The questionnaire was focused on collecting herd-level data such as management and husbandry practices, production objectives, constraints, health care, perception and restriction of colostrums intake, time and method of colostrum feeding, frequency of colostrum feeding, weaning age, history of calf morbidity and mortality in the herd and other variables. Additionally, calf-level data such as age of calf, time and season of birth, delivery status of the calf (normal, or dystocia or assisted birth), time of colostrums intake, and the parity of the dam were collected. Herders were also solicited to provide detail reproductive history profiles of each breeding female as well as fates of each pregnancy and calves borne alive using recall methods.

Monitoring of calves (cohort study)

A total of 205 calves born from July to November were enrolled and followed monthly until the calves completed the six months cohort period or lost to follow-up. At the start, new born calves (that aged birth

to 15 days using recalls on disease events) were included in the study. Other calves were enrolled progressively as they were born within the selected herds during the study period. During the cohort period, calves were regularly monitored every month to collect data on occurrence of morbidity and mortality and their causes. When calf loss occurred during the study period, date and reason of loss was documented. In this study, morbidity was defined as any illness with recognizable symptoms showed by calves which may ultimately results in death or recovery during the course of follow up; whereas, mortality was defined as any observed death of calves above the age of 24 hours, irrespective of the cause. Thus, a calf was considered to be at risk of acquiring a disease that occurred for the first time, and a calf recovered from a disease is also at risk of being affected by another illness. Likewise, any calf present alive in the cohort was considered to be at risk of dying of any disease.

During the study period, the calves were observed for clinical symptoms besides performing physical examinations for the presence of any disease conditions. Additionally, data on cleanliness of calves, house conditions, feeding and watering practices, and treatments and drug used were collected. Whenever any health problem encountered, the affected calf was examined clinically; appropriate samples, particularly fecal samples were collected and examined for parasitic identification.

Fecal sample collection and examination

Fecal samples were collected directly from rectum of camel calves for the examination of gastrointestinal parasite eggs and protozoal parasites. A total of 254 fecal samples were examined using floatation and modified McMaster eggs counting techniques for detection of parasites eggs and protozoal oocysts as described by Soulsby,(1982). All parasites eggs and coccidian oocysts were counted and the counts multiplied by 50 to obtain the total number of eggs per gram of feces.

Data Management and Statistical Analysis

Data obtained from the study were analyzed using Stata Version 13. Data from questionnaire survey were presented as frequency distributions by tables. Rank data were normalized by converting ranks into standardized values (0, 1), with values close to 0 and 1 indicating that a variable was ranked least and highest, respectively (Ouma *et al.*, 2011).

$$\text{Normalized Rank} = 1 - \left(\frac{(\text{rank} - \text{rank}_{\min})}{(\text{rank}_{\max} - \text{rank}_{\min})} \right)$$

As animals enrolled in this prospective study were at different times and followed for different periods of time and hence incidence density (true rate) was used in describing diseases occurrences. The speed at which an event occurs per unit time at risk (true rate) was calculated to define the risk of morbidity, mortality and other specific disease conditions (Muraguri *et al.*, 2005). Therefore, the incidence rates (IR) or cause specific rates were estimated by the following formula.

$$IR = \frac{\text{number of illness or death occurred during observation period}}{\text{total calf days at risk}}$$

The numerator is the total number of ill /dead calves, whereas the denominator is the number of calf-days at risk for the given period. Number of calf days at risk was calculated by adding the number of days at risk of obtaining a new case in each calf from birth up to end of the study (Martin *et al.*, 1987). For this

purpose, total calf days at risk were converted to calf months at risk, as the age of calf is defined up to six month. Furthermore, to simplify result comparisons with other findings and because of directly taking true rate results tend to overestimate calf morbidity and mortality rates (Gitau *et al.*, 1994), incidence rate was converted to incidence risk ratio (IR) or cumulate incidence rate (CIR) as probability of developing disease or dying over a time t (i.e. t=6 months) using the formula:

$$CIR = 1 - e^{-IR \cdot t}$$

For survival analysis, Kaplan–Meier method was employed to estimate hazard function of observed hazard differences for different explanatory variables with crude mortality and specific disease conditions. To assess the associations between potential risk factors and survival up to 180 days of age, Cox’s proportional hazard model was applied. Firstly, the association of individual risk factor with an outcome variable was screened by log-rank test and then variables with $p < 0.05$ in the univariable analysis were modeled using multivariable Cox-regression assess their independent effect.

RESULTS

3.1. Demographic characteristics of the respondents

The demographic characteristics of the camel owners were shown in Table 1. The age range of household varied between 23 and 95 with an average age of 49.79 ± 17.3 years. Most of the respondents (93.66%) were illiterate and the remaining few proportion (6.44%) had elementary education. Majority (82.14%) of households let the calf to start full suckling just after birth, but others (53.6%) restricted the amount of colostrum suckled by a newborn calf through different techniques such as limiting suckling time (58 %), half teat suckling (28.3%) and partial milking before suckling (13.3%). The practice was always associated to fear of fatal scouring. Weaning is found among the commonly known breeding practice in the study sites. Among the interviewed camel rearing households, 41.9 % and 40.2 % practice weaning in 1 and 2 years respectively and the remaining households (17.9 %) wean the calf any time when pregnancy is observed.

Table 1. Demographic Characteristic and calf management practice of respondents

Variables		Parameters	Borana (N=56)	Gabra (N=56)
Age of respondent (years)		Mean \pm SD	52.13 \pm 17.7	47.5 \pm 16.8
Family size (number)		Mean \pm SD	7.91 \pm 2.7	7.6 \pm 3.1
Sex	Female	Frequency	2 (3.6)	0
	Male	Frequency	54 (96.4)	56 (100)
Marital status	Single	Frequency	3 (5.4)	1 (1.8)
	Married	Frequency	52 (92.9)	55 (98.2)
	Widow	Frequency	1 (1.8)	0
Education	Illiterate	Frequency	54 (98.2)	48 (85.7)
	Grade 1 to 4	Frequency	1 (1.8)	6 (10.7)
	Grade 5 to 8	Frequency	0	2 (3.6)
Restrict colostrum	Yes	Frequency	29 (51.8)	31 (55.3)

intake	No	Frequency	27 (48.2)	25 (44.6)
Age at weaning	1 year	Frequency	22 (39.3)	23 (41.0)
	2 year	Frequency	24 (42.9)	23 (41.0)
Importance of colostrum	pregnancy	Frequency	10 (17.9)	10 (17.9)
	Strength	Frequency	32 (57.1)	50 (89.2)
	Natural	Frequency	4 (7.14)	1 (1.8)
	Fast growth	Frequency	18 (32.1)	2 (3.6)
	Immunity	Frequency	2 (3.6)	4 (7.1)

3.2. Camel herd structure and calf management

The camel herd structures of both community (Borana and Gabra) were indicated by Figure 1. Herds were dominated by breeding adult females, accounting for 50% of the animals, followed by young females and males.

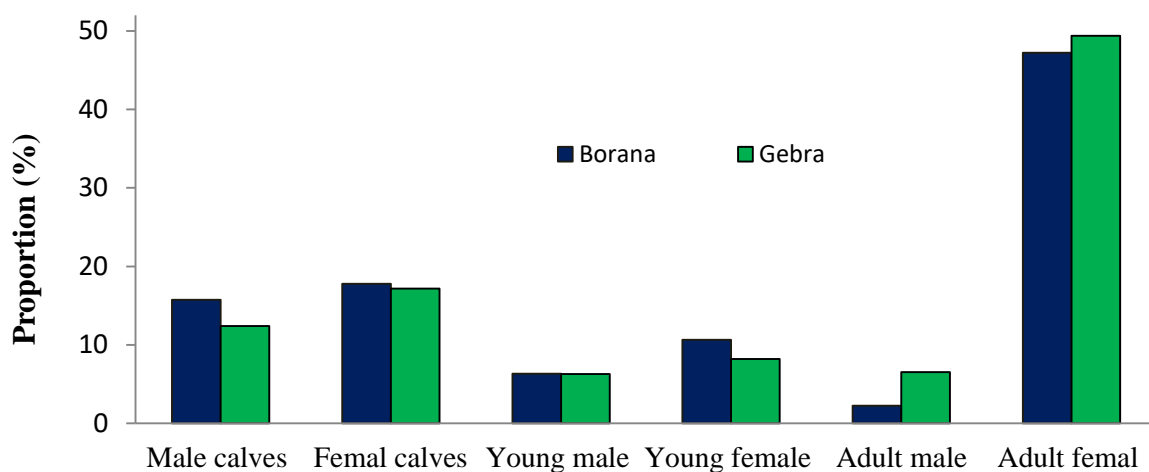


Figure 1. Proportion of camel herd structure by community

3.3 Constraints and diseases affecting camel calves

The major camel production constraints reported by respondent pastoralists were shown by figure 2. The major constraints of camel production in the study area were prevailing diseases (94%), feed shortage (81%), inadequate veterinary services (69%) and market constraints (50%).

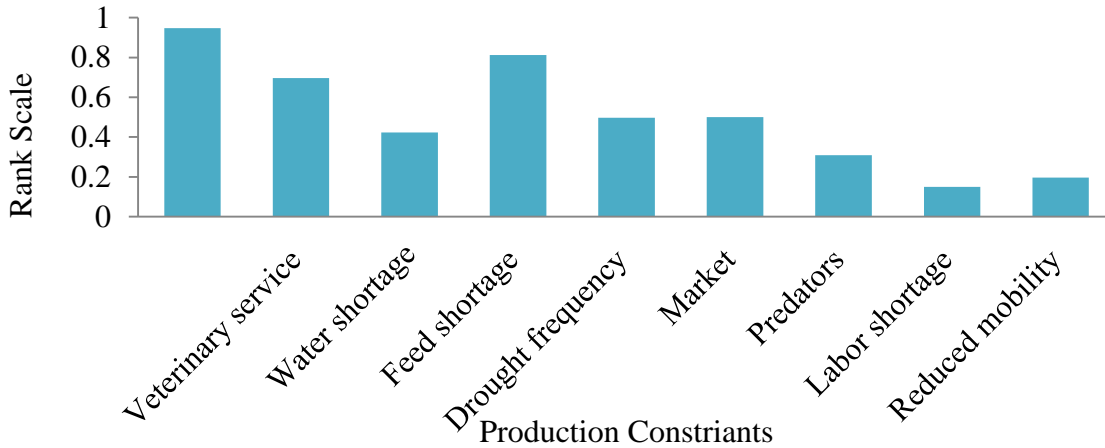


Figure 2. Camel production constraints of the study area as responded by interviewed pastoralist (n=112).

In the study area there are several diseases that cause calf morbidity and mortality (Figure 3). According to respondents, the major diseases that cause mortality on camel calves include camel pox, septicemia, swollen lymph nodes (kandhicha), neck paralysis (shimbirki), chronic wasting disease (Elgof), Calf diarrhea (alhati) and pneumonia (furi). while, camel pox (baga), Septicaemia and joint ill (dhidhiksi), calf diarrhea, pneumonia, respiratory infection (furi), swollen lymph nodes (kandhicha), chronic wasting disease (*Elgof*), neck paralysis (shimbirki), contagious skin necrosis (dhulla), mange and contagious ecthyma (amburur) were among diseases that causes morbidity on camel calves.

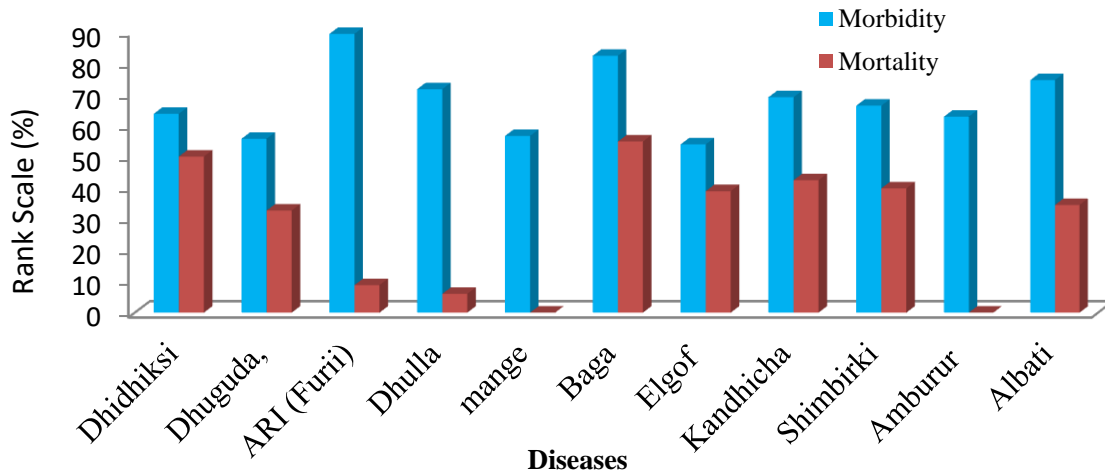


Figure 3. Reported camel calf diseases that cause morbidity and mortality (n=112)

3.4. Gastrointestinal tract parasites of camel calves

In addition to other health issues, parasitic infections have been the most widespread problems in the study sites (Table 2). The current study showed that 62.2% (95% CI = 48.1 - 64.9%) of the study animals had nematode and cestodes (Monezasppegs). Likewise, prevalence of coccidia oocyst was 7.1% which is relatively low. Average count of eggs per gram of feces was found to be 326.9 ± 20.9 .

Table 2. Parasitic infection of camel calves (N = 254) by helminthes eggs (mean EPG) and Coccidiaocyst (%) distributed by sex, season and PAs

Factors		Helminthes eggs			CoccidiaOocyst			
		Samples	positive	%	Mean EPG	sample	positive	%
Sex	Male	112	90	80.3	318.7	112	7	6.3
	Female	142	68	47.5	333.5	143	11	7.7
Seasons	Short wet	95	50	52.6	304.2	95	7	7.4
	Dry	159	108	68	340.5	159	11	6.9
District	Yabello	121	77	63.6	502.5	121	10	8.3
	Gomole	133	81	60.9	487.4	133	8	6
Total		254		62.2	326.9	254	18	7.1

3.5. Longitudinal cohort study

A total of 205 camel calves were enrolled and followed monthly for six months. Female and male calves contributed 112 (54.63 %) and 93 (45.37%) of the entries over the observation period, respectively. They contributed to a total of 34122 calf days at risk, which is also equivalent to 190 calvessix month at risk or 1137 calf month. The distribution of the study cohort is shown in Table 3. A total of 35 calves exited from the study before the termination of the cohort period (due to deaths and gift). The total exit rate was 17.07% of which 19 (9.26%) and 16(7.8%) were female and male calves, respectively

Table 3. Number of calves (cohort) monitored and reasons of withdrawals from the longitudinal cohort

Monthly visits	number of birth	withdrawals			total	number of cohort
		death	gift out			
July	52	0	0	0	52	
August	46	2	0	2	96	
September	44	2	0	2	138	
October	27	7	0	7	158	
November	36	5	0	5	189	
December	NI	10	0	10	179	
January	NI	2	0	2	177	
February	NI	2	0	2	175	
March	NI	3	0	3	172	
April	NI	1	1	2	170	
May	NI	0	0	0	170	
Overall	205	34	1	35	170	

NI =Not included

Table 4 shows summary of incidence Rate (IR), calf days and six months at risk in associated disease condition. The present 180 days longitudinal prospective study revealed that the incidence of crude morbidity and crude mortality risk rates were 60.0% and 16.4%, respectively. From disease conditions

encountered during the follow up period, diarrhea was the primary reason of calf morbidity with risk rate of 44.5%, followed by pneumonia (39.7%), camel pox (31.4%).

Table 4. Incidence rate of crude morbidity, crude mortality and incidence of selected diseases in Yabello and Gomoledistrict, Borana zone

<i>Disease condition</i>	<i>No cases</i>	<i>Calf days at risk</i>	<i>Calf months at risk</i>	<i>Incidence rate per calf months at risk</i>	<i>Incidence risk over six months (%)</i>
Crude morbidity	171	33558	1119	0.15	60.0
Crude mortality	34	34122	1137	0.03	16.4
Diarrhea	79	24063	802	0.10	44.6
Pneumonia	77	27403	913	0.08	39.7
Pox	66	31526	1051	0.06	31.4

In this study, major cause of death in most camel calves was pneumonia, septicemia (dhidhiksi), camel pox and calf diarrhea which were evidenced by 8, 5, 5 and 4 death records out of the total 34 dead calves respectively. Three calves died of kanicha (swollen lymph nodes) and the other 3 and 2 calves were died in association with chronic wasting disease (elgof) and bloat, respectively. Other observed causes of calf death were sudden death (3) and starvation (1).

Pre-weaning incidence risk rate of crude mortality and morbidity across study ethnic groups, PAs, livestock species diversity and type of management practice is given in Table 5. When calf health problems were compared by community the incidence of crude mortality and crude morbidity was not varied significantly between Borana and Gabra camel calves. But incidence of crude mortality was not similar across Pastoral Associations and higher incidence was observed in Dharito, Buya, Bakke and Surupa Badiya than Dida Yabello and Haro Bakke. Regarding crude mortality in relation to livestock species diversity, it increases in household that holds more than four types of livestock species.

3.6. Association of explanatory variables with camel calf Morbidity and Mortality

Analysis of camel calf mortality with respect to risk factors showed that Age, Colostrum feeding, Calf delivering status, season, type of pre-weaning management and animal species diversity were associated with mortality (Table 5). Accordingly, the result obtained from univariable analysis showed risk factors are found to significantly influence camel calf mortality. As a result, all the mentioned risk factors were considered to the next model (multivariable analysis). Consequently, multivariable Cox-regression model showed, calf age, method of colostrum suckling, calf delivery status, seasons and type of pre-weaning management practice were found to significantly influence crude mortality of camel calves (Table 5). According to the model, keeping the effect of other variables constant, the hazard of mortality was 7.38 fold higher in calves those consumed limited amount of colostrum than those fed adequate colostrum.

Table 5. Explanatory variables associated with the incidence of crude mortality in Cox regression model

Variables	levels**	Univariable		Multivariable	
		HR*	P-value	HR	P-value
Age	>3m vs.<3m	0.32	0.001	0.37	0.028
Colostrum feeding	Full vs. limited	17.8	0.000	7.38	0.022
Calf delivery status	Normal vs. assisted	0.07	0.000	0.14	0.000
Seasons	Dry vs. Minor wet	8.7	0.000	1.69	0.000
Pre-weaning calf mgt	KH vs. KGH	0.18	0.005	0.10	0.030
Animalspp diversity	>4 sppvs.≤4 spp	2.05	0.002	2.13	0.074

*Hazard ratio (comparable to relative risk). ** KGH=Kept in grazing herd and KH=Kept at homestead** levels written second or right side were reference

Primarily 29 independent variables were recruited in univariable analysis using Cox regression procedure. Accordingly, seven explanatory variables (short rainy season parasite load, dry season parasite load, Season, colostrum feeding management, pre-weaning calf management, livestock species diversity and sex of calf) for diarrhea were found significant at $P<0.05$ (Table 6). Variables that showed significant ($P<0.05$) association in univariable analysis subjected to multivariable Cox regression model (Table 6). Accordingly, GIT parasite load during dry period, seasons and pre-weaning camel calf management practice showed significant ($P<0.05$) association with incidence of diarrhea in the multivariable Cox regression model. Holding the effect of other risk factors constant, the hazard of diarrhea was 5.37 times higher in calves infected with GIT parasite in dry season than those without parasite which is clearly indicated in Kaplan-meier survival estimate.

Off the total 29 independent variables included in univariable Cox regression analysis for camel calf pneumonia (Table 6), only four (Calf delivery status, GIT parasite dry season, Seasons and pre-weaning calf management) were found ($P<0.05$) associated with subject matter. Among the variables include in multivariable Cox regression model GIT parasite burden in dry seasons, season, calf delivery status and type of management practice in pre-weaning were significantly ($P<0.05$) associated with pneumonia, used in multivariate Cox regression model and only extent of animal diversity owned found insignificant ($P>0.05$). Holding the effect of other risk factors constant, the hazard of pneumonia was 2.68 times higher in calves positive for GIT parasite in dry season than that of negatives. The association between Pre-weaning calf management and calf pneumonia is clearly demonstrated under Kaplan-meier survival estimate.

Table 6. Potential risk factors associated with the incidence of calf diarrhea, pneumonia and pox occurrence in Cox regression model

Variables	levels**	Multivariable	
		HR	P-value
Diarrhea			
GIT parasite in wet	Positive vs. Negative	0.97	0.967
GIT parasite in dry	Positive vs. Negative	5.37	0.036
Colostrum feeding	Partial vs. Full	0.72	0.605
Seasons	wet vs. dry	1.1	0.000
Calf management	kept homestead vs. grazing area	0.29	0.046
Animal spp diversity	>4 SPP vs. ≤ 4 SPP	1.14	0.777
Sex	Male vs. female	1.2	0.735
Pneumonia			
Calf delivery status	Assisted vs. Normal	0.43	0.045
GIT parasite DS	Positive vs. Negative	2.68	0.002
Seasons	wet vs. dry	0.48	0.043
Calf management	kept homestead vs. grazing area	0.41	0.039
Pox			
Age	>3m vs.<3m	3.51	0.001
Colostrum feeding	partial vs. full	2.22	0.002
Seasons	wet vs. dry	0.61	0.093

From various factors that were subjected to univariable Cox regression analysis, only 3 risk factors had significant effect on pox occurrence. Following the procedure of Cox regression analysis, these factors (age, type of colostrum feeding and season) were considered for subsequent analysis. Consequently, the result from multivariate Cox regression model showed only age group of the calf and method of colostrum feeding were found to be significantly associated with pox in the multivariable Cox regression model.

4. DISCUSSION

The present study revealed that the morbidity and mortality of camel calves are important problems of camel production. The most important diseases that cause morbidity and mortality in camel calves in this study is analogous to other study reported from different areas in Ethiopia such as Megersa (2010); Keskeset *al.* (2013); Tefera (2013) and from neighboring countries (Gilbert, 2012; Kuria *et al.*, 2011).

Four different types of gastrointestinal parasites were identified in camel calves and *Strongylus* species were found the predominant one which occurred in 111 (43.7%) out of 254 calves examined. The current prevalence was comparable to the previous prevalence (47%) reported in afar national regional state, Ethiopia (Gebruet *al.*, 2017) but lower than the prevalence of 74.4% in dry season and 69.8% in minor wet season reported in Borana by Megersa (2010).

The overall crude mortality of 16.47% in this study is similar to the previous mortality reports of 18.1% and 15 to 20% by Megersa *et al.* (2008; 2014) from Borana and 20.4% by Zeleke and Bekele (2000) from eastern Ethiopia. However, a higher crude mortality rates of 30% was reported by Tuffa and Baars (1998) and 45% by Getahun and Kassa (2002) from Eastern Ethiopia. Similarly, Kaufmann (2005) had reported mortality rates of 25%, 22% and 27% in Rendille, Gabra and Somali camel calves of Northern Kenya, respectively. The variation of research results observed countries is attributed to disparity in location and period of study.

In this study, camel calf diarrhea was the primary cause of calf morbidity with incidence rate of (44.6%), followed by pneumonia (39.7%), camel pox (31.4%). This result is congruent with previous cross-sectional survey research findings that identified diarrhea as most important survival constraints of camel calf (Megersa. 2010; Rirashet *et al.*, 2017).

In the present study, the major causes of death in most camel calves is considerably in agreement with the studies by Megersa (2010) in Borana and Ahmed and Hegde (2007). Sunken eye (12%) camel pox (7%), pneumonia (7%), contagious ecthym (6.9%) and contagious necrotic skin (4.6%) were reported cause of camel calf death. Farah *et al.* (2007) reported diarrhea as a major cause of death (73%) in Somalia. On the other hand, Mahmoud *et al.* (2012) were reported camel pox caused high morbidity (80%) and mortality (12.7%) on calves in Egypt.

The result showed age of the calf significantly influenced crude mortality. In this study, calves aged below three months were found at higher risk of death compared to those aged above three months and this is in line with other studies done by Farah, *et al.* (2004) in Somalia and Rirashet *et al.* (2017) in Somali region of Ethiopia. This higher crude mortality of camel calves in the present study could be attributed to undeveloped immunity system in the new born calves during the first 3 months of life and their susceptibility to environmental conditions. Restriction of colostrum consumption was found to negatively affect camel calf survival and accounted for higher risk of mortality compared to those calves with unlimited colostrum feeding. The finding is in agreement with previous research reports by (Kamberet *et al.*, 2001; Bishret *et al.*, 2013; Paxsonet *et al.*, 2008).

In the present study birth related disorders were significantly associated with camel calf mortality and morbidity of camel calf pneumonia. Calves that delivered through assistance were at higher risk of mortality than that normally delivered. The influence of this factor observed in the current study is found analogues with research finding by (Bellows, 1997). Mortality was significantly higher during dry season than minor wet season. This is because of the coincidence with shortage of feed and water especially during dry period. This finding is consistent with the research reports of Rirashet *et al.* (2017) who explained feed shortage during dry period found the major impact on death of calf in Borana area. In the contrary, a study by Megersa (2010) which conducted in the same area showed that the higher mortality rate camel calf were during the wet seasons than the dry season.

Calves kept on communal rangeland of this study were at higher risk as compared to that kept at homestead before weaning. This could be related to most of the diseases that cause morbidity and

mortality on camel calves were contagious which can be transmitted directly by contact or indirectly through objects. Overall, the calves that let to browse with the herd earlier may have a maximum exposure to various disease causing factors which logically can increase risk of mortality compared to calves kept at homestead.

Season of the year in which camel calves were monitored has significant impact on the health problems and survivability of camel calves. Regarding parasitic load in dry season, the present finding revealed that GIT parasites in dry season were found among the influencing factor of camel calf diarrhea and the result is in line with finding of Megersa (2014).

The risk of diarrhea on camel calves significantly higher in short rainy season than dry. This finding was congruent with Megersa, (2014) who reported that morbidity rate of diarrhea in camel calves was 4.2% during dry season and 6.5% during wet season. Similarly, Agab and Abbas (1999) reported a high incidence of calf diarrhea that affected 21.9% of one year-old calves studied, with the peak of occurrences during early summer coinciding with the peak of the calving period for camels in Sudan. The reason for that calves more infected in wet season is due to favorable environmental conditions for the pathogens and the tradition breeding calendar where most of the mating and parturition is tagged in wet season.

In the present study, diarrhea was higher in calves kept with grazing herd as compared to those kept at homestead. This difference could be ascribed to higher exposure of calves to contaminated production environment which usually work synergistically and increase the opportunity of being diseased due to increased duration of exposure to a higher quantity of pathogens (Larson *et al.*, 2004; Larson and Tyler, 2005).

Association of GIT parasitic with pneumonia indicated the impact of parasitic burden and feed shortage in dry season compared to short wet season that reduces the immunity of the calves and may predispose them to other infections. This is in agreement with previous research outputs that stated the indirect effects of several factors to a particular incidence (Anwar and Khan, 1998) and also definitive etiology of most respiratory diseases of camels has not yet been determined as a variety of viruses, fungi, bacteria and parasites are to be the possible causes of respiratory outbreaks among camels (Kebede and Gelaye, 2010).

The result of the study indicates incidence of pneumonia was significantly higher in minor wet season than dry season. This finding is in line with studies by Megersa (2014) in Borana area where respiratory infection, which has occurred in camel calves were high during minor wet season (7.1%) than dry (1.4%). Moreover, Agab and Abbas (1999) as well as with reviews where housing of camels in unsheltered pens are major predisposing factors as long as dust storms emerging during the first weeks of the rainy season in the African Sahel can contribute to respiratory disease in camels (Abbas *et al.*, 2002).

Calves let to graze with grazing herd to communal rangeland as early of their growing stage were at higher risk of pneumonia as compared to the one kept at homestead before weaning. This finding is matched with study by Hassen and Mustafa (1985) and Melaku and Feseha (1988) those reported crowding of pastoralist camels around limited watering points during the summer contributes to the

spread of respiratory pathogens as camels from different geographical regions often use the scanty open water sources.

Camel calves that were fed limited amount of colostrum were at higher risk of pox infection than calves those allowed to freely feed colostrum. This finding agrees with research report of Bishret *et al.* (2013) and Farah (2007) those explained camel calves are born with an immature immune system. So they require immediate passive immune transfer through colostrum (first-milk) IgG intake within 24 hours of birth. Because, calves rely exclusively on passive immunity from antibodies absorbed from maternal colostrum for their protection against infection during the first few weeks of their life. Similarly, camel pox has been characterized as strong maternal passive immune transfer and transmitting through tick (*H. dromedary*) vectors (OIE Terrestrial Manual, 2014). As a result, risk of infection would naturally increase when contact with potentially infected animals and/or vectors increase after maternal antibodies in circulation had dwindled

CONCLUSION AND RECOMMENDATIONS

In general a camel plays a vital role in contributing to food security and human wellbeing in vulnerable pastoralists of Borana lowlands. Particularly, Camels are found as key source of milk and meat productions, transportation, prestige and pastoral household income generations. In this study the most challenging constraints of camel productions were identified to be diseases prevalence, shortage of feed, poor veterinary service and access to drugs and vaccine. Calf age, colostrum feeding, calf delivery status, seasons and pre-weaning calf management were found significantly associated with crude mortality, diarrhea, camel pox and pneumonia. Diarrhea, Camel pox and Pneumonia were the predominant camel calf health problem responsible for the majority of camel calf illnesses and mortality. Therefore, to improve production and productivity of camel and camel calf health an intervention considering causes and risk factors associated with morbidity and mortality of camel calf with subsequent application of improved calf management, feeding complete colostrum, avoiding early release of camel calves to the rangeland and mixing them with matured camels before weaning should be implemented and further study on camel diarrhea, pox and pneumonia should be done.

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Spatial Sero-Prevalence of Contagious Bovine Pleuropneumonia in Pastoral and Agro-pastoral Districts of Borana Zone, Southern Oromia, Ethiopia

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Abstract:

This study, conducted from November 2019 to April 2020, aimed to assess the epidemiological distribution of the disease, to identify the risk factors involved and to know the perception of the community about the disease. Sera samples were collected from a total of 498 cattle from five randomly selected pastoral districts. The samples were tested using competitive enzyme-linked immunosorbent assay (cELISA). Out of a total of 498 sera samples tested 46 (9.246%) were positive for antibodies. The highest prevalence was observed in Teltele (12/95; 12.90%) followed by Yabello (12/104; 11.54%) and Arero (10/91; 10.99%), whereas the lowest prevalence was observed in Gomole (5/101; 6.42%) and Dubluk (7/109; 4.95) districts. The occurrence of contagious bovine pleuropneumonia (CBPP) in the five districts was not found statistically significant ($P > 0.05$). Age, herd size and animal movement of the animal were shown to have association with the prevalence of CBPP and significant difference ($P < 0.05$) was observed. Sex, season of the year and body condition of the animal was not significantly ($P > 0.05$) associated with the occurrence of CBPP. The study indicates that CBPP is one of the major problems that affect health and productivity of cattle in the study area. Awareness creation to the pastoralists in the study area about the effect of CBPP and designing appropriate control methods has a paramount importance to improve the health and productivity of cattle production in the area.

Keywords: Geo-spatial, Prevalence, CBPP, Borana, Ethiopia

1. Introduction

Even though the livestock sub sector contributes much to the national economy, its development is hampered by different constraints. The most important constraints to cattle productions are widespread endemic diseases including viral, bacterial, and parasitic infestation and poor veterinary service (Zewdie, 2004; Kuastros 2007). Of the health constraints, tick and tick borne disease Foot and Mouth Diseases (FMD), Contagious Bovine Pleuloro Pneumonia (CBPP), Brucellosis, Lumpy Skin Diseases (LSD) contribute to the great financial losses and the socio-economic development of poor farmers in the area (Zewdie, 2004).

Contagious Bovine Pleuro Pneumonia (CBPP) is a highly contagious disease of cattle that is caused by *Mycoplasma mycoides* subsp. *Mycoides* small colony (*Mmm* SC). Cattles infected with CBPP (caused by *Mmm* SC) has a tendency of becoming “carriers” which may consequently lead to the spread of the disease (Musa et al., 2016). The disease is usually spread by movement of animals across international boundaries with devastating consequences on cattle, particularly in severe outbreaks (Tambuwal et al.,

2011 and Thiaucourt et al., 2011). CBPP causes production losses, increases production costs via increased disease control costs, compromises food security through loss of protein and draft power, disrupts livestock and livestock products trade, retards genetic improvement and inhibits sustainable investment in livestock production and causes pain and suffering to animals, causes high morbidity and mortality losses especially in newly affected areas or among susceptible herds that may show 100% morbidity with mortality exceeding 50% (Tambi *et al.* 2006; Lesnoff *et al.* 2004). The CBPP induced productivity losses are associated with significant financial losses to cattle owners (Tambi *et al.* 2006), especially in production systems with poor control of cattle movements that is typical of the predominant communal grazing system like in Borana. Absence research works conducted in Borana zone Southern Oromia, on CBPP indicated as there is no information about the epidemiological status of the CBPP and especially on molecular identification of the causative agent; but there are frequent reports of the disease from zone which borders Kenya and where most of export animals for the country exit. Therefore this study was conducted with the objectives to assess the epidemiological distribution of the disease, to identify the risk factors involved and to know the perception of the community about the disease.

2. MATERIALS AND METHODS

2.1. Description of the Study Area

The study was conducted in five districts of Borana pastoral zone namely Arero, Dubluk, Gomole, Teltele and Yabello. Borana zone, predominantly inhabited by the Borana Oromos, extends to the Kenyan border in the South Somali Region in the South East, Southern Nation Nationalities and People Region (SNNPR) in the West and North, and Guji zone in the North East. Borana rangelands characterized by a semiarid to arid climate (Haile *et al.*, 2011; Kamara *et al.*, 2005). Geographically the area is located between from 4° to 6°N latitude and 36° to 42°E longitude with altitude ranging from 1,000 to 1,700 meters above sea level. The topography of the area consists of isolated mountains and valleys (Coppock, 1994; McCarthy *et al.*, 2002).

The mean annual rainfall of the area ranges from 250 to 700 mm. The annual mean temperature varies from 19 to over 25°C. Extensive pastoralism is the main means of livelihoods for the Borana people. Nomadic pastoralism is the rational livestock system in the area (Gelagay *et al.*, 2007). The area has a bimodal rainfall distribution, with the long rains “*Ganna*” extending from March to May and the short rains “*Hagaya*” from September to November. A cool dry period “*Adoolessa*” (June to August) bridges the two rainy seasons, while a warm dry season (major dry season) “*Bona Hagaya*” runs from December to February (Coppock 1994; Angassa and Oba 2007).

2.2. Study Design

A cross-sectional study design was employed using field survey and serum sample collection. The design includes both sex groups above six months of age to estimate sero-prevalence. Cattle selected for sampling were identified and data on household owning the animals, district, pastoral association (PA), age, sex, flock size and date were also recorded. Their ages were determined using information from the owner and dentition.

2.2.1. Sampling strategy

Multistage random sampling coupled with systematic sampling method was applied to select the study animals.

Sampling frame

The sampling frame includes a list of all districts in the zone and Pastoral associations (PAs) or villages. Villages where vaccinations were not given against CCPP were selected purposively.

2.2.2. Sample size determination

The determination of sample size for serum collection was based on the formula given by Thrusfield, (2005) for simple random sampling method with 95% confidence level and 5% absolute precision was considered.

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

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

Study on CCPB conducted in Borana pastoral areas using cELISA were not available so that prevalence of 50%. Thus, 50% was used as the expected prevalence. Using the 95% confidence level and an expected prevalence of 50% and 5% absolute precision a minimum sample size of 384 cattle were determined, however, 498 cattle were involved in the study.

2.2.3. Sample collection

Blood sample for Serology

Five to ten millilitres of blood sample for serum extraction were collected from the jugular vein of each cattle with no history of vaccination using sterile plain vacutainer tubes and needles. Collected blood samples were labelled and kept in icebox and then transported to Yabello Pastoral and Dryland Agriculture Research Center Microbiology laboratory. The samples were kept in a slanting position for 6-8 hours and centrifuged to separate sera from coagulated blood; then the separated serum was transferred to a sterile tube and stored at -20°C until the samples were transported to National Veterinary Institute (NVI) for test using c-ELISA.

GIS data

GPS coordinate were taken and finally the spatial prevalence was mapped for Positive animals Using Quantum GIS software

2.2.3. Competitive ELISA (cELISA)

Serum samples were examined for the presence of specific antibodies using cELISA. The 498 test sera were examined according to the test protocol supplied with the kit.

2.3. Data Management and Statistical Data Analysis

All data were entered and stored in separate data bases in MS-excel files. Data was screened for proper coding and then transferred to stata 13.0 (Stata corp.1985-2013) statistical software. Seroprevalence of CCPP was calculated based on c-ELISA positive results. Flock and animal level seroprevalence was calculated by dividing the number of c-ELISA reactors by total number of tested flocks and animals, respectively. Logistic regression analysis was used to determine the associations between the potential risk factors and seroprevalence. The strength of association of each risk factor was determined by the magnitude of Odds Ratio (OR). Questionnaire data was analyzed by descriptive statistics using SPSS statistical software version 20.

3. Results and Discussions

3.1. Sero-prevalence and Associated Risk Factors CBPP

One of the most powerful benefits of GIS has been its ability to integrate different databases into a single environment for possibilities of improving surveillance and control programs. From a total of 498 cattle sera tested 46 (9.24%) of them were found positive (Table 1) and the finding is similar with previous work done by (Ahmed, 2004, Birhutesfa et al., 2015, Teklue et al., 2015 and Mamo et al., 2018). The current overall prevalence of CBPP is higher than the reports of (Kasaaye and Molla, 2012 and Wakgari et al., 2018) who reported 4% and 6.9% prevalence respectively in Ethiopia. However; lower than the prevalence reported by (Alhaji et al., 2016; Daniel et al., 2016; Malicha et al., 2017 and Fulasa et al., 2020) in different parts of Ethiopia.

The prevalence of CBPP was 12.90%, 11.54%, 10.99%, 6.42%, and 4.95% in cattle tested from Teltele, Yabello, Arero, Gomole and Dubluk districts respectively and no statistical difference was observed in CBPP occurrence between the districts. This result contradicts with the previous findings by (Sery et al., 2015 and Wakgari et al., 2018) who reported statistically significant difference among the districts. This could be due to agro ecology; animal management and movement in these districts are the same. With respect to prevalence of CBPP according to age category, the prevalence was higher in adult cattle (11.46 %) than in younger ones (5.14%) and the association shown was significant in this study. This result is in agreement with the previous report by (Kasaaye and Molla, 2012; Emanuel et al., 2013; Bashiruddin et al., 2015 and Wakgari et al., 2018). To the contrary (Daniel et al., 2016 and Malicha et al., 2017) reported as there is no significant difference between the adult and young age categories. The difference in prevalence of CBPP observed between adult and young animal was attributed to the fact that young animals do not move far away from home which decreases the chance of contact with other infected animals. In addition, young animals are susceptible to acute form of CBPP and may die and not get a chance for test. Moreover, c-ELISA is more sensitive in detecting cattle with chronic stage than any other test and the chance to miss individuals at early stage of infection is high.

Herds with history of movement revealed higher prevalence (11.49%) than herds not move place to place (5.114%). Animal movement indicates significant risk factors ($P < 0.05$). The prevalence seemed to increase significantly with the level of animals' movement. The result is similar with (Radostitis et al., 2010). CBPP is a contagious disease so that animal movement increases the chance of contact with infected animal.

Even though, the prevalence is higher in male (10.61%) than female (8.74%) animals; the difference is not statistically significant ($P > 0.05$). This finding corresponds with reports of (Danie et al., 2016 and Fulasa et al., 2020). However, disagrees with findings done by (Malicha et al., 2017) who reports a significant difference between male and female in seropositive to CBPP. This absence of difference may be due to CBPP is mainly transmitted through aerosols and the chance of getting infection is equal for both sexes.

The current prevalence of 6.71%, 6.37% and 16.92% observed in small, medium and large herd size respectively and it shows significance difference. Higher prevalence was observed in cattle sampled from larger herd size as compared to those cattle tested from medium and small size herds. The finding is in line with the report of (Fulasa et al., 2020; Teklue et al., 2015). This is due to CBPP is transmitted by aerosols and has contagious nature which is increased as herd size increases.

In this study season is one of the risk factors and even though higher prevalence was observed in dry season (11.20%) and lower in wet season (7.26%). However, the difference is not statistically significant. Absence of the difference was because of the animals get infection during dry season could be seropositive in wet season and vice versa. As to body condition a prevalence of 10.84%, 9.64% and 7.23% were observed in poor, medium and good body condition respectively. In the present study there was no significance difference among body condition ($p > 0.05$) in the sero-status of the animals.

Table 1: Sero-prevalence and Associated Risk Factors of CBPP

Risk Factor	No of Examined animal	No of Test Positive	Prevalence (cELISA)%	Chi-Square	P-Value
District					
Arero	91	10	10.99	5.725	0.221
Dubluk	101	5	4.95		
Gomole	109	7	6.42		
Teltele	93	12	12.90		
Yabello	104	12	11.54		
Age					
Adult	321	40	12.46	11.198	0.001
Young	177	6	3.39		
Herd Movement					
No	176	9	5.114	5.52	0.019
Yes	319	37	11.491		
Sex					
Female	366	32	8.74	0.402	0.526
Male	132	14	10.606		
Herd Size					
Small	164	11	6.707	12.409	0.002
Medium	204	13	6.373		
Large	130	22	16.923		
Season					
Dry	250	28	11.20	2.308	0.129
Wet	248	18	7.258		
Body Condition					
Poor	166	18	10.843	1.341	0.511
Medium	166	16	9.639		
Good	166	12	7.229		
Total	498	46	9.24		

Results of multivariate logistic regression analysis to fit the model revealed that among the risk factors considered in the analysis Age, herd movement in search of water and feed during drought period and herd size of the animals had statistically significant effect on positivity ($p < 0.05$). The odds of cattle being positive for CBPP infection were 26.65 times greater in adult to the odds of young cattle. The odds of sero-prevalence were significantly higher by 6.15 times in animals with history of herd movement than in animals without herd movement during drought period. The odd of positivity for CBPP in large herd size were about 3.21 times higher than that of small herd size animals (Table 2).

Table 2: Result of Multivariate Logistic Regression Analysis of Sero-prevalence of CBPP

Risk Factor	Odds Ratio	Std. Err.	Z	P>Z	(95% Conf. Interval)
Age					
Adult	26.652	24.778	3.530	0.000	(4.309 - 164.853)
Young	Reference				
Herd movement					
Yes	6.154	5.269	2.120	0.034	(1.149 – 32.959)
No	Reference				
Herd size					
Large	3.214	1.295	2.900	0.004	(1.459 - 7.081)
Medium	0.918	0.395	-0.200	0.842	(0.394 - 2.135)
Small	Reference				
Constant	0.004	0.004	-5.650	0.000	(0.001 - 0.025)

3.2 Questionnaire Survey Result

From questionnaire survey the major livestock diseases described by respondents in the study area were Contagious bovine pleuropneumonia (CBPP), Foot and mouth disease (FMD), Blackleg, Pasteurellosis and Trypanomiasis (TRYPS). The top three diseases were ranked in each district (Figure 1).

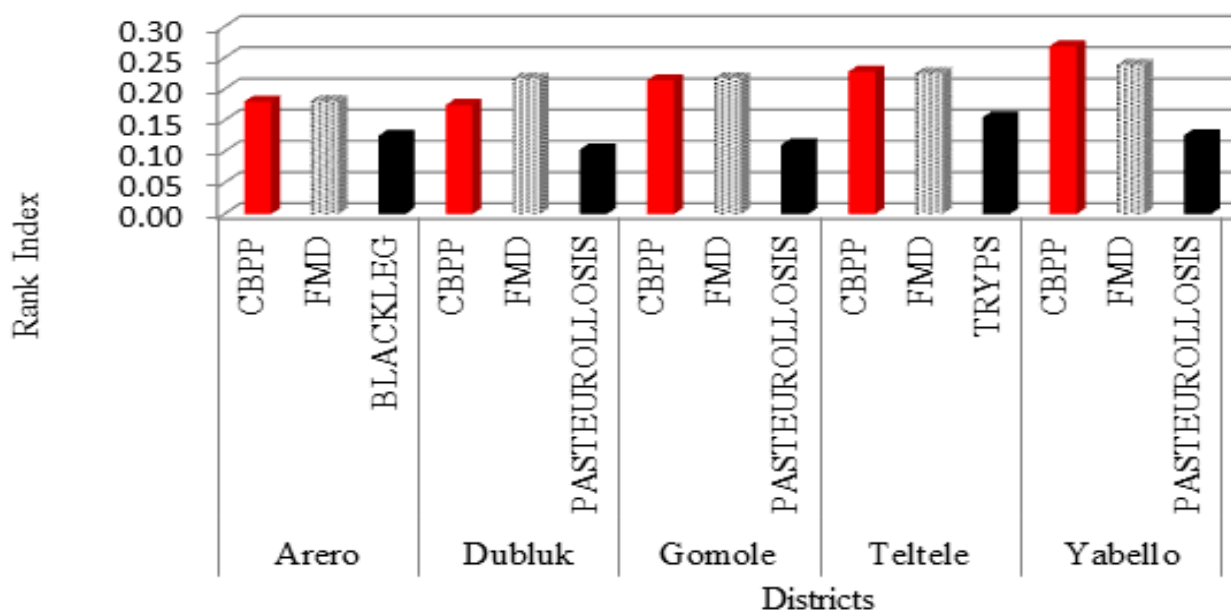


Figure 1: Top three diseases ranked in each district

Most of cattle owners in the study area were not practice separation of diseased animals from healthy herd to control the transmission of the disease. Some of them kept in separate house to treat the diseased

animals; while a few of the respondents kept near the cattle barn and in the compound. This indicated that cattle owners in each district need awareness on disease control (Figure 2).

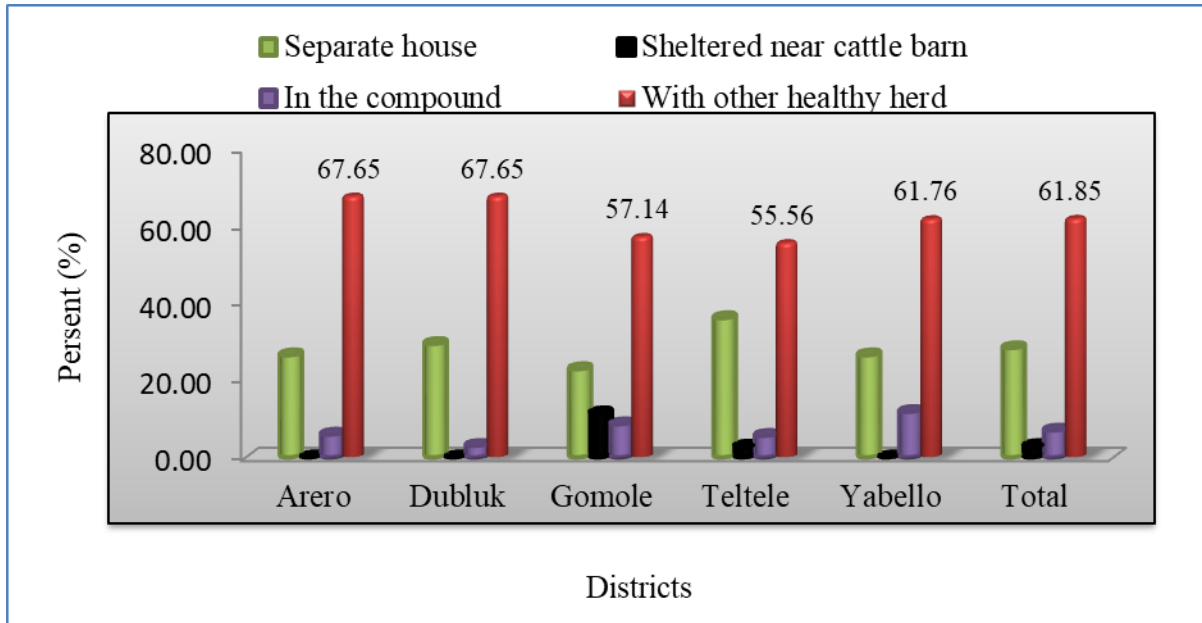


Figure 2: Practice of pastoralists towards managing animal showing clinical symptom

3.3 Geo-spatial prevalence of CBPP

Spatial analysis can be a useful tool in epidemiology and able to add considerable value and insight into animal health problems and their relationship with the physical environment. Geospatial techniques are the state-of-the-art techniques in epidemiology to identify the spatial trends of a disease. It is hard to think of an epidemiological investigation without location being at least inferred. In the current study we have tried to infer the location where animals positive to the disease were found. The GPS coordinates taken for positive animals were shows that there is no difference in spatial distribution of the disease among the study districts (Figure 3).

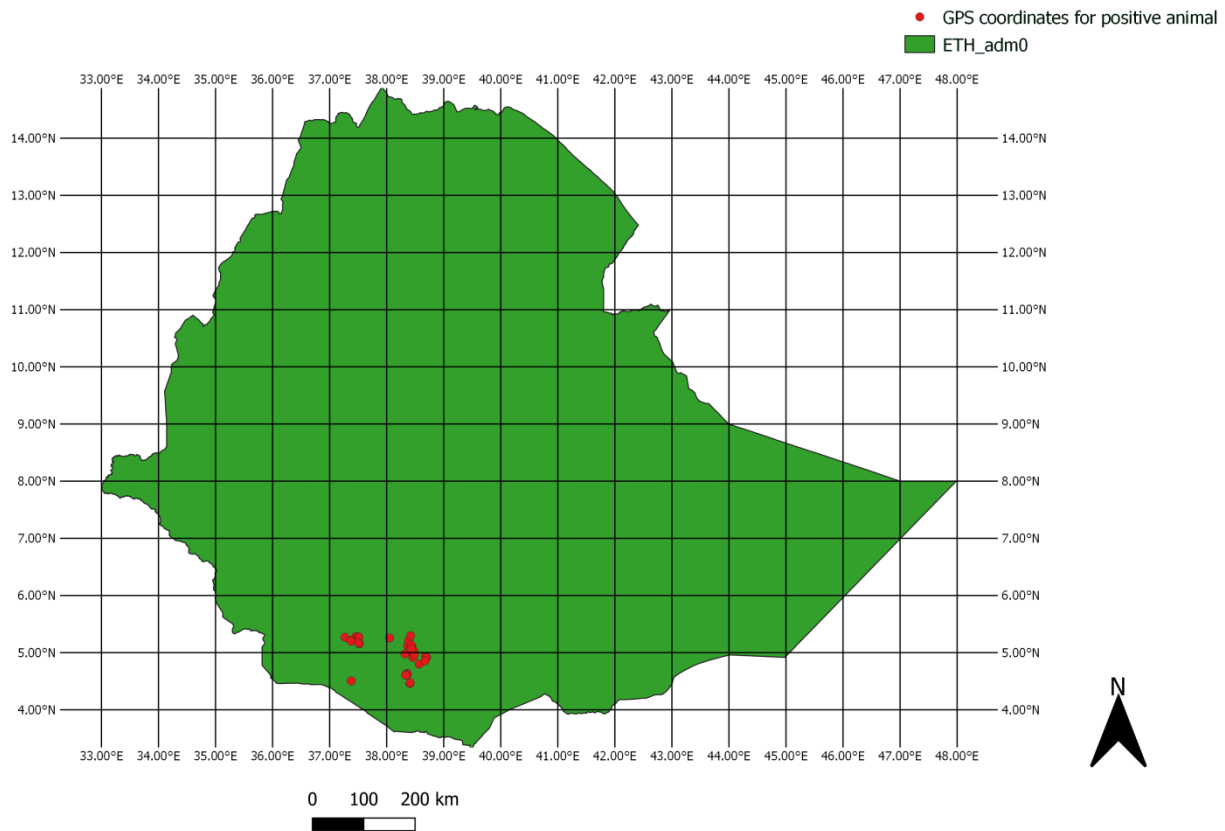


Figure 3: Map of Geo-spatial prevalence of CBPP in the study area

From questionnaire survey most of the respondents has traditional knowledge about the disease. However the practice they use in disease control is not good. For instance they keep infected animals with health.

Conclusion and Recommendations

Based on questionnaire survey and serological test; the present study indicated that CBPP is prevalent in the study area. Age, body condition and animal movement were found to be risk factors of sero-prevalence of CBPP in the area. Shortage of awareness on mode of transmission of CBPP and prevention methods to decrease disease transmission has seen from cattle owner. Based on these findings; the following recommendations are forwarded:-An integrated control and prevention approach; vaccination coupled with strict sero-surveillance and monitoring of CBPP with GIS based work is warranted, Strengthening of the community awareness about the disease, source, transmission and prevention, Restriction of illegal animal movement especially on borders to prevent the spreads of the disease, Animal feed should get attention as animals with good body condition relatively show low prevalence and Age of the animal should be considered during control and prevention

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Conflict of Interest: The authors have no conflicts of interest regarding this work.

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Prevalence and Risk Factors of Brucellosis in Goats in Borana Pastoral Area, Southern Oromia, Ethiopia

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Abstract

Goat production plays an important role in the livelihood of Borana pastoralists next to cattle husbandry. Optimal utilization of the goat population, is however, impaired by diseases such brucellosis. Brucellosis is considered one of the serious diseases incurring considerable loss to goat industry through reproductive wastages. The situation of brucellosis has not been investigated in goats in Borana pastoral areas despite the frequent occurrence of abortion. This study, conducted from November 2016 to April 2017, aimed at determining the prevalence of infection with Brucella organisms and identify the risk factors of infection.. Sera samples were collected from a total of 789 goats from three randomly selected pastoral districts. The samples were tested using competitive enzyme-linked immunosorbent assay (cELISA). Out of a total of 789 sera samples tested 137 (17.36%; CI: 14.78, 20.19) were positive for anti-Brucella antibodies. The highest prevalence was observed in Elwoya (71/252; 28.17%; CI: 22.71, 34.16) followed by Moyale (48/332; 14.46%; CI: 10.86, 18.71), whereas the lowest prevalence was observed in Yabello district (15/208; 8.78%; CI: 5.29, 13.52). The results of multivariable logistic regression analysis revealed that age and sex of goats were significantly associated with prevalence of brucellosis. The odds of infection was nearly 7 times higher in female goats than in males ($P < 0.001$). Adult goats are 12 times more likely to be infected than their younger counterparts ($P < 0.001$). For goats raised in large sized flocks ($OR = 2.57$; $P = 0.028$) and for those goats originated from Elwoya district the risk of infection was significantly higher ($OR = 7.91$; $P < 0.001$). The history of occurrence of reproductive problems in female goats is significantly associated with seropositivity to Brucella infection ($OR = 5.32$; $P < 0.001$). This study showed that significant proportion of goats in Borana pastoral districts were infected with Brucella organisms suggesting its economic implication and zoonotic importance.

Keywords: Brucellosis, Borana, Ethiopia, Goats, cELISA, Risk factors, Sero-prevalence

Introduction

Goats are important domestic animals capable of surviving in harsh environments such as common in arid and semi-arid areas (Tharwat and Al-sobayil, 2017). In Borana pastoral areas, they are the second most important livestock species and serve the society mostly as source of cash and collaterals for the family to cover school fees for children and other family expenses (Teshome et al., 2018). The optimal utilization of goats, however, is hampered by infectious diseases. One possible disease is brucellosis, which has been

recognized as one of the neglected tropical zoonotic diseases and of worldwide public health importance (Olufemi et al., 2018). It is a sub-acute or chronic disease caused by *Brucella* species (Gondal et al., 2017). In livestock it is mainly caused by *Brucella abortus* (*B. abortus*), *Brucella melitensis* (*B. melitensis*), *Brucella suis* (*B. suis*), *Brucella canis* (*B. canis*) and *Brucella ovis* (*B. ovis*). Among these species, *B. melitensis* and *B. ovis* are the common cause of brucellosis in sheep and goat (Tekle et al., 2019). Although several species of *Brucella* can infect goats, *B. melitensis* is the primary cause of brucellosis (Constable et al., 2017). There are three biovars of *B. melitensis* that have differing geographic distribution, but no difference in pathogenicity or host preference. Brucellosis caused by *B. melitensis* is regarded as a neglected but very important disease of livestock and humans in developing countries (Constable et al., 2017).

Brucella melitensis infection has major veterinary and human importance in areas where it occurs and causes considerable economic losses associated with abortion, neonatal death (Rajala et al., 2016), reduced fertility and decreased milk production (Ran et al., 2018). The considerable costs of preventive programs and restriction on trade of animals and their products constitute an important loss to infected countries. In addition, there is huge loss associated with human illness (Constable et al., 2017).

In Ethiopia, the occurrence of brucellosis in goats has been known for long time. Conservatively the overall prevalence was estimated to be about 5.3%; 95% CI = 3.5, 7.5 (Tadesse, 2016). The involvement of *Brucella* species among goat herds has also been confirmed using bacteriological methods (Tekle et al., 2019). The nationwide average prevalence, however, does not reflect the situation in the pastoral areas since pastoralism is a distinct production system from mixed crop livestock farming. Within the pastoral areas itself depending on the local husbandry system, the prevalence can vary. For example, prevalence of 1.7% was observed in Somali pastoral region of Ethiopia (Lakew et al., 2019) whereas prevalence of 4.8% was reported in goats in Afar pastoral region (Ashenafi et al., 2007). In one study comparing seroprevalence of brucellosis in goats in Somali and Afar regions prevalence of 13.2% was recorded in Afar region where commingling of animals due to communal grazing is the common practice. In contrast in Somali region where herding and rangeland utilization is based on clan basis, the prevalence was 1.7% (Teshale et al., 2006). In the Borana pastoral area, however, except the reports on the occurrence of *Brucella* infection in goats information on factors affecting its occurrence is scarce. The previous studies employed mostly CFT and other screening tests such as RBPT and indirect ELISA (Currò et al., 2012; Mohseni et al., 2017). An alternative diagnostic approach is the competitive ELISA (Yahaya et al., 2019). There is empirical evidence of frequent occurrence of unconfirmed cases of abortion and stillbirth in goats in the area. Most of those cases are being handled by farmers and community animal health workers. Hence, reliable information is needed on the disease prevalence and the risk factors to reduce the veterinary and public health impacts of brucellosis in goats. This study was conducted to estimate the prevalence of brucellosis in goats and identify its risk factors in Borana pastoral area using competitive ELISA.

Materials and Methods

Description of the Study Area

The study was conducted in three randomly selected districts of Borana pastoral zone namely Elwoya, Moyale and Yabello. Borana is characterized by a semi-arid to arid climate (Kamara et al., 2005; Haile et al., 2011). Geographically it is situated between 4° to 6° N latitude and 36° to 42° E longitude. The altitude of the Borana zone ranges from 1,000 to 1,700 meters above sea level featured by isolated mountains and valleys (Coppock, 1994; McCarthy et al., 2002). Elwoya, Moyale and Yabello districts are located at a distance of 590, 770 and 570 km South of Finfine (capital city of the Country), respectively (Teshome et al., 2018). The mean annual rainfall of the area ranges from 250 to 700 mm. The mean annual temperature varies from 19 to over 25°C. Extensive pastoralism (nomadic pastoralism) is the main means of livelihood for the Borana people (Gelagay et al., 2007). Cattle, goats, sheep and camels are important livestock species raised in the area.

Study Design and Sampling Strategy

A cross-sectional study was employed to collect sera samples and information on potential risk factors such as age, sex, parity of animals; history of occurrence of reproductive problems and herd size. The age of animals was estimated based on information obtained from the owner and dentition (Abebe and Yami, 2008). A multistage clustered sampling method was employed in which the study districts were selected randomly. This was followed by random selection of villages among all villages registered under the selected districts. Within the selected villages 161 households who have goats were purposively selected and all goats greater than 3 months of age were sampled if the herd size was ≤ 5 . For households having more than five goats 4 to 9 goats were selected per household. Accordingly a total 789 goats were sampled from the 161 households selected. Questionnaire was administered to the households to collect information on risk factors of brucellosis.

Blood Sample Collection

Approximately 5-7 mL of blood was collected from the jugular vein of each goat for using sterile plain vacutainer tubes and needles. The tubes were labeled individually and were kept on ice and transported to Yabello Pastoral and Dryland Agriculture Research Center. The samples were allowed to stand overnight to allow serum separation. The sera were separated after centrifugation at 1500 g for 10 minutes. The sera were then collected into sterile cryogenic tubes and stored at -20°C until transportation to National Animal Health Diagnostic and Investigation Center, Sebeta, Ethiopia for further analysis.

Laboratory Analysis of the Samples

Commercial competitive ELISA (SVANOVA Biotech, *Brucella* Ab c-ELISA, serial number: 10-2701-10, Uppsala, Sweden) was used for detection of anti-*Brucella* antibodies in the samples according to manufacturer's instructions. Positive and negative controls provided along with the kit were used for validation of the assay. A sample was considered positive to anti-*Brucella* antibody when its percent inhibition (PI) was ≥ 30 ; which was computed as $100 - ((\text{mean OD}_{\text{samples}} \times 100) / (\text{mean OD}_{\text{conjugate control}}))$. The c-ELISA, used in this study has a sensitivity of 93.6% and a specificity of 99.4% in goats (Nielsen et al., 2005).

Data Management and Analysis

The analysis of data such as the effects of risk factors on the prevalence of brucellosis was carried out using STATA software version 13.0 (StataCorp, 4905 Lakeway Drive, College Station, Texas 77845

USA). Multivariable logistic regression was used to determine the associations between the risk factors and prevalence. Odds ratio was reported as the measure of association between prevalence and risk factors. The effect of clustering was assessed using multilevel mixed-effects generalized linear models. A 95% confidence interval and a P value < 0.05 were considered significant.

Results

From a total of 789 goats sera tested 137 (17.36%; CI: 14.78, 20.19) of them were found positive (Table 1). The prevalence of brucellosis was 28.17% (CI: 22.71, 34.16), 14.46% (CI: 10.86, 18.71) and 8.78% (CI: 5.29, 13.52) in goats tested from Elwoya, Moyale and Yabello districts, respectively. Female goats were more frequently infected with *Brucella* species (20.19%) than their male counterparts (11.42%). The odds of infection was nearly 7 times higher in female goats than in males (P < 0.001). Similarly the prevalence was higher in adult goats (26.17%) than in younger ones (8.07%). Adult goats are 12 times more likely to be infected than their younger counterparts (P < 0.001). Higher prevalence was observed in goats sampled from larger herd size compared to those goats tested from medium and small size herds. The prevalence seemed to increase with the parity level of animals and in those goats having history of reproductive problems. For goats raised in large sized flocks (OR = 2.57; P = 0.028) and for those goats originated from Elwoya district the risk of infection was significantly higher (OR = 7.91; P < 0.001). The history of occurrence of reproductive problems in female goats is significantly associated with seropositivity to *Brucella* infection (OR = 5.32; P < 0.001). When univariable logistic regression was used to analyze the data parity number was significantly associated with prevalence of *Brucella* infection (OR = 1.43; P = < 0.001; CI: 1.17, 1.73). The prevalence was 11.19% (16/143), 18.19% (27/146), 25.40% (32/126), 26.67% (24/90) and 30.00% (9/30) in nulliparous pregnant goats, in primiparous goats, in goats having two parity, three parity and four parity, respectively. In multivariable logistic regression analysis, however, its effect was reversed due to the occurrence of multicollinearity with age. It was, therefore, omitted from the multivariable logistic regression model used.

Table 1: Results of multivariable logistic regression analysis on the effects of risk factors on prevalence of brucellosis in goats in Borana zone.

Risk factors	N tested	N positive	Percent OR	95% CI	P value
District					
Yabello 205	18	8.78	Ref.		
Elwoya 252	71	28.17	7.91	3.24 - 19.29	0.000
Moyale 332	48	14.46	1.64	0.66 - 4.08	0.289
Sex					
Female	535	108	20.19	6.97 2.72 - 17.87	0.000
Male	254	29	11.42	Ref	
Age					
Adult	405	106	26.17	12.19 3.55 - 41.86	0.000
Young	384	31	8.07	Ref	
Flock size					
Small	175	24	13.71	Ref	
Medium	267	33	12.36	0.96 0.39 - 2.33	0.920
Large	347	80	23.05	2.57 1.11 - 5.98	0.028
History of reproductive problems					
Yes	299	84	28.09	5.32 3.14 - 9.00	0.000
No	440	53	10.82	Ref	

Discussion

Goats remain valuable resources for the pastoral community inhabiting fragile and marginal lands. Optimum utilization of this resources requires control of infectious diseases such as brucellosis, which is highly communicable resulting in considerable economic loss due to reproductive wastages (Rashid et al., 2017) and a risk for public health. The present study documented serological evidence of brucellosis in goats and level of awareness of brucellosis in occupationally exposed household members in three selected districts of Borana pastoral region in Southern Ethiopia. Since we employed competitive ELISA, which is more sensitive, more specific and discern *Brucella* infection from cross-reacting bacteria, the results reflect the status of brucellosis in goats in the area. The prevalence observed is considerable ranging 8.78% in Yabello to 28.17% in Elwoya districts. The livestock authorities, veterinary services and public health sectors should take this into consideration.

The high seroprevalence observed shows the widespread occurrence of brucellosis in goat population in an area where there is no control program in place. It is comparable to the results of authors who reported brucellosis in goats from different parts of Ethiopia such as Teshale et al. (2006), Ashenafi et al. (2007) and Tsehay et al. (2014) who reported seroprevalence in goats in the range of 15 -16.45% in pastoral areas. It is also comparable to seroprevalence observed (16.2%) in goats from Sudan (El-Ansary et al., 2001), 16.1% from Nigeria (Bertu et al., 2010) and 18.70% from China (Wang, 2012). The higher seroprevalence observed in this study similar to the previous authors could be due to similarity in the animal husbandry practiced in the study areas. The pastoralists often mix animals of different ages and species in communal grazing areas and during night enclosures. Such activities favor transmission and

spread of *Brucella* among animals. The absence of control measures could also be another factor contributing to high seroprevalence. McDermott and Arimi (2002) also referred to the occurrence of a great variation in the prevalence of brucellosis in sub-Saharan Africa (ranging from 4.8 to 41%) in the pastoral areas. However, the current seroprevalence is lower than the reports of Al-Majali (27.7%) and Hamidullah et al. (34.88%) in Jordan and Bale et al. (2003) who reported seroprevalence of 34.8% and that of Ojo et al. (2007) who reported prevalence of 45.75% in Nigeria. In contrary, our report is higher than the findings of Ashenafi et al. (2007), Megersa et al. (2011) Adugna et al. (2013) Dabasa et al. (2013), Tegegn et al. (2016) and that of Teferi and Yeshibelaw (2019). The difference in seroprevalence could be attributed to difference in animal husbandry, disease control activities and the types of laboratory tests used for the detection of evidence of infection.

The high prevalence of brucellosis observed in adult goats in this study is due to the biology of the *Brucellae*, which is associated with the sexual maturity of the hosts. It is also partially attributed to the cumulative effects of age in which older animals are more likely to be exposed over lifetime than adults and young animals. That is, adult animals have greater chance of becoming infected and coming into contact with other animals resulting in continued exposure to *Brucella* as they remain in the herds over a long period of time serving as breeding stock. In the pastoral production system young animals are sold for immediate family expenses while adult animals especially females remain to produce offsprings. This observation has also been reported in Ethiopia and elsewhere in the world (Bertu et al., 2010; Radostits et al., 2010; Zubairu et al., 2014; Asmare et al., 2013; Olufemi et al., 2018; Edao et al., 2020). In addition, it is adult animals which move frequently from place to place during the dry season in search of pasture and water. This increases the chance of contact with other animals and the chance of infection. However, this finding is in contrast with the findings of Wubishet et al. (2018).

The statistically significant difference observed in the prevalence of brucellosis between female and male goats is probably due to the production of a sugar erythritol, which stimulates the growth and multiplication of *Brucella* organisms during consequent pregnancies (Radostits et al., 2010). This observation is in agreement with the reports of Rahman et al. (2011). Interestingly, in female goats the occurrence of reproductive problems is associated with infection with *Brucella*. Reproductive loss due to abortion, birth of weak offspring, and infertility are recorded as the common clinical signs of brucellosis in infected hosts. In this study, seropositivity to *Brucella* infection in goats was significantly associated with history of reproductive problems. Although the proportion of *Brucella* infected goats developing reproductive problems remains to be explored in the future, the observed association reveals the possible effects of brucellosis on the optimal utilization of the goats by pastoralists and the nation. Our observation is, however, in contrast to the findings of previous authors (Teshale et al., 2006; Ashenafi et al., 2007; Junaidu et al., 2010; Adugna et al., 2013; Sintayehu et al., 2015; Olufemi et al., 2018 and Wubushet et al., 2018).

In general, the distribution of anti-*Brucella* antibodies among different districts and herds was found to be variable. This could be associated with variability of the herd sizes and the geographical locations of the districts. Borana pastoralists trek their livestock, with the exception of lactating and few pregnant animals, to different districts, or even crossing national borders by traveling several kilometres in search of pasture, water and sometimes market. This results in massive concentration of animals in areas with

relatively better pasture and watering points. This in turn, may contribute to the increased transmission of *Brucella* organisms among different herds and districts.

Conclusion and recommendations

The present study documented high prevalence of brucellosis (17.36%) in goats in Borana pastoral area suggesting its widespread occurrence. This may result in considerable economic loss in terms of reproductive wastages like abortion, still birth, infertility, sterility and delivery of weak offspring. In addition to this it has a public health importance. Location, age, sex, history of reproductive problem and flock size were risk factors of brucella infection. Ethiopian government should institute brucellosis control measures and possible eradication strategies as well as awareness creation for the community on the economic and public health implication of brucellosis for the contribution to successful prevention and control of the disease.

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Compliance with ethical standards

Ethical clearance on the use of goats for this study was obtained from animal research ethics review committee of Addis Ababa University, College of Veterinary Medicine and Agriculture, before the start of this study. All procedures performed in this study were in accordance with the ethical standards of the institution. The owners of goats used in this study and the local administration were informed about objectives of the study and the owners revealed their consent in the presence of administrative bodies and elders.

Conflict of Interest: The authors have no conflicts of interest regarding this work.

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Apiculture Research Results

Assessment of pesticides use and its effects on honeybee (*Apis mellifera* L.) colonies in some selected districts of West and Kellem Wollega Zone, Western Ethiopia

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ABSTRACT

Honeybees are a fundamental part of our agricultural system and their presence contributes to food security via providing income and increase crop yield and quality through pollination. However, today unwise applications of various pesticides are killing a number of honeybee colonies annually and thus have become critical in the developments of the apiculture subsector. This study was proposed to assess and evaluate the impacts of commonly used pesticides on adult honeybee health. Sayo, Dale Wabara, Guliso, and Nedjo districts were purposively selected Zonal agricultural bureau which is based on the potential of beekeeping activity, crop production and pesticide application. Household survey was conducted and commonly used pesticides were tested for their toxicity effects on honeybees in laboratory via feeding, fumigation and contact method. From the total 177 respondents, 98% used pesticides for their crop production. And Glycel 41% SL, Glyweed 48% SL, Roundup 360% SL, Diazinon 60% EC, Mancozeb 80% WP and 2, 4- D were the most commonly used pesticides while their toxicity effects were compared with a standard positive control (Dimethoate) and negative control (Water). All the applied Pesticides showed toxic to the experimental adult honeybees within a 24 hours exposure via all testing methods (Feeding, Contact and Fumigation) as compared with controls. Therefore, proper utilization is significant from poison of bee.

Key Words: Contact, Feeding, Fumigation, Honeybee, Pesticides

Introduction

With about seven million honeybee populations, Ethiopia's annual honey and beeswax production is estimated to be over 54,000 and 5000 tons, respectively (MoA, 2013). With this, the country is ranking ninth highest honey producer in the world and the leading producer of honey and beeswax in Africa (CIAFS, 2012).

Honey bees are an essential part of our agricultural system, as many other species of pollinators. The value of honey bee pollination services to U.S. Agriculture has been estimated to be greater than 14 million dollars (Morse, 2000) with their value topping \$215 billion worldwide (Gallai, 2008). More than three-quarters of all flowering plants must be pollinated by an animal visitor; usually an insect (Klein *et al.*, 2007). In addition, it often takes several floral visits by pollinators to ensure maximum fruit set and quality. The increased use of pesticides, reduction in the number of wild colonies, and the increased value of both bees and the crop they pollinate have all added to the importance of protecting bees from pesticides (Rader *et al.*, 2009).

The use of agro chemicals for crop pests, weeds, mosquitoes and household pests control brings into focus the real possibility of damaging the delicate equilibrium in the colony, as well as the contamination of hive products (Kerealem *et al.*, 2009). In addition, bee colonies are maintained for their honey and wax production. Potential exposure of bees to pesticides can vary greatly depending on the type of pesticide, formulation, application method, label restrictions, and other factors (Ellis *et al.*, 2014). The goal in using a pesticide is to achieve maximum benefit (success) with minimum negative impact, and these factors should always be considered in pesticide selection.

The introduction of pesticide in Ethiopia to control agricultural pests' dates back to the 1960's (EPA, 2004). Although the volume fluctuates across the pesticide types, the country on the average imports 3346.32 metric tons of pesticides annually (Gizachew Assefa, 2011). Using pesticides is widely spread following modern agriculture and areas with high crop farming parts of Ethiopia are yearly receiving different types and amounts of pesticides.

Today unwise application of herbicides and various pesticides are killing a number of honeybee colonies annually and thus have become critical in the developments of the subsector. By selecting the least options and applying them when pollinators are not present, harm can be minimized (Medrzycki *et al.* 2013). Creation of awareness, having pre warning regulation and strong coordination among beekeepers, chemical applicators and crop growers are very essential to minimize the effect of honeybee poisoning. There is evidence that extensive use of pesticides in agricultural practices has resulted in presence of residues in honey and wax and this changes our honey products from organic into inorganic and decreases the foreign income of the country.

Honeybees can be exposed to agro-chemicals through: - Direct contact during foliar applications, contact with residues on the plant surface after foliar application, ingestion of residue in nectar and pollen or vapor drift (Gizachew Assefa, 2011). Whenever apicultural trainings are given, the primary concern that beekeepers of the study area always raise are the increasing and indiscriminate use of agrochemicals and bee forage resources. However, the information of agrochemicals utilization and hazardous effects to the local honeybee in the study area has been limited. Therefore, this work was proposed with the aim of identifying the commonly used pesticides in the study area and evaluating their toxicity effects on adult honeybees in the laboratory.

Materials and Methods.

Scope of the study area

The study was conducted in Kellem Wollega (Sayo and Dale Wabara district) and West Wollega Zones (Guliso and Najo) were selected (Figure 1). From each district three Peasant associations were purposely selected based on beekeeping potentiality and intensive pesticides application for crop production. From each Peasant associations, beekeeping technician experts and fourteen model farmers (beekeepers and crop growers) were selected and interviewed on pre structured questionnaires for their attitudes and knowledge of chemicals used in the area and impacts on bee colonies as well as hive products in general. In addition pesticides applicators were interviewed whether they are using chemicals according to the prescription labeled and recommended doses. Farmers' data collection check lists were developed and farmers' attitudes or perceptions were recorded through individual and group interviews or discussion at each Peasant association of the area.

Additionally, pesticides and veterinary drug shops were interviewed on the sources and supply of the chemicals. Also the prescription labeled on chemical containers and their legal registrations were assessed for its safety. Finally toxicity of the most commonly used chemicals were tested under laboratory

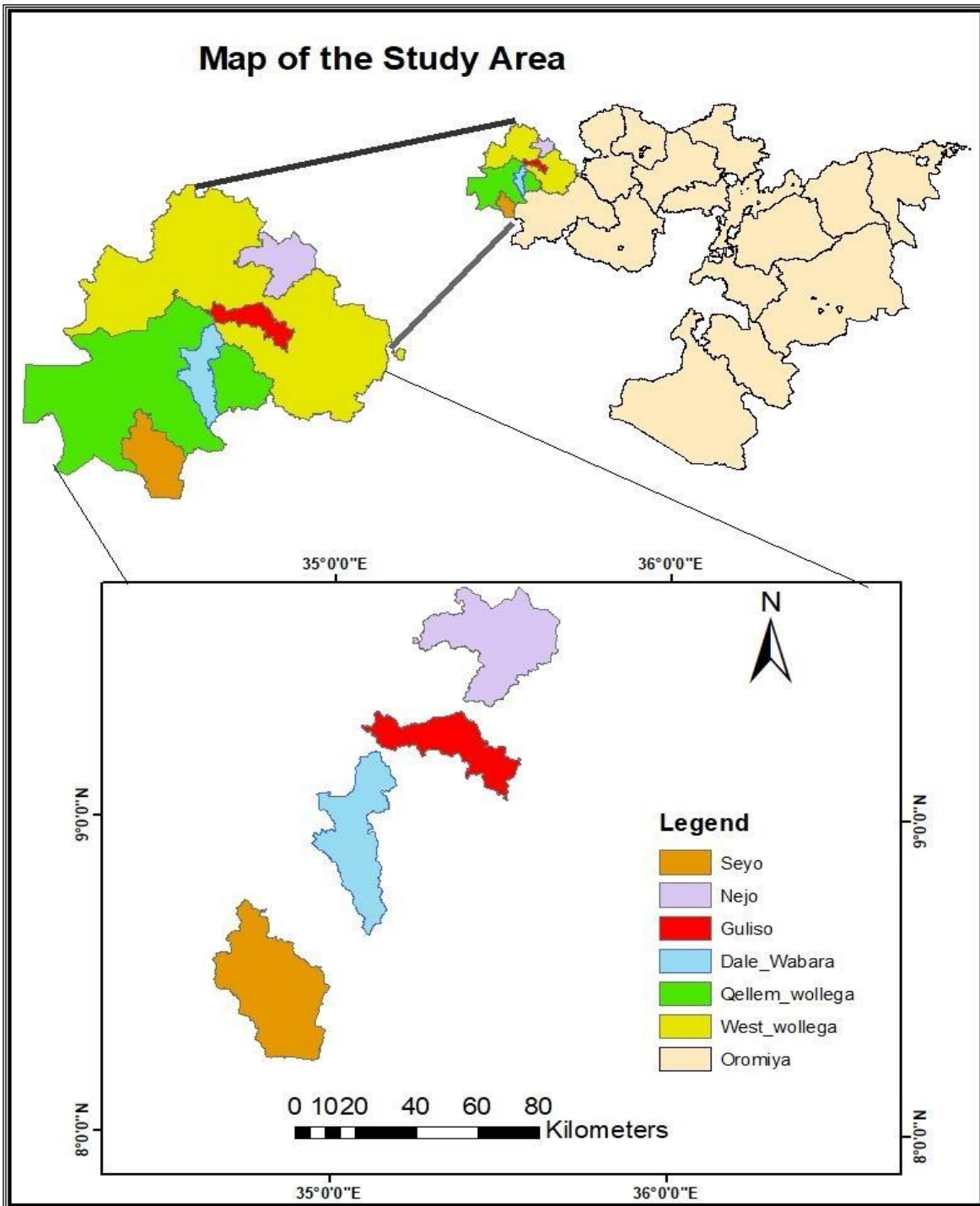


Figure 1: Map of the study area

Laboratory analysis

Standard methods for toxicology research in *A. mellifera* were used to test the toxicity (Medrzycki *et al.* 2013). Acute toxicity of widely used pesticides identified during survey works were tested in the laboratory. For this, 30 (thirty) healthy adult worker bees, collected from frame without brood were anesthetized with CO₂ and held in well ventilated laboratory cages (5.5 x 8.5 x 10 cm) and placed in 25 ± 2°C temperature and 60-70% of humidity over study periods. The acute toxicity of these agro-chemicals to honeybees was tested through feeding, contact and fumigation. The concentration of each test pesticides causing 50% death of experimental bees and degree of toxicity hazard were determined. The mortalities caused by individual pesticides were also compared with positive and negative controls and amongst pesticides.

Table 1: Field recommended concentrations of frequently used agrochemicals in the study area

Common name	Types	Recommended concentration
Glycel	Herbicides	0.5ml/31.25ml H ₂ O
Diazinon	Insecticides	0.5ml/50ml H ₂ O
2, 4-D	Herbicides	0.5ml/80ml H ₂ O
Glyweed	Herbicides	0.5ml/40ml H ₂ O
Round up	Herbicides	0.3ml/50ml H ₂ O
Mancozeb	Fungicides	1gm/500ml H ₂ O
Dimethoate	Insecticides	0.1ml/30ml H ₂ O

Feeding test

30 (thirty) pre-determined healthy worker bees were placed in laboratory cages and starved for up to 2 (two) hours before the beginning of the test. Bees were provided with 50% honey solution containing the recommended concentration of 10µg/bee (300µg/30bee) of each test Agrochemicals to determine the toxicity (OEPP 2010). Each treatment was replicated 3 times. Death of honeybees and injured honeybees were recorded in 15, 30 and 45 minutes, 1, 2, 4, 6, 12, 24, 48 and 72 hours and compared with 50% honey solution (non-toxic) and 0.3 µg of reference standard toxic chemicals, Dimethoate or E-605 Forte (i'e Parathion). Food consumption in each test has also been recorded and replenished every 24 hrs. (Gough *et al.* 1994).

Vapor or fumigation test

Thirty (30) health bees were held in Laboratory cage and placed over the petridish filled with the recommended concentration of each pesticide in three replications. The deaths of bees and injured honeybees were recorded in an hour interval for a maximum of three days. The death rate is compared with concentration of 0.0009% of the standard toxic chemicals (Dimethoate) and non-toxic control (petridish filled with water). All bees in the cages were fed 50% honey solution until the end of study (Gough *et al.* 1994).

Contact test

Filter papers were immersed in the recommended concentration of each test agrochemical and allowed to dry. These papers were enclosed separately in a lab cage containing 15 worker honeybees. Toxicity effects of each concentration of test materials were compared with 0.0009% standard chemicals E-605

forte (i.e. Parathion) and control (paper immersed in pure tap water). Each treatment was replicated three times (Gough *et al.* 1994). Finally in all laboratory tests percent of mortality caused by each agrochemical in each test was calculated using Abbott method (Abbott 1925) indicated below.

$$\% \text{ of mortality: Correct mortality (Abbott)} = \frac{\% \text{ mortality treatment} - \% \text{ mortality control} * 100}{100 - \% \text{ mortality control}}$$

Data analysis

Data means \pm SD were calculated using SAS Software (SAS Institute, 2003; 14). Significant differences in mortality rate against experimental workers with each acute toxicity test methods were obtained with one-way ANOVA, and means were separated with LSD tests.

Result and Discussion

Survey results

Different type of pesticides were identified by the survey conducted and they were Glycel 41% SL, Gillyweed 48% SL, Round up 360% SL, Coneo 41%SL, Glymax, Linkosate 48%SL, Diazinon 60% EC, Primagram, Ethiolathion 50%EC (Malathion), Mancozeb 80% WP, Aura72 SL (2, 4-D amaine salt), 2, 4-D zura® and Dimethoate which are concurrent with brands of pesticides listed by Dessalegn Begna (2015). This study showed that 85%, 8%, 6% and 1% of pesticides were from local market, local market and informal dealer (smuggler), informal dealers and national market respectively (table 2).

Table 2: Percent of market categories

Market categories	Percent (%)
National market	1
Local market	85
Informal dealers/smuggler	6
Local market and informal dealers	8
Total	100

From the total 177 respondents, 98% of the farmers are used agrochemicals for their crop production. And when purchasing agrochemicals, about 66.7 % of farmers were supplied with information on the pesticides such as pamphlets, or instructions describing safety issues or procedures (Table3); from this about 11% of chemical users cannot understand the instruction on the pamphlet or on the container. According to this assessment 7% and 12.8% of pesticide users apply pesticides at any stage of crop if pests occur and at vegetative and flowering stages respectively (Table 4). And 57.2% and 22.5% pesticides were applied during morning (7:00 am- 12:00 am) and afternoon (12:00 am – 4:00 pm) respectively (Table 5). This time is active time for honeybees and hence can cause death of honeybee foragers in the field, honeybees in the hive, and absconding there by lead to low honeybee products production.

Table 3: Percent of respondent supplied with information on pesticides

	Response	Percent (%)
Supplied with information on pesticides	Yes	66.7
	No	33.3

Table 4: Stages of crop pesticides are applied

Stages of crop	Percent (%)
Only at vegetative	72.7
At vegetative and flowering stage	12.8
At any stage if pesticide occurred	7
Only at vegetative and at seed setting	7.3

Table 5: Time of pesticide application

Time	Percent (%)
Before noon (7:00 am- 12:00 am)	57.2
Afternoon (12:00 am - 4:00 pm)	22.5
Any time of the day	20.3

Laboratory acute toxicity test result

Feeding test result

The cumulative mortality rate result within 24 hours of pesticides exposure through direct feed to adult honeybees was indicated in table 6 and figure 2 below. Dimethoate which is a toxic standard reference was used as a positive control while Water was used as a negative control for comparison with the tested pesticides. This study showed that pesticides were significantly differed ($P < 0.05$) in causing mortality to honeybees among themselves and negative control (water). However, Roundup and Diazinon caused statistically comparable toxicity effect ($p > 0.05$) with a positive control (Dimethoate) (Table 6) and killed 100% of the experimental adult honeybees within 24 hours exposure (Figure 2).

100% killing of Diazinon against the experimental sample adult *A. mellifera scutellata* L. through feeding was also observed by Dinku *et al.* (2020). Result of this study agrees with Dawit *et al.* (2015) and Atkins *et al.* (1981) found that Diazinon mixed in the honeybee food was highly toxic. The tested pesticides exhibits tumble, mortality and sub lethal to honeybees. From our study Mancozeb was the least (38.6%) killing which is followed by 2, 4-D (52%) as compared with the other pesticides but more toxic than water (8.6%). Even though the percentage killed higher than ours, Dinku *et al.* (2020) recorded 66.6% and 46.6% adult *A. mellifera scutellata* L. mortality rate by 2, 4-D and Mancozeb via feeding in the laboratory and toxic than negative control water. Moreover, Gebrecherkos (2019) has observed 2, 4-D took long 24 hours, to kill 94.9% of the experimental adult *A. mellifera jemenetica* as Dimethoate.

In contrary with ours and above researchers, Amssalu *et al.* (2012a) have observed a 2, 4-D is not toxic to adult central high land; *A. m. bandasii* when ingested with food. The toxicity variation between and within the agrochemicals might be due to environmental condition, temperature and honeybee race variations. For many agrochemical substances, a linear relation links ambient temperature and LD50s,

positively with some and negatively with others; hygrometry is also a factor of variation (Medrzycki *et al.* 2013).

Furthermore, the occurrence of different in toxicity by the same chemical against adult honeybees is investigated by different researchers. For instance; after emergence, the age-susceptibility relation is variable depending on the target species and toxin (Hodgson 2004). The weight of an individual is an important factor influencing the LD₅₀ and it is often negatively correlated with toxin susceptibility. The amount and quality of pollen ingested in the first days of life can affect the pesticide susceptibility of young and older worker bees independently of their weight (Wahl and Ulm 1983). Agro chemicals are fast ingredients acting at warm and humid air condition than cool climate condition (Dinku *et al.* 2020).

Table 6: Mortality rate of adult honeybees within 24 hours agrochemical exposure via feeding

Pesticides	N	Mean ± SD
Dimethoate	3	30.0±0a
Round up	3	30.0±0a
Diazinon	3	30.0±0a
Glyweed	3	22.3 ± 0.57b
Glycel	3	20.6± 0.57c
2, 4-D	3	15.6± 0.57d
Mancozeb	3	11.6± 0.57e
Water	3	2.6± 0.57f
LSD (5%)		0.82
CV		2.30

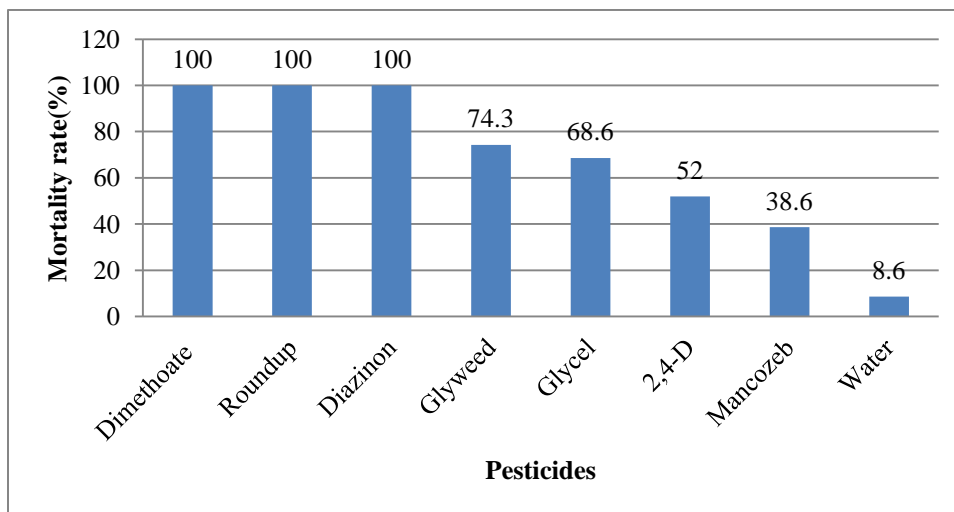


Figure 2: The percentage mortality rate of adult honeybees within 24 hours pesticide exposures via feeding

Contact test result

The killing effect of different pesticides against adult honeybees within a 24 hours evaluation via contact was indicated in Table 7 and Figure 3 below. A toxic standard reference (Dimethoate; positive control)

and non-toxic reference (water; negative control) were used to compare with the tested Pesticides. Statistically there was no significant ($p > 0.05$) mortality effect among the tested Pesticides and with a positive control (Dimethoate). Including standard toxic chemicals (Dimethoate), the tested Pesticides namely Round up, Diazinon and 2, 4-D killed 100% while Glyweed, Glycel and Mancozeb killed 97.3% of experimental adult honeybees within 24 hours and all showed significant ($p < 0.05$) with a non-toxic negative control water which killed 28.6% (Figure 3).

Similarly Dawit *et al.* (2015); Dinku *et al.* (2020) have reported the 100% honeybee mortality when contacted with Diazinon 60% EC. However, Dinku *et al.* (2020) made a different observation with less toxicity of 2, 4-D and Mancozeb 80% like a negative control which killed $< 20\%$ of the experimental adult honeybees via contact test. Furthermore, Gebrecherkos (2019) 2, 4-D and Mancozeb 80%WP didn't exhibit any significant difference in imposing *A. mellifera jemenetica* mortality when comparing with the negative non-toxic control (water) via contact mode of exposure.

Table 7: Mean mortality of adult honeybees within 24 hours agrochemical exposure via contact

Pesticides	N	Mean \pm SD
Round up	3	15.0 \pm 0a
Diazinon	3	15.0 \pm 0a
Dimethoate	3	15.0 \pm 0a
2, 4-D	3	15.0 \pm 0a
Glycel	3	14.6 \pm 0.57a
Glyweed	3	14.6 \pm 0.57a
Mancozeb	3	14.6 \pm 0.57a
Control	3	4.3 \pm 0.57b
LSD (5%)		2.02
CV		8.39

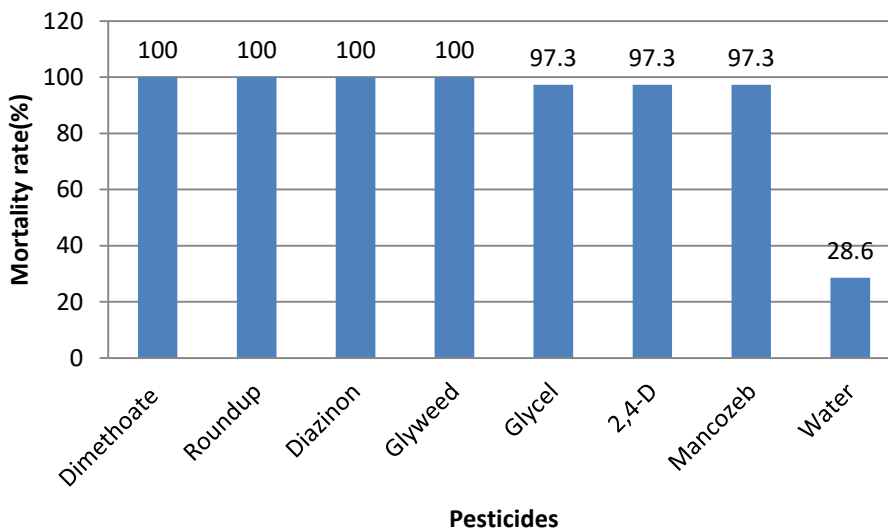


Figure 3: The percentage mortality rate of adult honeybees within 24 hours pesticide exposure via contact

Fumigation test results

Within 24 hours fumigation there was statistically not significant ($p>0.05$) among all the tested Pesticides (Table 8) and positive control (Dimethoate) and all killed 100% experimental adult honeybees (Figure 4). However, all Pesticides showed statistically a significant ($p<0.05$) toxic with a non toxic water which killed 14.3% of adult honeybees within 24 hours fumigation.

This study indicated that if these chemicals are fumigated against to the bees, they would be highly toxic which might be due to their fume property. The tested fumigant Pesticides may have great diffusion and penetration characteristics which bring them to penetrate into honeybees' body and kill them. According to Oregon Health Authority (2013), toxic agrochemicals spraying during windy conditions and using nozzles that create fine spray particles increase the risk of spray drift; high temperatures and low volatility also increase the risk of drift. Furthermore, May *et al.* (2015) drift can be reduced by using coarse sprays and by not applying pesticides during windy conditions and it is also important to consider the formulation carefully because water based and oil based sprays have different drift characteristics.

Table 8: Mortality rate of adult honeybees within 24 hours pesticides exposure via fumigation

Treatments	N	Mean \pm SD
Dimethoate	3	30.00 \pm 0a
Round up	3	30.00 \pm 0a
Diazinon	3	30.00 \pm 0a
Glyweed	3	30.00 \pm 0a
Glycel	3	30.00 \pm 0a
2, 4-D	3	30.00 \pm 0a
Mancozeb	3	30.00 \pm 0a
Water	3	4.3 \pm 0.57b
LSD (5%)		0.35
CV		0.76

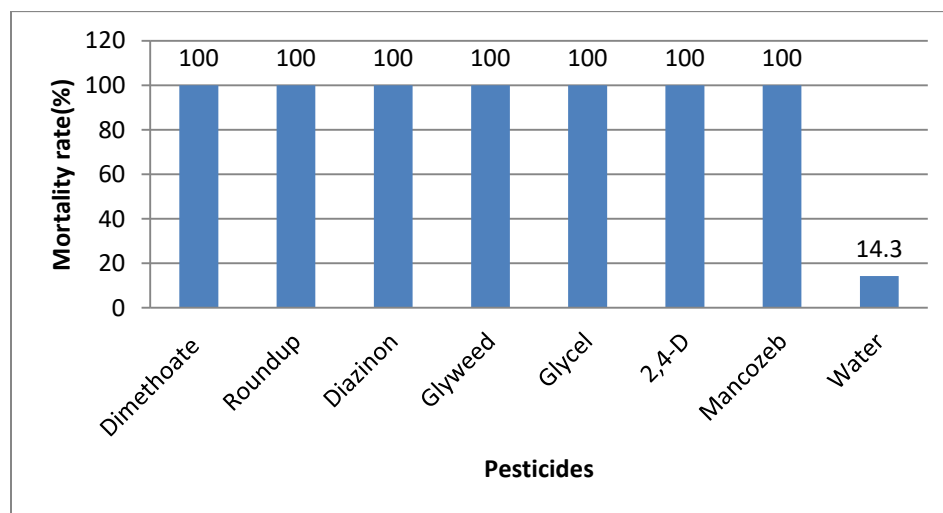


Figure 4: The percentage mortality rate of adult honeybees within 24 hours pesticide exposure via fumigation

Conclusion and recommendation

In the study area farmers extensively used thirteen different brands of pesticides at different stages of crop production including the flowering stage. Most pesticides were applied at an active time for honeybees. Based on the laboratory result, all Pesticides tested were found to be toxic to honeybees in all testing methods (feeding, contact and fumigation) as compared with the controls. Farmer beekeepers, crop growers and pesticide applicators should be trained on how to keep out the safety of bees from agrochemicals through integrated pest management, time of application, apiary site selection, the effect of pesticide not only on honeybees but also on hive products, proper pesticides handling, management, utilization, and appropriate safety precautions through the concerned staff.

Crop growers should be educated and demonstrated on the impact of insects in general and honeybees in particular about their pollination service on quantity and quality increments. Hence this would initiate them to weed by their hands and organic product would be harvested. This sector needs a governmental action in excluding the use of highly toxic pesticides to honeybees and to replace with less toxic. Further laboratory and field toxicity test work will be expected on the existing national wide pesticides against different local bee races.

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Physicochemical composition of honey (*Apis mellifera* L.) types Produced in West and Kellem Wollega Zone

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Abstract

Honey is a sweet natural product synthesized by honeybees from the nectar of bee flowering plants. Its physicochemical composition varies based on botanical, geographical, entomological and seasonal honey types. The aim of this study is, therefore, to compare the physicochemical properties of honey types collected from Kellem and West Wollega Zones of Western Ethiopia in different seasons. The pollen grain was analyzed by the Methods of Melissopalynology while physicochemical analyses of honey were determined using the Harmonized Method of the International Honey Commission. From the first season honey sample (FSHS), 87 % of total pollen count was that of *Guizotia* species hence it was *Guizotia* monofloral honey while 46% of *Coffea arabica* L. pollen was recorded from the second season honey sample (SSHS) which is also named as Coffee monofloral honey type. A significant reducing sugar difference ($p < 0.05$) was observed between *Coffea* honey and that of *Guizotia* honey the highs being in *Coffea* honey (80.00 ± 5.71). *Guizotia* honey is significantly more acidic with a pH of 3.34 ± 0.12 compared to *Coffea* honey (3.59 ± 0.05). The sucrose, moisture, free acidity, hydroxymethylfurfural (HMF) and ash contents between the tested monofloral honey types were not shown statistical differences ($p > 0.05$) and all found within the acceptable national and international standard. Therefore the honey samples harvested in the study area had acceptable physicochemical compositions.

Key words: *C.arabica*, *Guizotia*, Honey, Physicochemical

Introduction

Honey is the natural sweet substances synthesized by bees, from the nectar and secretion of living parts of plant. The bee collect nectar from the plant, deposit and reduce the water content, store and leave it in honey combs or honey pots to ripen and mature for their own consumption (Codex Alimentarius, 1989). Honey production has gone from traditional hunting to a more elaborate production stages. This has led to an increase in the availability of honey stock. According to FAO (2018) report 45,300 metric tons of honey is produced per annum in Ethiopia, makes the country to rank first honey producer in Africa and ninth in the world. It is a complex mixture and presents very great variations composition and characteristics based on geographical and floral types or the nectar foraged by bees (Saxena *et al.*, 2010). The composition and the quality of honey are strongly influenced by geographical and environmental factors (Jones *et al.*, 2011). They depend on the maturity of honey; the mode of production, climatic conditions, and treatment and storage conditions as well as source of nectar (Was *et al.*, 2011). A quality product will go a long way in developing the confidence that encourages return, customers and efficient production of a product to any marketing scheme (Geno, 2005). In marketing of honey, consumers should

have confidence that they are getting good quality for what they are paying so that the country able to earn foreign currency to revamp the national economy (Geno, 2005).

The quality of honey is a key factor for both local and international markets to enable attainment of competitive premium prices and ensure human health. Honey quality consideration is an aspect disregarded by producers and processors especially in developing countries. The quality of honey is often judged by few physicochemical factors. Good quality honey has low water or moisture content less than 20%, since higher proportion of water may enhance honey spoilage through fermentation. Hydroxymethyl furfural (HMF, 5-hydroxymethyl-2-furaldehyde) is a recognized indicator of reduced quality in numerous foods that contain carbohydrate (Rattanathanalerk *et al.*, 2005). The presence of HMF in levels higher than 40mg/kg is a sign of honey degradation through heating or long storage in hot condition (Morales *et al.*, 2009). pH- values have great importance during the extraction and storage of honey, as they influence the texture, stability and shelf life of honey (Terrab *et al.*, 2002).

Because of multiple importance of honey from food to medicine, it is of great interest to carry out complete analysis of honey and to formulate values ranges of various honey constituents, and characteristics. Honeys industries have shown great interest in these constituents as they influence the storage quality, granulation, texture, and flavor, nutritional and medicinal values of the honey. Honey is generally evaluated by a physicochemical analysis of its constituents. Acceptability of honey depends on its quality which can be assessed by among other things its physicochemical characteristics. The physicochemical properties of honey produced indifferent geographical location of Ethiopia have been reported by several researchers (Sisay Gobessa *et al.*, 2012; Gebreegziabher Gebremedhin *et al.*, 2013).

Honey is harvested two or three times annually in all the study areas depending on the availability of bee forages. In the late September or early November, herbaceous bee plants such as *Bidens* spp (Meskel flower or Adey Abeba in Amharic), *Trifolium* spp and *Plantago lanceolate* L. (Yebeglat in Amharic whereas literally Qorxobbii in Afaan oromoo) are the dominant species which release both pollen and nectar sources (Nuru Adgaba,2007). This makes the main honey harvesting season to be practiced at the first of December and honey type is said to be Meskel flower honey which is Monofloral (Literally DammaTuufoo in Afaan oromoo) at all studies areas. In addition to this, other honey harvesting season is practiced in February from *Vernonia amaygdalina* L. and *Coffee arabica*. Since honey composition predominantly varies based on the botanical origin, this research was initiated with the objective of analyzing the botanical origin and physicochemical qualities of honey types between the seasons of the study area.

Material and Method

Honey Sample Collection

The honey samples were collected from honey production potential of Sedi chanka and Haro Sebu district of Kellem Wollega Zone while Najo and Guliso district of West Wollega zone. From each district three representative kebeles, a total of 12 kebeles were selected for honey sample collection. For these work two different sources of honey samples of *A.mellifera* was used to determine the physicochemical composition between honeys harvesting season of the study area. The first source was the honey samples that was harvested in a month of October through November and labeled as the first honey harvesting season (FHHS), while the second source was the honey samples that was harvested in a month of January through February and labeled as the second honey harvesting season (SHHS). For each sources, 24 ripen

honey samples (two samples from each kebele) from each season, a total of 48 ripen honey samples from both sources of honey season were collected. The collected honey samples were brought to Holota Bee Research center Laboratory, using sterile glass cup honey containers for analysis.

Analyzing botanical origin of honey

Standard procedure by Louveaux *et al.* (1978) was used to analyze the botanical origin honey samples. Ten grams of honey was dissolved in 20 ml of warm distilled water in centrifuge tube at temperatures that ranged from 20-40°C and centrifuged at 3800 rpm for 10 minutes and the supernatant was decanted. Again 20 ml distilled water was added to completely dissolve the remaining sugar crystals and centrifuged at 3800 rpm again for 5 minutes and supernatant was removed completely. The sediment was spread evenly using a sterile micro spatula on microscope slide and the sample was dried for a while. Thereafter, one drop of glycerin jelly was added to the cover slip and the pollen was identified using pollen atlas (Nuru, 2002). The percentage of pollen types in each honey samples was calculated based on the total number of different types of pollen grains counted in each sample. If pollen grains counted was greater than 45 %, used as predominant pollen (monofloral honey), Secondary pollen (16-45%), important minor pollen (3-15%), and minor pollen (<3%) (Louveaux *et al.*, 1978). The pollen count was done under light microscope (Swift instrument international, serial number 8750038, Japan, high power 400x) linked to a computer. Physicochemical and pollen grain analysis of honey samples were done at Laboratory of HolotaBeeResearch Center.

Physicochemical properties

Moisture content

It was determined using an Abbérefractometer (ABBE- 5 Bellingham Stanley. Ltd, United Kingdom) that can be thermo stated at 20°C and regularly calibrated with distilled water. Honey samples were homogenized and placed in a water bath until all the sugar crystals were dissolved. After homogenization, the surface of the prism of the refractometer was covered with honey and after 2 minutes refractive index for moisture was recorded. The value of the refractive index of the honey sample was determined using standard table (Bogdanov, 2009).

pH and Free Acidity

Ten gram (10 g) of honey was dissolved in 75 ml of distilled water in a beaker and stirred using magnetic stirrer. The electrode of pH meter (METTLER TOLEDO, CHINA) was immersed in the solution and the pH of honey was recorded. For measurement of free acidity, the solution was further titrated with 0.1 M sodium hydroxide (NaOH) solution to pH 8.30. For precision, the reading to the nearest 0.2 ml was recorded using a 10 ml burette. Free acidity is expressed as mill equivalents or a mill mole of acid/kg honey and is equal to ml of 0.1M NaOH x 10.

Acidity =10 V, Where: V = the volume of 0.1N NaOH in 10 g of honey (Bogdanov, 2009).

Determination of total ash content

Honey samples was incinerated at 600°C in a muffle furnace (BioBase JKKZ.5.12GJ, Shandong.ltd, China) to constant mass (Bogdanov, 2009). First, the ash dish was heated in an electrical muffle furnace

at ashing temperature and subsequently cooled in a desiccators to room temperature and weighted to 0.001 g (M2). Then 5 g (M0) of each honey sample was weighed to the nearest 0.001 g and taken into a platinum dish and two drops of olive oil were added to prevent foaming. Water was removed and started ashing without loss at a low heat rising to 350 – 400°C using electrical devices. After the preliminary ashing, the dish was placed in the preheated furnace and heated for at least 1 h. The ash dish was cooled in desiccators and weighed. The ashing procedure was continued until constant weight was reached (M1). Lastly, % of weight of ash in g/100 g honey was calculated using the following formula: -

$$WA = \frac{M1 - M2}{M0}$$

Where, M0= Weight of honey taken, M1= Weight of ash + dish and M2= Weight of dish.

Determination of sugars

High performance liquid chromatography (HPLC- 1260 Infinity Series Agilent Technologies, Germany) was used. Five grams of honey was dissolved in 40 ml water. A 25 ml of acetonitrile was pipetted into a 100 ml volumetric flask and the honey solution was transferred to a flask and filled to the mark with distilled and the solution of each honey sample was filtered using syringe filter (0.45 µm) before chromatographic analysis. The HPLC separation system was composed of analytical stainless steel column, 4.6 mm in diameter, 250 mm length, containing amine modified silica gel with 5-7 µm particle size. Flow rate 1.3 ml/min, mobile phase Acetonitrile: water (80:20, v/v) and sample volume 10 µl. The sugars was detected by a Refractive Index Detector thermo stated at 30°C temperature regulated column oven at 300°C. The identification of honey sugars was obtained by comparison of their retention times with those of the standard sugars (Bogdanov, 2009).

Determination of hydroxyl methyl furfural (HMF)

UV–Vis spectrophotometer (JENWAY, United Kingdom) was used (Bogdanov, 2009). A 5 grams honey sample was mixed in 25 ml distilled water and transferred into 50 mL volumetric flask. A 0.5 mL carrezz solution I (15 g K₄Fe (CN) 6. 3H₂O /100 mL distilled water) was added and mixed into 0.5 mL carrezz solution II (30 g Zn acitate /100 mL distilled water). The solution was mixed into the honey solution. A droplet of alcohol was added into the solution. The solution was filtered through a filter paper and the filtrate (10 mL) was discarded. A 5 mL filtrate was added into each of two test tubes and 5 mL distilled water was added into the first test tube (sample solution), while 5 mL sodium bisulfite solution (0.20% of 0.20 g NaHSO₃/100 mL distilled water) was added into the other test tube (reference). The contents of both test tubes were well mixed by vortex mixer and their absorbance was recorded spectrophotometrically by subtracting the absorbance measured at 284 nm for HMF in the honey sample solution against the absorbance of reference (the same honey solution treated with sodium bisulfite, 0.2%) at 336 nm and the result was calculated and expressed according to International honey commission (Bogdanov, 2009), which is:

$$\text{hydroxylmethyl furfural (HMF)/100 g honey} = (A_{284} - A_{336}) \times 14.97 \times 5/\text{g sample},$$

Where A₂₈₄= absorbance at 284, A₃₃₆ = absorbance at 336, 14.97= constant, 5= theoretical nominal sample weight and g = mass of honey sample.

Honey color analysis

Pfund classifier was used to measure the color of honey samples. Briefly, homogeneous honey samples free of air bubbles were transferred into a 10-mm light path cuvette until the cuvette was approximately half full. Then the cuvette was inserted into a color photometer Pfund honey color grader (No. 0061, made of USA) and the color grades were expressed in millimeter (mm) Pfund grades compared to an analytical grade glycerol standard following the procedure of Codex Alimentarius Commission Standards, 2001. Measurements were performed for each sample using approved color standards of the United States Department of Agriculture, 1985.

Data analysis

Data means \pm SD were calculated using SAS Software (SAS Institute, 2003; 14). For botanical origin analysis, pollen grain morphology between the honey types were counted from the slide microscopically and their percentage were calculated by dividing the single plant species pollen grain morphology over the total different plant pollen grain morphology, and then multiplying by 100.

Result and discussion

Botanical origin of honey types

The honey plant species with their pollen frequency classes of FSHS and SSHS are listed in Table 1 below. It shows that seven herbaceous species and one tree species, a total of eight plant species from five families were identified from FSHS. On the other hand SSHS was synthesized from ten plant species under six families including herbs, shrubs and tree life form. All of the identified plant species (eighteen in number of plant species) provides both pollen and nectar sources for the bees (Tura Bareke and Admassu Addi, 2019), even though their pollen frequency class varies.

The predominant pollen source for FSHS and SSHS were *Guizotia* species and *C.arabica* respectively. *Sesamum indicum* and *Trifolium* spp were secondary pollen source for FSHS while *Vernonia amygdalina* and *Vernonia auriculifera* were secondary pollen source for SSHS. Pollen of a particular plant species is said to be predominant (monofloral honey type) if its occurrence in the honey sample is more than 45% of total pollen count, secondary pollen (16-45%), important minor pollen (3-16%), and minor pollen (<3%) (Louvaeux 1978; Agashe and Caulton, 2009). Therefore, FSHS of this study could be called a *Guizotia* (monofloral) honey (figure 1). Similarly, *C.arabica* species was a dominant plant and its pollen grain counted 46% from a total pollen count from SSHS and could be said *C.arabica* (monofloral) honey (figure 2).

Fechner *et al.* (2016) investigated that the collection of pollen and nectar by the bee foragers depends on the availability of botanical resources within their foraging ranges which are affected by the environmental and seasonal factors. Basically *Guizotia* species flowered from mid October to November at the study area (Field observation and personnel communication). During this period it grows in abundance in a very wide range habitats and available everywhere like in cultivated fields, forest margin and open grasslands which might be a reason for its pollen grain dominance from FSHS.

The flowering period of *C.arabica* at the study area depends on the raining condition. Mostly it flowers either in January or February following the rain soon. This plant is widely cultivated for its fruits and is a much known cash crop in the study area. When flowered it is abundantly available for the forager bees and releases plenty of nectar and pollen.

Table 2: The characteristics of identified honey plants with their pollen frequency class from honey

Honey type	Scientific name	Family name	Vernacular name	Life form	Resources for bees	Frequency class	Honey type
FSHS	<i>Guizotia</i> species	Asteraceae	Tufu/hada/nuugi i	Herb	Pollen & nectar	PP	Monofloral honey- <i>Guizotia</i> honey
	<i>Sesamum indicum</i>	Pedaliaceae	Saalixa	Herb	Pollen & nectar	SP	
	<i>Trifolium</i> species	Fabaceae	Siddisa	Herb	Pollen & nectar	SP	
	Grass species	Poaceae	Gosamargaa	Herb	Pollen & nectar	MP	
	<i>Parkinsonia aculeata</i>	Fabaceae		Tree	Pollen & nectar	MP	
	<i>Vicia faba</i>	Fabaceae	Baaqelaa	Herb	Pollen & nectar	MP	
	<i>Plectranthus assurgens</i>	Lamiaceae	Ajooftuu	Herb	Pollen & nectar	MP	
	<i>Andropogon</i> species	Poaceae	Marga	Herb	Pollen & nectar	MP	
SSHS	<i>Coffea arabica</i>	Rubiaceae	Buna	Shrub	Pollen & nectar	PP	Monofloral honey- <i>C. arabica</i>
	<i>Vernonia amygdalina</i>	Asteraceae	Eebicha	Shrub	Pollen & nectar	SP	
	<i>Vernonia auriculifera</i>	Asteraceae	Reejjii	Shrub	Pollen & nectar	SP	
	<i>Syzygium guineense</i>	Myrtaceae	Baddeessaa	Tree	Pollen & nectar	IMP	
	<i>Terminalia</i> species	Combretaceae	Dabaqqaa	Tree	Pollen & nectar	IMP	
	<i>Cirsium</i> species	Asteraceae	Qoraattiiharree	Herb	Pollen & nectar	MP	
	<i>Vernonia leopoldii</i>	Asteraceae	Sooyyama	Shrub	Pollen & nectar	MP	
	<i>Pterolobium stelatum</i>	Fabaceae	Harangamaa	Shrub	Pollen & nectar	MP	
	<i>Grevillea robusta</i>	Proteaceae	Botoroo	Tree	Pollen & nectar	MP	
	<i>Guizotia</i> species	Asteraceae	Tufo/hada/nuugi i	herb	Pollen & nectar	MP	

Where PP; predominant pollen (> 45% of the pollen grains counted), SP; secondary pollen (16-45%), IM; important minor pollen(3-15%), MP; .minor pollen (less than 3%).

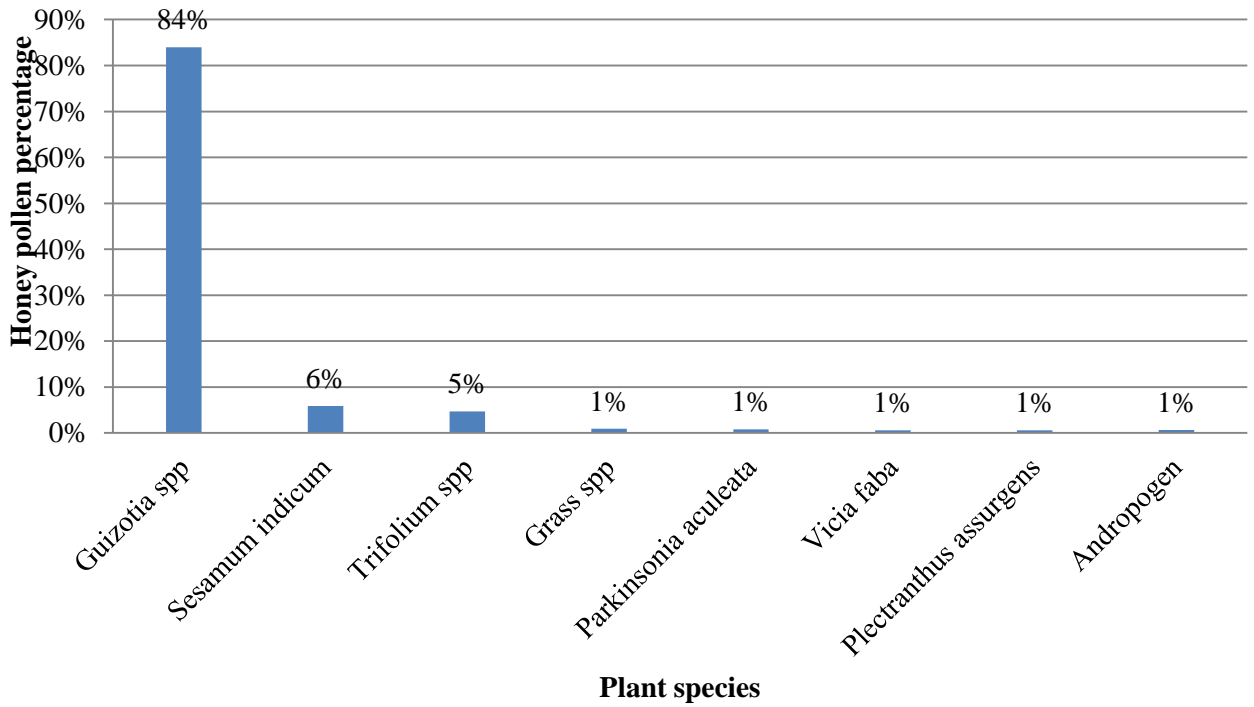


Figure 5: Relative frequency of nectariferous plant species from FSHS (% distribution)

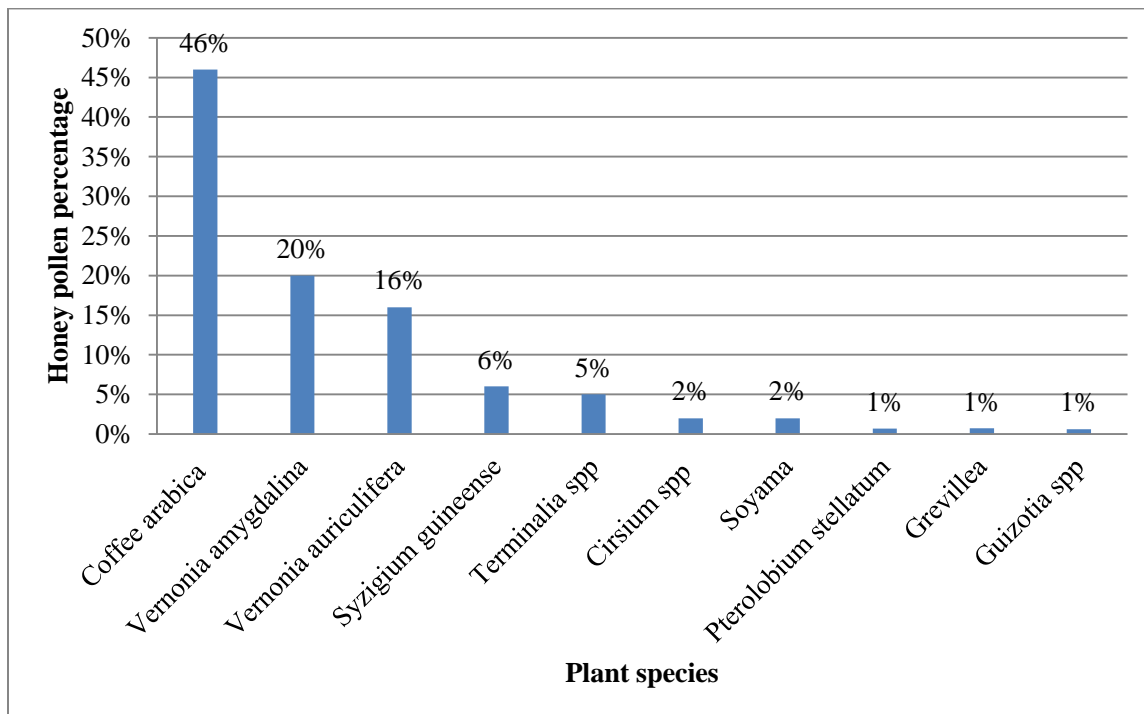


Figure 6: The percentage relative frequency of nectariferous plant species from SSHS

Physiochemical composition between honey types

Sugar profile

The sugar composition results (%) of the analyzed honey types are indicated in table 2 below. A significance difference ($p < 0.05$) was observed on fructose content with a mean of 42.20 ± 0.74^a by *C. arabica* honey and 37.71 ± 3.65^b by *Guizotia* honey. However, the mean of glucose and maltose content between honey samples didn't show significant variation ($p > 0.05$). *C. arabica* and *Guizotia* had recorded glucose content of $36.10 \pm 5.60a$ and $33.95 \pm 4.33a$ respectively while maltose content from *C. arabica* and *Guizotia* was $1.72 \pm 0.90a$ and $1.70 \pm 1.20a$ respectively. Our study showed that fructose is the predominant monosaccharide sugar which followed by glucose and maltose, and are more recorded from *C. arabica* than *Guizotia*. The disparity between the honey types of this study might be due to the possible effects of plant species.

The predominance of fructose over glucose and glucose over maltose of our result was in line with the finding by Makarewicz *et al.*, (2017) from Poland country. They encountered a mean fructose content of 41.36 ± 1.42 (*Eucalyptus* honey type) to 47.18 ± 0.78 (*Lime* honey type); a mean glucose content of 26.34 ± 1.84 (*Thyme* honey type) to 37.93 ± 1.42 (*Eucalyptus* honey type), and a mean maltose content of $1.88b \pm 0.11$ (*Eucalyptus* honey) to $6.64a \pm 0.15$ (*Coriander* honey). Moreover, Abara Belay *et al.*, 2017 analyzed the sugar profile between Ethiopian monofloral honeys and they obtained a mean fructose content from $35.30 \pm 3.53d$ (*Becium grandiflorum*) - $43.07 \pm 0.37a$ (*Acacia*); a mean glucose content of $29.34 \pm 2.75e$ (*B. grandiflorum*) to $37.20 \pm 0.35a$ (*Leucas abyssinica*), and a mean maltose content of $0.55 \pm 0.34f$ (*Schefflera abyssinica*) to $2.04 \pm 0.45a$ (*Eucalyptus globules*). Although the concentration of both sugars varies depending on the botanical, entomological origin of the honey, it is generally expected that fructose will be found in a higher proportion than glucose and maltose (Yücel *et al.*, 2013).

Statistically significant difference ($p < 0.05$) was seen on the mean of reducing sugar (a cumulative mean of fructose, glucose and maltose) and $80.00 \pm 5.71a$ and $73.35 \pm 5.30b$ was recorded from *C. arabica* and *Guizotia* respectively (table 2). These was within the acceptable limit of international standard as determined by Codex, 2001 and EU, 2002 which should be $\geq 60\%$, and National standard which should be $\geq 65\%$ (ESA, 2013). The mean reducing sugar of ours was exceedingly higher than what observed for *Pineapple* type ($61.17 \pm 0.17b$) to $63.89 \pm 0.25a$ (*Acacia* type) from Malaysian honey (*A. mellifera*) sample (Moniruzzaman *et al.*, 2013) and Bangladesh *A. mellifera* honey ($63.3 \pm 1.5\%$) (Isalm *et al.*, 2017). Furthermore, Gebreegziabher Gebremedhin *et al.*, (2013) from Amhara, Ethiopia reported a mean reducing sugar of $71.2 \pm 2.5\%$; while from Tigray, Ethiopia Tewodros *et al.*, (2013) informed a mean of $67.3 \pm 2.42\%$, which all are lower than ours. From our study, *C. arabica* produced more sugar than *Guizotia* honey which might be due to the more availability of enzyme and sugar in *C. arabica* nectar. This was in line with the observations of Cavian (2002) the presence of enzyme in bees and nectar as well as the presence of sugar in nectar of plant is the main factor for sugar production of any honey. The difference of reducing sugar between honey samples might be due to the conversion of sugars into organic acids (Cavia *et al.*, 2007)

Both of the analyzed honey types yielded the sucrose content within the accepted international standard; Codex, 2001 and EU, 2002, which should be ≤ 5 and national standard (ESA, 2013) (≤ 10). This study yielded sucrose content of $2.17 \pm 1.75a$ from *Guizotia* and $2.15 \pm 0.61a$ from *C. arabica* and no significant variation ($p > 0.05$) was observed between them (table 2). A comparable observation were made from

Nigerian *A. mellifera* honey with a mean of 2.32 ± 0.01 - 2.42 ± 0.02 (Nwezze *et al.*, 2017). The amount of sucrose is determined by the degree of maturity and origin of the nectar compound of the honey and is used to detect adulteration of honey by addition of cane or other sugars. A related result between ours honey types might be the effect of the harvested material. Even though the season between honey samples were different with plant species, both honey types were harvested by the bee technician and only sealed honey comb was selected. The sucrose content in matured honeys could be low, due to the invertase enzyme, which degrades the disaccharide (sucrose) into two simple sugars (glucose and fructose) (Fonte *et al.*, 2013). According to Scripca *et al.*, (2019), the sucrose content is an important parameter of the authentication of honey and the presence of a high level of sucrose in honey indicates adulteration with different syrups and also harvesting the product before maturation. Our study distinguished the maturity (ripeness), natural and free from foreign material (adulteration) of the tested honey samples.

Table 3: The percentage sugar composition between monofloral honey types

Parameters	Honey type (Mean \pm SD)		X	LSD	P-value	International Standard		National standard
	<i>Guizotia</i>	<i>C. arabica</i>				Codex, 2001*	EU, 2002**	
Fructose (%)	37.71 \pm 3.65b	42.20 \pm 0.74a	39.6	3.33	0.01	-	-	-
Glucose (%)	33.95 \pm 4.33a	36.10 \pm 5.60a	34.9	5.76	0.43	-	-	-
Maltose (%)	1.70 \pm 1.20a	1.72 \pm 0.90a	2.16	1.28	0.97	-	-	-
RS (Fru + Glu+Malt) (%)	73.35 \pm 5.30b	80.00 \pm 5.71a	76.2	6.44	0.04	\geq 60	\geq 60	\geq 65
Sucrose (%)	2.17 \pm 1.75a	2.15 \pm 0.61a	1.7	1.64	0.94	\leq 5	\leq 5	\leq 10

Means with different superscript (a, b, c) within the rows are statistically different at $p \leq 0.05$.

Note : SD = Standard deviation, X = overall mean, LSD = Least Significant Difference at $\alpha = 0.05$, RS (%) = percent of reducing sugar in honey , * = Codex Alimentarius Commission (2001), ** = The Council of European Union (2001) and *** = Ethiopian Standards Agency (2013)

MC (moisture content), FA (Free acidity), and pH

The mean results of MC (moisture content), FA (Free acidity), and pH of the analyzed honey types were indicated in table 3 below. Statistically no variation ($p > 0.05$) was observed between honey types with $19.70 \pm 1.43a$ and $18.80 \pm 0.54a$ for *Guizotia* and *C. arabica* respectively. The recorded MC results are within the limits (should be not more than 20%) set by international standard (Codex, 2001 and EU, 2002) while it should be not more than 21% set by national standard (ESA, 2013). Both of our honey types were fell within the range of the finding by Abara Belay *et al.*, (2017) who investigated the highest MC by *S. abyssinica* (20.54 ± 1.28) whilst the lowest MC by *E. globules* (14.14 ± 0.19) from Ethiopian *A. mellifera* honey sample. However, below ours result were observed from Romania country with the

lowest water content in the polyfloral (16.78 ± 0.65) sample and the highest content in acacia (17.13 ± 0.48) honey sample (Scripta *et al.*, 2019).

Moisture is the second most important constituent of honey following the sugar from our study in particular and as a world in general. The comparability between our honeys types might be the harvesting procedures. Our honey samples were collected by the bee technician following the standard procedures and waiting for the proper level of maturity within the modern hive. Generally, uncapped honey that contains more water is not recommended for harvesting. Evidently, Scripca *et al.*, (2019) has been claimed high water content indicates extraction of a product in high humidity conditions or premature extraction. Moreover, Acquarone *et al.*, (2007) have observed that the variation of MC in honey samples is fundamentally influenced by (a) geographical position from where the nectar and pollen producing plant and the bee colony was found, (b) the level of honey maturity in the hive, (c) botanical origin of honey, and (d) harvesting and post harvesting manipulation. When the MC is high in honey it increases the honeys water activity and influences the shelf-life of honey. According to Codex Alimentarius (2001) standard, the water content should not exceed 20% in honey to ensure safety against fermentation caused by the action of osmotolerant yeasts during storage.

The free acidity result is presented in Table 3 below. Statistically no variation ($p > 0.05$) on free acidity between honey types and $31.43 \pm 1.52a$ by *C. arabica* while $27.18 \pm 3.70a$ by *Guizotia* was observed. The mean free acidity of this study fits the national ($< 40 \text{ meqkg}^{-1}$) (ESA, 2013) and international (Codex, 2001 and EU, 2002) quality standards which should be a maximum of 50 meqkg^{-1} from *A. mellifera* honey. Comparably, this mean value was close to the result reported from Tigray ($29.89 \pm 5 \text{ meqkg}^{-1}$; Gebreegziabher Gebremedhin *et al.*, 2013) and Amhara ($27.34 \pm 5.06 \text{ meqkg}^{-1}$; Tewodros *et al.*, 2013). However, the mean free acidity of ours was by far higher than the honey (*A. mellifera*) obtained from Nigeria ($18.67 \pm 0.64 \text{ meqkg}^{-1}$; Nweze *et al.*, 2017) and Polish market ($14.40 \pm 0.58 \text{ meqkg}^{-1}$; Makarewicz *et al.*, 2017). Free acidity indicates one of the quality parameters of honey samples and it reveals whether the honey is fermented or not (Silvano *et al.*, 2014) and corresponds to the presence or absence of organic acids in the product.

Statistically significant variation ($p < 0.05$) between our honey types with a pH of $3.59 \pm 0.05a$ by *C. arabica* and $3.34 \pm 0.12b$ by *Guizotia* was recorded (Table 3). The mean pH result of honey (*A. mellifera*) observed from the present investigation was within the pH range of international standard (3.2-4.5). The mean pH value of ours fell within the range of the finding by Abara Belay *et al.*, (2017) who investigated the highest pH value ranged from 4.6 ± 0.1 by *E. globulus* to 3.4 ± 0.1 by *Hypoestes* and a significance difference was observed. The pH of honey affects its texture, stability and shelf life and a pH unit between 3.4 and 6.1 indicates the freshness of honey samples (Khalil *et al.*, 2012). The variations in pH values of honey from different locations are due to the different geographic origins as the nectar's pH and soil conditions may influence honey physicochemical properties (Khiati, 2014).

HMF (Hydroxymethyl furfural), ash and color

The mean results of HMF (Hydroxymethyl furfural), ash and color of the analyzed honey types were indicated in table 3 below. The enzymatic activity and concentration of hydroxymethylfurfural (HMF) in honey sample are one of the important indicators of honey's quality (freshness) indicating whether the honey is aged or over-heated (Mairaj *et al.*, 2008). Statistically similar ($p > 0.05$) amount of HMF was observed with a mean of $14.85 \pm 6.72a$ by *Guizotia* and *C. arabica* by $12.50 \pm 4.91a$. None of the

investigated samples exceeded the allowed limit of National (ESA, 2013) and International (Codex, 2001; EU, 2002) quality standards which should be not more than 40 (mg/kg). The HMF concentration which could be comparable with ours was reported from Nigerian *A. mellifera* honey and ranged from 11.97 ± 0.05 mg/kg to 16.12 ± 0.12 mg/kg (Nwezze *et al.*, 2017). The low HMF concentration of our honey types indicates that they are fresh honey and not taken into a high temperature which made them to have a good quality.

The mean ash content of *Guizotia* and *C. arabica* was 0.11 ± 0.04 (g/100g) and 0.27 ± 0.06 (g/100g) respectively. Our result founds within the acceptable limit of international standard (Codex, 2001; EU, 2002) which should be a maximum of 0.6 (g/100g). From Ethiopian monofloral honey samples Abara Belay *et al.*, (2017) have observed a range from 0.19 ± 0.07 by Hypoestes honey to 0.39 ± 0.04 g/100g from *C. macrostachyus* honey. Ash content expresses the richness of honey in mineral content. The minerals calcium (Ca), magnesium (Mg), iron (Fe), copper (Cu), cadmium (Cd) and zinc (Zn) in the form of sulfate (SO_4^{2-}) and chloride (Cl^-) are found in small amounts. The ash content result depends on the floral origin and soil features, and honeys with ash content of $\leq 0.6\%$ have the nectar source (Andrade *et al.*, 1999). The blossom honeys (nectar of plants) have lower ash content than the honeydew (secretions of living parts of plants or excretions of plant-sucking insects on plants) (Bogdanov, 2009)

The color of this study honey types were indicated in table 3 below. *Guizotia* and *C. arabica* exhibited a mean of 91.38 ± 27.17 and 114.67 ± 5.27 Pfund value respectively. According to the USDA-approved color standards (USDA 1985), the mean of *Guizotia* and *C. arabica* could be classified under Amber and Dark Amber color respectively. Similar observation with our *Guizotia* color was observed from Dellomenna district (91.0 ± 9.24 b-Amber; mm pfund) and *C. arabica* color with Dinsho district (118.0 ± 6.91 a- Dark Amber; mm pfund) of Bale natural forests (Bekele Tesfaye *et al.*, 2016). Color of untreated honey can be used for its floral origin and is the single most important factor determining import and whole sale prices (Bertoncelj *et al.*, 2007). Furthermore, the use of old comb, contamination with metals, exposure of honey to high temperature and long storage might changes the color between honey samples. Variations in the color of honey are related to its floral origin, mineral content, storage and product processing, climatic factors during nectar flow and the temperature at which the honey matures in the hive, as well as factors such as the proportion of fructose and glucose present, nitrogen content and the instability of fructose in an acid solution. The dark color is related to the content of minerals, pollen and phenolic, and is characteristic of floral origin. Darkening of honey during storage may occur because of Maillard reactions (chemical reaction between amino acid & reducing sugar), fructose caramelization and reactions of polyphenols.

Table 4: The physicochemical composition between monofloral honey types

Parameters	Honey type (Mean ± SD)		X	LSD	P-value	International standard		National standard
	<i>Guizotia</i>	<i>C. arabica</i>				Codex, 2001*	EU, 2002**	
Moisture content (%)	19.70±1.43a	18.80±0.54a	19.3	1.35	0.16	≤20	≤20	≤21
FA (meqkg ⁻¹)	27.18±3.70a	31.43±1.52a	29.3	6.52	0.88	≤50	≤50	≤40
pH(pH units)	3.34±0.12b	3.59±0.05a	3.4	0.11	0.00	3.2-4.6		-
HMF (mg/kg)	14.85±6.72a	12.50±4.91a	13.9	7.10	0.49	≤40	≤40	≤40
ASH(g/100g)	0.11±0.04 ^a	0.27±0.06a	0.2	0.15	0.05	≤0.6	≤0.6	-
Color (mm pfund)	91.38±27.17a	114.67±5.27a	101.3	24.74	0.06			

Means with different superscript (a, b, c) within the rows are statistically different at $p \leq 0.05$. Note: SD = Standard deviation, X = overall mean, LSD = Least Significant Difference at $\alpha = 0.05$, FA (meqkg⁻¹) = Free acidity, HMF (mg/kg) = Hydroxymethylfurfural, * = Codex Alimentarius Commission (2001), ** = The Council of European Union (2001) and *** = Ethiopian Standards Agency (2013)

Conclusion

Based on the laboratory analysis obtained, honey harvested during the months of mid November through December (FSHF) was predominantly produced from *Guizotia* spp, and while honey cropped during February dominantly produced from *C.arabica*. Thus the study confirmed that at least two different monofloral honeys can be obtained from the study areas. *Guizotia* honey had more acidic whilst less sugar production compared to *C.arabica* honey. The quality of honey produced from the study areas meet the international and national quality standards.

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The Role of Honeybee (*Apis mellifera*) Pollination in Enhancing Seed Yield and Yield related parameters of *Coriandrum sativum* L

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Abstract

Coriander (Coriandrum sativum L.) is one of the most important annual spices and medicinal herb plants. It is an open pollinated crop and honeybees are effective pollinators for open pollinated crops because of ample of nectar and pollens are available on the flowers of Coriander. An experiment was conducted to evaluate the effect of honeybee's pollination on Coriandrum sativum seed yield and yield related parameters at Sinana Agricultural Research center at on-station. The study was designed into three treatments. These include plots caged with honeybees (T1), plots caged without honeybees (T2) and open pollinated plots (T3). All collected data were analyzed using One-way-Analysis of Variance (ANOVA). There were no significant different ($P>0.05$) on date of flowering, primary and secondary branches among the three treatments. Whereas, there were a significant difference ($P<0.05$) on flowering period, shading time, Number of capsule per plant, Thousand kernel weight and total seed yield per hecter. The result also revealed that about 29.70% of seed yield advantage of Coriandrum sativum pollinated by honeybees over control. From this result it was concluded that visits of honeybees at flowering time of Coriandrum sativum is veryhelpful in increasing seed yield and yield related components of this crop.

Keywords: Honeybees, Pollination, Seed yield, Seed weight

Introduction

Pollination is one of the most important ecological interactions and the first step for the sexual reproduction of the most plant species (Murcia 1996). Pollination carried out by animals and other abiotic agents it is a considered an important ecosystem service with 35% of the plants cultivated in the world benefitting from this interaction (Klein *et al.*, 2007). Among the various pollinating agents, insects played a major role. The global annual economic value of insect pollination is estimated to be € 153 billion (Gallai *et al.*, 2009). Of the total pollination activities, over 80 per cent is performed by insects and honeybees contribute nearly 80 per cent of the total insect pollination, and they are considered the best pollinators (Robinson and Morse, 1989) due to their suitable body size, hairiness, thoroughness, steadfastness, floral constancy, and manageable populations. It is also known that honeybees (*Apis mellifera* L.) play an important role in pollination of some cultivated plants, notably allogamous species for which cross pollination is essential. Cross pollination of entomophilous crops by honeybees is considered as one of the effective and cheapest methods for triggering the crop yield both qualitatively and quantitatively. Being the pollination service provider bees contribute handsomely in enhancing the

productivity and production of cross pollinated crops through efficient pollination in an inconspicuous and silent manner (Singh *et al.*, 2005 and Mohapatra *et al.*, 2010).

Coriander (*Coriandrum sativum L.*), which belongs to the family of Apiaceae is one of the most important annual spices and medicinal herbs which is cross pollinated plants. It is grown in Ethiopia and throughout the world for its seeds as well as leaves and it has immense uses such as spice and medicinal values (Parthasarathy *et al.*, 2008; Nowak and Szemplinski, 2014). Coriander originated from the Mediterranean and Western Asian regions (Burdock and Carabin, 2009). There is a long standing tradition of cultivation of coriander in Ethiopia (Geremew *et al.*, 2014) for spice and medicinal uses. The flowers are hermaphrodite and protandrous where pollen release precedes stigma receptivity, so pollen from a different flower is required for seed set (Wierdak, 2013; Mandal and Mandal, 2015). According to the observations by several authors (Sinacori *et al.*, 2009; Bendifallah *et al.*, 2013), the flowers of coriander attract many groups of insects, in particular the Diptera, Coleoptera and Hymenoptera. However, honey bees are the potential pollinators of this crop.

Although, honeybees have a great contribution in giving pollination service for ecosystem many people do not understand it. Most of the time people only know by their honey and bees wax production, but the values of honeybees for pollination services had 10 folds' times the values that they have for honey and beeswax. Moreover, the role of honeybees for pollination of local farming system is still poorly understood and till not sufficiently appreciated (Jacobs *et al.*, 2006). So far, there is no detailed information regarding the pollinators or foragers requirements of coriander growing in Ethiopia. Therefore, the aim of this study was to assess the role of honeybees in enhancing the yield and yield related components of *Coriandrum sativum* in the highland of Bale Zone of Oromia Regional State in Ethiopia.

Materials and Methods

The study was conducted at the high land of Bale in Sinana Agricultural Research Centre (SARC) at station during the 2017-2019 main cropping seasons. It is found at a distance of about 463 km from Addis Ababa in the south-easterly direction, and 33 km from the nearby town, Robe. The geographic location is 07^o 07' N latitude and 40^o 10' E longitudes. The elevation is 2400 meters above sea level. The area is characterized by bimodal rainfall pattern. The annual average rainfall is 548 mm and mean maximum and minimum temperatures are 26.5°C and 11.8°C, respectively (SARC, 2006).

Experimental set up

The experiments were arranged in a randomized complete block design (RCBD) with five replications. For the experiment Waltahi variety of Coriander (*Coriandrum sativum*) was used and all recommended agronomic practices were also followed. The plots were kept from any damaging condition throughout the cropping season. The treatments were: plots caged with honeybees (T1) the plots were covered with an insect proof mesh cage and a honeybee colony were placed inside the cage during the flowering peak (50% florets open). Caged without honeybees or pollinator exclusion (T2) the plots were covered with an insect proof mesh cage before the ray florets started opening, plots kept open to all pollinators (T3)-plots accessible to all flower visitors or left open for natural pollination as control. Insect proof mesh cages (4m x 3m and 2.5m high) were made of wood covered with 20% shade cloth. All insects were removed from

all the cages before blooming, to exclude unwanted pollinators. Honeybee colonies used in this experiment received supplementary feeding (dissolved sugar) and water before and after they were placed in the cages. At the time of maturity 10 mature pods were selected randomly from each replication and the number of seeds produced was counted manually. Harvesting was done from each plot after seeds are matured. The seeds were separated manually from the pods and yield had calculated per plot for all the treatments.



Fig. 1. Coriander pollination experimental set up (cage with bees, caged without bees and open plots)

Flower visitor identification

During the whole flowering period, flower visitor identifications were done in each of the plots open plots which is accessible to all flower visitors, to assess types and frequency of insect species were visiting the *Coriandrum sativum* crop. According the number of bees and other pollinators in the open treatment was observed in one m² area for five minutes seven days a week during the whole flowering period and the data was recorded at every two hours interval (9.30am, 11.30am, 1.30 pm and 3.30pm) hours a day. Visiting insects were collected and identified by the entomologist at Sinana Agricultural Research Center. An increase in seed yield and quality of *Coriandrum sativum* seeds due to managed honeybee pollination was calculated using the formula as follows developed by Beyissa, (2006).

$$\text{Yield increment(\%)} = \frac{(\text{Yield from honey bees pollinated} - \text{Yield from insect excluded})}{\text{Yield from from open pollinated}} \times 100$$

Data collection and measurement

Flowering Period: The flowering period was determined by recording the flower starting and ending date of the plants. For this purpose ten plants were purposely selected to investigate the effects of honeybee of pollination on flowering period of the plant. Number of primary and secondary branches per stem was randomly counted from selected ten middle row plant final harvests. Number of Capsule: On individual plant basis, number of capsule in the tagged plants counted manually. The mean capsule per plant was taken for each treatment. Shading Time was determined when plants reached about the 50% physiological maturity and its flowers were totally shades.

Thousand kernel weight (TKW) (g): was determined based on the weight of 1000 seeds sampled from the grain yields of each plot by counting using an electric seed counter and weighed with an electronic balance. Grain yield was determined by harvesting plants from the net middle plot area to avoid border effects. Seeds, which were obtained from the corresponding net plot were cleaned manually and weighed using sensitive balance and recorded as mean values of seed yield per hectare in kilograms.

Data analysis:

All collected data were subjected to analysis of variance using statistical software package (SAS 9.1.3). The data were statistically analyzed using one-way-analysis of variance (ANOVA) and the differences among treatment means were compared using Least Significance Difference (LSD) test at 5% level of significance.

Results and Discussions

Coriandrum sativum flowers were visited by 7 species of insects belonging to 4 orders (Table 1) such as *Apis mellifera*, *Xylocopa*, orthoptera, *Colletes succinctus L*, *Musca* sp, *Danaux plexippusL* etc. *Apis mellifera* was found the most frequent visitors (64.62%) at maximum activity at 11:30 am and minimum activity at 3: 30 pm. This is probably due to the bee's activity being limited by environmental factors like daily temperatures and relative humidity. Counts were made on one meter (1 m²) for 5 minutes, when the flowers were open. *Apis mellifera* as the most dominant floral visitor of coriander has also been reported earlier by (Ricciardelli and Ambrosio 1979; Chaudhary O P and Jage Singh, 2006) respectively. It is generally thought that the more visits made, the more efficient is the pollinator, though this also depends on the per visit pollen contribution to the pistillate flower part (Herrera, 1989).

Table 1. Coriander (*Coriandrum sativum L*)visitor's frequency and percentages

No	Insect order	Common Name	Scientific Name	frequency	Percentage
1	Hymenoptera	Honeybees	<i>Apis mellifera</i>	42	64.62
		Carpenter bee	<i>Xylocopa</i>	2	3.08
2	Orthoptera	Grass hopper	<i>Orthoptera</i>	3	4.62
		Wasp	<i>Colletes succinctus L</i>	6	9.23
3	Diptera	Housefly	<i>Musca</i> sp.	5	7.69
4	Lepidoptera	Butterfly	<i>Danaux plexippus L</i>	3	4.62
		Spider	<i>Achaearana Trpidariorum</i>	4	6.15
Total pollinator count				65	100

The mean date of flowering (Table 2) showed no significant differences among the treatments. This might due to date of flowering is not affected by mode of pollinations; however it depends on environmental factors like daily temperate, relative humidity and also on soil type. In the current study shading time of *Coriandrum sativum* was significantly affected by pollinating agents. The early shading time were observed in treatment caged with honeybees (107.2days) and followed by open pollination (111 days). This showed that shading time highly depends on mode of pollinations.

The flowering period of *Coriandrum sativum* were significantly affected by mode of pollination. Plots caged without honeybees had the longest flowering period (41.4days), followed by open pollination, and while caged with honeybees had the smallest flowering period (32.47days). This is in agreement with Oz *et al.* (2009) reported flowering period was affect by mode of pollination and the longest flowering period was observed in Canola crops caged without bees followed by open pollinated crops. This indicated that mode of pollination has a great contribution for early maturation of *Coriandrum sativum*.

Table 2. Mean comparison of three years' data collected on Date of flowering date of blooming, Shading time and Flowering period

Treatments	DF	ST	FP
Caged with honeybees (T1)	74.73±2.06	107.2±2.297 ^b	32.47±0.703 ^c
Caged without honeybees (T2)	74.8±2.031	116.2±2.46 ^a	41.4±0.515 ^a
Open Pollination (T3)	74.8±2.041	111±2.332 ^{ab}	36.2±0.518 ^b
Over all mean	74.78±1.153	111.5±1.445	36.69±0.643
LSD	NS	6.75	1.67
CV (%)	10.59	8.21	6.18

a,b,c= means with different superscripts within a column are significantly different (P<0.05), NS= none Significant, DF = Date of Flowering, ST= Shading Time, FP= Flowering period.

The number of primary and secondary branches were not significantly different (p>0.05) among treatments of *Coriandrum sativum*. This may probably because of the primary and secondary branches were not affected by mode of pollination, but it is affected by environmental factors and soil type. Significant difference (P<0.05) was observed in capsule setting among treatments (Table 3). Plots caged with honeybees had the highest number of capsule setting per plant (184.93), while plots caged without honeybees had the lowest number of capsule setting per plant (108.67). Similarly, in Sunflower crops caged with honeybees increased significantly the percentage of seed setting, number of filling seed per head compared with crops caged without honeybees (Oz *et al.*, 2009).

Table 3. Mean comparison of three years data collected on Primary branch, secondary branch and Number of Capsule

Treatments	PB	SB	NCP
Caged with honeybees (T1)	23.87±5.09	59.73±8.87	184.93±20.02 ^a
Caged without honeybees (T2)	24.33±4.87	55.8±7.76	108.67±12.04 ^b
Open Pollination (T3)	26.33±6.02	63.33±9.88	149.8±17.16 ^{ab}
Over all mean	24.84±3.02	59.62±5.03	147.8±10.54
LSD	NS	NS	3.4434
CV(%)	43.85	83.40	57.66

^{a,b,c}= means with different superscripts within a column are significantly different (P<0.05), NS= none Significant, PB= Primary branches, SB= secondary branches and NCP= Number of capsule

The present result revealed that there was a significant different (P<0.05) among treatments regarding thousand seed weight (TKW). Plots caged with honeybees the highest TKW (3.05 g), whereas plots caged without honeybees had the lowest TKW (2.16 g). Munawar *et al.*, 2009 found a similar result from Pakistan on the Coriander crop caged with honeybees.

Mode of pollination had a significant effect on the total yields per hector. From the current study, the total yield of plots under different treatments were compared and significant (P<0.05) differences were found. The yield from all treatments were different and the highest yield per hectare was obtained in treatments plots caged with honeybees (19.38Qt/ha) and followed by open pollinated crop(16.80Qt/ha). The lowest yield per hectare was gained from caged without honeybees (14.39Qt/ha) (Table4). The higher yield of crops caged with honeybees might be due to the higher pollination efficiency of honeybees inside the cage. These results are in agreement with the already recorded observations by Dhore (2009) and Kumar and Jaiswal (2012) on the effect of pollinators in increasing seed yield of *C. sativum*. The results are also in agreement with previous result of Gebremedhin and Tadesse, (2014) on *Guizotia abyssinica* at Tigray Region of Ethiopia.

Table 2. Mean comparison of three years' data collected on thousand Kernel weight and total seed yield

Treatments	TKW(g)	TSY(Qt/ha)
Caged with honeybees (T1)	3.05±0.09 ^a	19.38±0.46 ^a
Caged without honeybees (T2)	2.16±0.11 ^c	14.39±0.28 ^c
Open Pollination (T3)	2.55±0.09 ^b	16.80±0.75 ^b
Over all mean	2.57±0.08	16.88±0.43
LSD	0.27	1.52
CV(%)	14.42	12.25

a,b,c= means with different superscripts within a column are significantly different (P<0.05), NS= none Significant, TKW= thousand Kernel weight, and TSY= Total seed yields

In the present investigation conducted at Sinana Agricultural Research center at on-station, the result revealed that plots caged with honeybees had yield advantage of 29.70% in Coriander crop over control.

Insect pollination enhanced average crop yield between 18 and 71% depending on the crop Bartomeus *et al.* (2014). This might be because of Coriander pollination was highly affected by mode of insect pollination.

Conclusion and recommendation

Visits of honeybees in Coriander of total seeds yield increment by 19.38 quintal per hectare and seed yield related components produced. Thus strategies to promote pollination by honeybee may be helpful in enhancing seed yield of *Coriandrum sativum*. Therefore it is recommended that honeybees should be used as an input to increase seed yield of *Coriandrum sativum*.

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Minimizing effect of honey badger (*Mellivoracapensis*) through management controlling methods in Borana Zone Southern Oromia

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Abstract

The experiment was conducted at Yabello and Arero districts of Borana zone to evaluate effectiveness of honey badger controlling methods and reduce honey badger destruction in the study area. For this study locally adapted honeybee Apismellifera colonies were used. Three honey badger controlling methods: Wood lock, Wire tying, Iron sheet and Control were used and compared. In the experiment, there was significant (χ^2 $p < 0.05$) variation between Honey badger controlling methods in terms of colony absconding rate and degree of damage to honeybees. Accordingly, it was predicted that there would be minimal damage to the produced honey when iron sheet was encircled 1.5m high hives stand method is use followed by wood lock method. Honey badger resulted intensive trial damage to wire fixed and conventional (control) beehive units. Overall, the actual damage/absconding rate to bee colonies was minimal (7.64%) in all controlling units except the control (53.85%). The benefit-cost ratio analysis was 5.09, 4.01 and 3.44 for wire tying, wood lock and iron sheet encircled 1.5m high hive stand honey badger controlling methods, respectively. The values of benefit cost ratio of all controlling methods were greater than one implying that all methods were profitable in honey production. The study shows that all treatments effectively prevented honey badger attack from hives with significant variance in terms of preventive success rates and economic feasibility. But, placing the hive at 1.5m high above the ground with hive stands rapped by smooth iron sheet was found to affect monitoring as easily as the other methods. Therefore, Wood lock honey badger controlling management method is suggested for further promotion for an area where this animal is a honey production concern.

Key words: Honey badger, Apismellifera, absconding, Borana zone

Introduction

Different mammals may be considered enemies of honey bees. In general, they prey on colonies for honey and brood. Honey badger is a mammalian bee enemy characterized by short, strong legs; elongated feet and straight; strong toes adapted to burrowing. It is heavily furred, distinctly marked, and very strong. They live in dens and are mostly nocturnal. They have perineal glands, which emit a fetid odor. This secretion is causing honeybees to abscond and making others to become moribund (Neal and Cheeseman 1996). According to Kigatiira (1984) honey badgers, empty a hive by repeatedly holding their tail in front of the entrance. The disturbed bees attach themselves to the tail, whereupon the badger transports them away and returns to the unguarded honey.

In Africa, honey badgers are considered the most destructive mammalian predators of honeybees (Hepburn and Radloff, 1998) and capable of destroying more than twenty hives in a single night (Guy

1972). Kingdon (1989) showed that despite the precautions taken by traditional beekeepers in Tanzania, 2,700 hives out of 24,000 (11.25%) were damaged in a single year and it concluded that commercial beekeepers would not be prepared to lose 10% or more of their production to these animals. Reports of honey badgers raiding apiaries have been recorded from South Africa, Botswana, Angola, Zimbabwe, Malawi, Mozambique, Tanzania, Zaire, Kenya, Uganda, Senegal, Togo-Benin, Nigeria, Somalia and Ethiopia (Hepburn and Radloff, 1998).

In Ethiopia like other African countries, the honey badger is reported as the most widespread enemies of honeybee (Desalegn B., 2015). Eventhough, it is the most common predator and challenging concern of the beekeeping sector in Ethiopia, there is no strategic mechanism for prevention and controlling of honey badger to minimize the loss of honeybee colonies and their product. Some reports indicated that traditional beekeepers prevent honey badger by rapping corrugated iron sheet on a tree trunk and tying a bunch of thorny branches on the stem of the tree on which the bee hive is hanged on; therefore, these two methods make it uncomfortable for the animal to climb. Generally, both traditional and modern beekeeping systems of Ethiopia are making use of these methods to protect this enemy. According to (Tamirat, *et.al*, 2013 unpublished) this predator was identified as a serious bee enemy and one of the top challenging predators next to ants for beekeeping development in the Borana zone. For this reason seeking different mechanism of controlling methods is found to be crucial for developing and implementing cost effective controlling methods against honey badgers in order to boost honey production and beekeeping practice in the zone.

Objectives

- To evaluate effectiveness of honey badger controlling methods
- To reduce honey badger destruction in the study area

Materials and methods

Study site

The experiment was conducted in Borana zone at Yabello Pastoral and Dry-land Agriculture Research Center (YPDARC) apiary site and Arero district where the animal occurrence was confirmed.

Experiment setups

For the study three different honey badgers controlling methods were applied with untreated check. At each site, 12 Movable frame (modern) hives and a total of 24 bee hives were used to undertake the activity. For the movable frame hives foundation sheets were prepared and fixed to the frames. At each study site, local honeybee colonies were transferred to all the movable frame (modern) hives at the beginning of active season. All honeybee colonies were managed under the same condition and brought to the uniform strength.

Experimental design

Three honey badger controlling methods (Wood lock =group 1, Wire tying=Group 2, Iron sheet=Group 3 and Control=Group 4) were developed and compared: The detail of preparation is presented as follow.

Wood lock

Three colonies were placed on a standard hive stand between two woods supporting posts and strong upper restricting stick. For these two strong wood posts that has 1.08m long were selected and placed on either side of the hive. While 30cm of the posts part will be used to fix the either posts in the ground, the left portion of the post will be used to restrict the movement of the hive side wards. At 78cm from the ground a single hole will be created using on the two posts used with any piercing material to place a strong horizontal stick that has 1m length to restrict the upward movement of any hive. For the second supper the hole will be created at 1.06m long from the ground to place the second horizontal stick for the first supper and this process will continue for the rest of the suppers with 25cm gap. Methods like placing iron sheet on the posts will be used to protect bees from ants.



Figure 1: Wood lock



Figure 2: Wire tying

Wire tying

Three colonies were placed on standard hive stand and hives were fixed to the hive stand by strong wire.

Iron Sheet

Three colonies were placed on the hive stand prepared at 1.5m high above the ground with hive stands rapped by smooth iron sheet.



Figure 3: Iron Sheet

Control

Three colonies were placed on the standard hive stand with no honey badger controlling methods applied and used as untreated check

Data collection

At each site researchers, technical assistants (TAs) and experienced beekeepers were visited every morning throughout the study period.

Data on number of colonies attacked and destructed by honey badger were recorded by observing attack and any sign of trail to attack the colony which include damaged combs with brood and honey. In addition to this average honey yield per harvest/ colony and colony absconding tendency were also recorded for each treatment group during the study period.

Except the experimental colonies, bee hives and seasonal managements all costs/expense used to make treatments were recorded to compare the cost effectiveness of the improved practice of controlling methods.

Data analysis

To examine the effect of honey badger controlling methods on absconding, Chi-Square (χ^2) procedure of SASv.9 was applied.

Gross Margin (GM)

To identify the economically feasible honey badger controlling methods of used alternatives Gross margin (GM) was computed

Gross margin the difference between the Gross Return (GR) and the Total Variable Cost (TVC) was computed with the following empirical formula

$$GM = GR - TVC$$

Benefit-Cost Ratio (BCR)

Benefit-Cost Ratio (BCR) which is given by the ratio of gross return to total variable costs was used to determine the most beneficial honey badger controlling methods among the implemented treatments.

$$BCR = \frac{GR}{TVC}$$

If the ratio is less than one, then the costs exceed the benefit. However, if the ratio is more than one then the benefits exceed the costs (Gittenger, 1982; Jehanzeb, 1999).

Results and Discussion

Hive damage by honey badger

From the current study there is significant ($\chi^2 p < 0.05$) variation between Honey badger controlling methods. Accordingly, it was predicted that there would be minimal damage to the produced honey when iron sheet encircled at 1.5m high hives stand method above the ground is used. The remaining two, Wood Lock and Wire tying are found to be better than the control. However, there is no significant difference was found between controlling methods in the extent of hive damage. Honey badger resulted intensive trial

damage to wire fixed controlling method as compared to conventional (control) beehive units. Overall, the actual damage/absconding rate to honeybee colonies was minimal (7.64%) in all controlling units except the control (53.85%) Table 1. Commutatively, the alternative honey badger controlling methods were found seven times (7) times better than the conventional/control.

Table1. The effect of different honey badger controlling methods on absconding rate honeybee colonies,

Treatment/Deterrent	Abscond		Stayed		Chi-Square Tests		
	Frequency	Percent	Frequency	Percent	χ^2 Value	DF	$p < 0.05$
Iron Sheet	0	0 ^a	11	100.00 ^a	14.43	3	0.0024
Wood post	1	6.25 ^a	15	93.75 ^a			
Wire	2	16.67 ^a	10	83.33 ^a			
Control	7	53.85 ^b	6	46.15 ^b			
Total	10	19.23	42	80.77			

Percent with different superscript under the same column are significantly different at $p < 0.05$

Of the entire treatments used, the iron sheet was found the most promising controlling method than other treatments. This is due to the fact that smooth iron sheet rapped to hive stand made unsuitable for honey badger to climb.

Other scholars (Shankute *et al*, 2012; Alemayo A. *et al*, 2016 and Bekele T. *et al*, 2017) reported that the use of indigenous knowledge of beekeepers by hanging bee hives to long smooth base trees encircled with smooth iron sheet on the trunk of the tree for making it not conducive for honey badger to climb. However, the higher a hive is raised, the harder it is for the beekeeper to work with the hive. The hive should be hung at approximately waist level to the beekeeper for ease of working.

Cost benefit analysis

Regarding total variable cost (TVC) the iron sheet encircled at 1.5m high hive stand method is found the most expensive (1310 ETB) followed by wood lock (1090 ETB) and wire tying (800 ETB) methods. Consecutively the control is found the least in terms of TVC (760 ETB). However, regarding BCR the control least of all tested honey badger controlling mechanisms. The benefit-cost ratio was 5.09, 4.01 and 3.44 for wire tying, wood lock and iron sheet encircled 1.5m high hive stand honey badger controlling methods, respectively, implying that wire tying method is the most profitable than others.

Table2. Cost-benefit analysis of honey badger controlling methods

Particular	Honey badger controlling methods			Control
	Iron sheet	Wood lock	Wire tying	
Honey yield/hive (kg)	7.76	7.76	7.76	7.76
Honey price /kg (birr)	250	250	250	250
Total cost /treatment (Birr)	1310	1090	800	760
Total revenue/treatment (Birr)	5820	5456.25	4874.25	2685.93
Benefit cost ratio	3.44	4.01	5.09	2.53
Gross margin/ treatment (Birr)	4510	4366.25	4074.25	1925.93

The data of costs for the respective honey badger controlling method is collected to analyze benefit cost ratio (BCR). Accordingly, costs of wire, nails, wood lock, iron sheet, hammer and labor for those methods that incorporated the mentioned expenses are all considered. However, the detail is not presented and rather the summation of all costs under a particular controlling is given in table 2. Based on the collected data, the t test analysis of the benefit cost ratio (BCR) was performed (Table 3). Accordingly, the result showed that there is a significant difference ($p < 0.05$) between controlling methods.

Table3. Analysis of Benefit Cost Ratio (BCR) from one sample statistic

N	Mean	T	Df	P < 0.05
4	3.75±0.48	7.833	3	0.004

Conclusions and Recommendations

All treatments effectively prevented honey badger attack from hives with significant variance in terms of preventive success rates and economic feasibility. Unmanaged/conventional placing of hives was mostly exposing honey bee colony to predator (honey badgers)

The tested Honey badger controlling methods were performed differently in minimizing honey badger damage with variable cost. In terms benefit cost ratio (BCR), all controlling methods in the study area within the study period was showed values >1 implying that all methods were profitable in honey production. Overall, in terms of longterm service and higher level efficiency is required, the iron sheet encircled to 1.5m high method is the preferable one followed by wood lock method. But, placing the hive at 1.5m high above the ground with hive stands rapped by the smooth iron sheet was found to affect monitoring as easily as the other methods. Because, the higher a hive is raised, the harder it is for the beekeeper to work with it. Therefore, based on price, ease frequent hive management and decent level of efficiency wood lock honey badger controlling method is the most promising one. Based on the current finding points of view, the following recommendations is proposed

Training beekeepers to reduce persecution through the use of honey badger controlling methods is needed. Wood lock honey badger controlling management method is suggested for further promotion for an area where this animal is a honey production concern.

Further studies on beekeeper's preference and adoption to controlling methods for its applicability.

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Assessment of current status, nesting ecology and potential threats of stingless bees in selected districts of East Wollega Zone, Oromia Regional State, Ethiopia

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ABSTRACT

The study was conducted in selected districts of east Wallega zone (Diga, Sasiga and WayuTuka) from December, 2018 to April, 2020 to determine current status, nesting ecology and potential threats of stingless bees. Based on respondents and visual observation, the beekeeping activities have been practiced as a sideline with other agricultural activities. Majority of the respondents have been finding stingless bee nest during their farm practice. The technique to identify the natural nesting site of stingless bee were; Lay on the ground with their chest around the nest site, Searching the nest entrance whole and Searching the stingless bee during foraging. Most of the farmers harvested stingless bee honey from natural colonies in the forest and farmland by digging out the ground. Stingless bee colonies were decreasing with its products. In terms of Meliponiculture, some of respondents were shifting traditional ways of stingless bee keeping to adaptation of stingless bee in man-made box hive. The stingless bee honey production in farm land area was more decreasing than in forest area due to natural habitat losses. Based on respondents and visual observation, the stingless bee's colonies and nests were found in natural forests, in enclosure area, around the river, in communal grazing area and in farm area respectively. The more stingless bee colonies and its nest found in natural forest and protected area that provide them with a suitable cavities and nest preparation. The major opportunities for stingless bee keeping in the study area was; non-aggressive and has no side effect of stingless bees, availability of bee forage and water, honey as medical value, available of stingless bee colonies and indigenous knowledge of farmers on stingless bee colonies. The major problems in Meliponiculture were; Habitat loss, lack of awareness and extension service, Poor indigenous knowledge sharing culture, agrochemical application.

Key words: *stingless bee, threats, ecology, Oromia,*

INTRODUCTION

Stingless bees have populated tropical earth for over 65 million years – longer than *Apis*, the stinging honey bees (Michener, 2000). Stingless bees (Hymenoptera, Apidae, and Meliponini) are a group of small- to medium-sized bees with vestigial (non-functional) stings. They belong to the meliponinae, one of three subfamilies of the family Apidae and occur in recurrent colonies where they store honey, pollen, propolis and royal jelly. Social organization in stingless bees is highly developed and can be comparable to that of honeybees (Sakagami, 1982).

Stingless bees, like the honey bees of the genus *Apis*, live with many individuals in a nest where honey and pollen are stored. Although the amounts of honey are generally smaller than in the nests of honey bees, people have used stingless bee honey for many centuries. They are eusocial insects that play an

important role in the pollination process of plant life, particularly plants in natural habitats in most tropical countries (Heard, 1999). The raising and farming of stingless bees, has been sparking interest for the past few years, not only in rural communities income generation, but also in the gourmet market, where professionals use the product, either for its medicinal properties or for its outstanding flavors. Honey produced by stingless bees have been widely relished in the past and besides their putative medicinal properties there are overbearing traditional reasons to harvest honey from pots, either from the forest or with the comfort of a well-established meliponary (Vitet *et al.*, 2004).

Stingless bee honey is a valuable natural product from a diverse group of highly eusocial bees comprising the tribe Meliponini in the family Apidae. Stingless bees produce honey from nectar of flowering plants. It is stored in pots that are made of wax cerumen. Honey is mainly made up of glucose and fructose but contains minerals, vitamin and other nutrients. It is the main energy source for the bees and can serve as energy booster for humans (Peter K., 2010). Stingless bees store honey in pots rather than in combs. Their honey is also nectar or honeydew that has been concentrated and transformed. The honey from stingless bees is highly appreciated traditionally considered to be more powerful as a natural medicine for treating common diseases than honey of honey bee, (Lubertus B *et al.*, 2006). One possible reason for such medicinal application is that the honey is usually kept in pots of cerumen. During storage, the honey may acquire some components of cerumen, known to have several medicinal effects (Drummond, 2013).

Unlike honey bees, which store honey in neat, regular combs, stingless bees fashion lumpy honey pots. Although their honey and other hive products are less abundant, it is more difficult to collect than those of honey bees. Their nests is typically in hollow trees or other cavities, the bees stockpile both honey and pollen in lumpy little pots fashioned from cerumen which is a mixture of beeswax and plant resins. Honey is frequently collected from natural colonies in the forest. This often leads to the destruction of the nests, and often to that of the tree as well. One of the main issues for practical Meliponiculture is how to extract honey. When a beekeeper wanted to remove the honey, traditionally, they open at the rear the log or chamber and then perforate all honey pots, to allow the honey to drain and be collected. Here the honey passes through the garbage area of the colony and is contaminated. Also, this procedure causes a lot of damage to the colony, which loses several food pots and causes a high mortality of adult bees and the loss of a great percentage of the laid eggs. (Villanueva *et al.*, 2005).

Stingless bees (Apidae: Meliponini) are major pollinators of many wild and cultivated plants, and both indigenous and non-indigenous populations use their products for diverse purposes including food, crafts, and medicine. However, many aspects of both stingless bee nesting ecology and traditional knowledge of these culturally significant bees by diverse human populations remain unknown or poorly documented. (Victor H *et.al.* 2018).In the studyarea the current status, nesting ecology and potential threats of stingless bees was not identified, well organized, documented and the study was focusto explore the nesting ecology, current status of stingless bee population, challenges and opportunities that affect stingless bee colony and its products.

2. Materials and Methods

2.1. Description of the Study Area

The study was conducted in East Wollega Zone, Oromia Regional state at about 332km away from Addis Ababa, and the capital city of Ethiopia. It is bordered on the southwest by Buno Bedele Zone, on the west by the Didessa River, which separates it from West Wollega, on the northwest and north by

the Benishangul Gumuz Region by the northeast by Horo Guduru Wollega, on the east by West Shoa, and on the southeast by the Gibe River which separates it from Jimma Zone. The zone is located in the area stretching from 36 0 30'00" to 36 0 45'00" longitude and 9 0 05'00" to 9 0 15'00" latitude with elevation ranging from 1000m to 3207m. The range of annual rainfall of the zone is from 1500mm to 2200mm with mean annual temperature 15-20 degree centigrade (CSA 2005, 2007). The study was specifically conducted in three districts; Diga, Sasiga and WayuTuka.

Diga district is one of East Wollega Zone, Oromia Regional State. The Woreda is located at about 346 km away from Addis Ababa and 15km from Nekemte town to the West. Based on agro-climatically conditions namely: Highland altitude ranges 2100-2342m and Midland ranges 1200-2100m with annual rainfall of 2400mm (JoshaO,*et al*; 2010, CSA 2007).

Sasiga district is one of the wored in the Oromia Regional state and a part of the EastWalaga Zone. Sasiga is bordered on the south byDiga, on the west by the Benishangul-Gumuz Region, on the northwest by Limmu, on the north by an exclave of the Benishangul-Gumuz Region and on the east by GutoGidda. The administrative center of this woreda is Galo and 42 Km from Nekemte, the capital of East Walaga Zone.

WayuTuka district is located 324 km from the capital Addis Ababa at an altitude of 1700–2200 m above sea level and has an average annual rainfall of 2400 mm (CSA 2007).

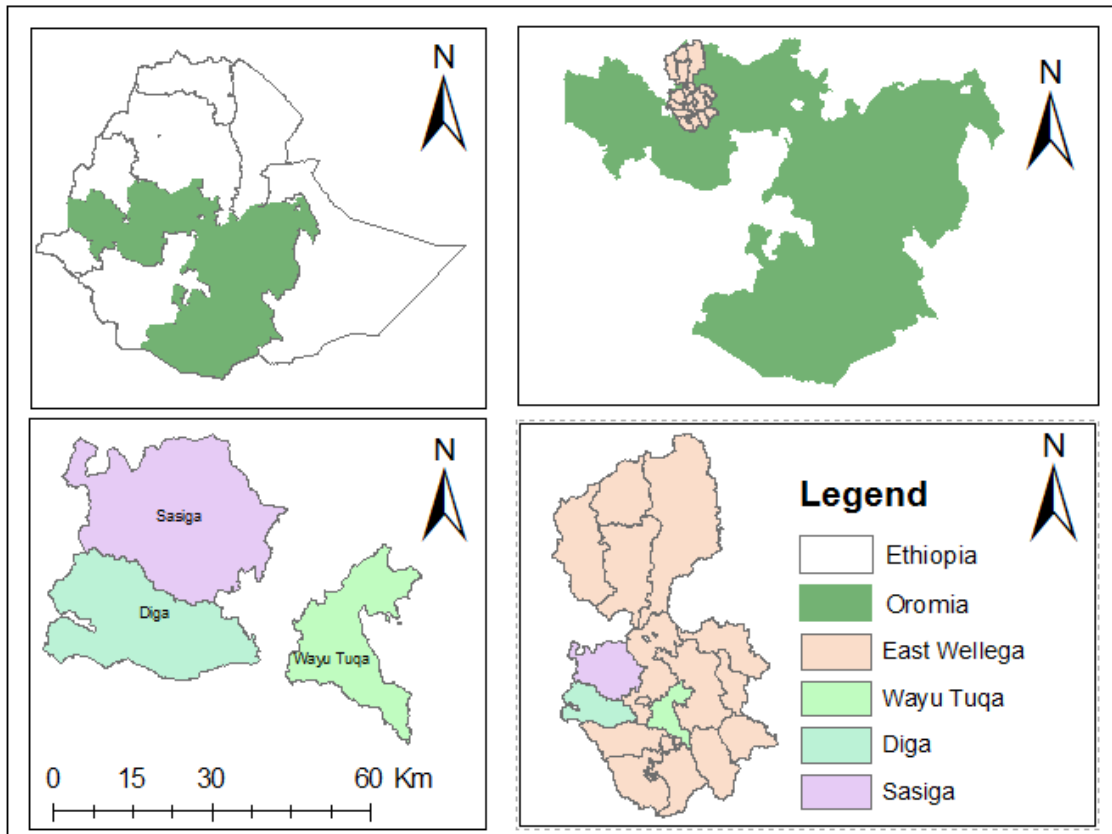


Figure 1 Map showing the location of the study area

2.2. Data sources and methods of collection

In this study, both primary and secondary sources of data were used. The primary data was collected from sample household beekeepers through a semi-structured questionnaire and by field examination of stingless bee nests. Secondary data was obtained from various sources through desk review.

2.3. Sampling methods

The study was purposively select three districts based on their intervention in stingless bee harvesting and domestication. The entire farmers who were involved in stingless bee honey harvesting were clustered in to those who were engaged in domestication and those who do not involve in domestication. Atotal 153 farmers and out of these 10 farmers who are involved in domestication were selected purposively and the rest of 143 farmers were selected using simple random sampling from those farmers who are not involved in the domestication of stingless bee but involving in harvesting of stingless bee honey.

2.4. Data Collection Methods

The study was based on qualitative and quantitative data collection methods. The qualitative approach was the dominant approach because it tends to give more attention to the subjective features of human knowledge and behavior (Powell and Connaway, 2007). Accordingly, interview, focus group discussions (FGD), and field observation were used.

2.5. Field Observation

Field observation was used to obtain information on farmers' indigenous knowledge that was not captured through interview and group discussions, to crosscheck the actual practices and to capture pictures.

2.6. Data management and statistical analysis

The collected data were stored in Microsoft Excel and SPSS software programs (SPSS @, version 20) for analysis. The statistical analysis used in the study varied depending on the type of variable and information obtained. Summarized data was presented in the form of tables and figures.

The data collected through semi structured questionnaires were analyzed using descriptive statistics and the ranking of the different types of beekeeping constraints, Common Cause of stingless bee colony and yield decrease obtained in the study were done by using the rank index formula as described by (Musa *et al.*, 2006):

Rank index=*sum of (5 * number of household ranked first + 4 *number of household ranked second + 3 *number of household ranked third + 2 *number of household ranked fourth + 1 * X number of household ranked fifth) for an individual reason divided by the sum of (5 * number of household ranked first + 4 *number of household ranked second + 3 * number of household ranked third + 2 * number of household ranked fourth + 1* number of household ranked fifth) for overall reasons.*

3. RESULT AND DISCUSSION.

3.1. Beekeeping activities and potentials

Based on respondents and visual observation the beekeeping activities have been practiced sideline with other agricultural activities. There were no any respondents who depend only on beekeeping. Most beekeepers were started beekeeping before 2005(3.80%),2006-2010 (10.89%), 2011-2015 (18.41%)and after 2016(28.03%)in increasing respectively (table 1)and this indicate these beekeepers were related within age of between 31-42 years. Based on household respondents, beekeeping practice was increasing in the studyarea with beekeeping technology. Majority of beekeepers started after year 2016.

Table 1. Beekeeping starting time

Year	Frequency	Percent
Before 2005	9	5.88
2006-2010	26	16.99
2011-2015	42	27.45
After 2016	76	49.67

3.2. Indigenous Knowledge system in stingless bee honey production

The study interviewed 153house hold respondents who have been involved beekeeping and stingless bee production in order to extract the indigenous knowledge use and practices in stingless bee honey production.

3.2.1.Occasion of nest site location finding

Majority of the respondents have been finding stingless bee nest during arm practice since all respondents were farmers and some of respondents have been got stingless bee nests during keeping their livestock Table 2.

Table .2 Occasion of nest site location finding

Occasion of nest finding	Frequency	Percent
Keeping livestock	42	27.45
Farm practice	67	43.79
Fuel wood Collection	13	8.49
Only singles bee nest finding	31	20.26

3.2.2. Identification methods of the wild nesting site

According to the respondents, however, it is a difficult task to identify the wild nesting site of stingless bee. The technique to identify the natural nesting site stingless bee presence by ranks were Lay on the ground with their chest around the nest site (as 1st), Searching the nest entrance whole (as 2nd) and Searching the stingless bee during foraging (as 3rd) and others method of identifying the natural nesting site Table 3.

Table. 3 Identification methods of nest site

Identification methods of nest site	Relative degree of importance's					index	Rank
	1st	2nd	3rd	4th	5th		
Lay on the ground with their chest around the nest site	19	15	9	1	0	0.32	1
Searching the nest entrance whole	12	19	6	4	2	0.29	2
Searching the stingless bee during foraging	1	9	22	17	3	0.19	3
Searching the stingless bee during watering	0	3	7	8	27	0.071	5
Ting the stingless bee using thread	0	5	3	7	23	0.04	6
Using indicator insects or honey to attract the stingless bee	1	2	7	28	2	0.09	4

The farmers lie on the ground by their chest around the nest site to see the stingless bee while entering and leaving their nest/ observe bees flying in and out of the nest. The farmers used to search this tube in the areas around homestead, farmland and forest (Kwapong *et al.*, 2010) and also other respondents stated that the stingless bee's nest can be identified by searching the entrance holes on trunks of trees and fallen logs. However, according to the respondents this method is time consuming and less effective compared to the other methods.

3.2.3. Domestication of stingless bee and colony transfer methods

The survey result shows that in the study area some farmers are starting to maintain stingless bee by artificial hives through domesticating these wild bees. The domesticators employ two ways of maintaining the stingless bee species in traditional hives made mud greased by cow dung (6.54) and most of the respondents maintain at their original nest site cavity (93.45%).



Figure 2. Domestication of stingless bee

3.2.4. Stingless bee honey harvesting methods

Stingless honey harvesting was take place in four ways, from the wild colony and from domesticated colony in their home. Honey harvesting in domesticated colony was more sustainable way compare to harvesting honey from the wild colony. The respondents clarify that honey harvesting from domesticated colony was easier, efficient and quicker compare to natural nest colony in the wild.

Table 4: Ways of stingless bee honey harvesting.

	Way of harvesting	Frequency	Percent
From wild colony	Without taking care of the colony	27	17.65
	With taking care of the colony	19	12.41
	By taking care of honey	71	46.40
	Taking care for colony and honey	36	23.53
From domesticated colony	Without taking care of the colony	0	0
	With taking care of the colony	0	0
	Only fir taking care of honey	1	0.65
	Taking care for colony and honey	9	5.88

Most of the farmers harvested in stingless bee honey from natural colonies in the forest, farmland and around homestead by digging out the ground. The ground-nesting stingless bees build their nests in the soil, with only the nest entrance visible. According to the respondent, the height of the excavated underground cavity varied from 0.5m up to 0.75mand also 0.75m up to 1m based on the property of the soil area. From this, it can be understand that how honey harvesting in wild colony is difficult. The result shows that, stingless bee honey was harvested from the wild colony in two ways; harvesting the honey in their natural underground cavity without taking care of the colony and harvesting the honey with taking care of the colony by maintaining the nests in their natural underground cavity.

3.2.4. Stingless Bee Honey Yield status

According to respondents the amounts of stingless bee colony and their product was decreasing. In terms of Meliponiculture, some of respondents were shifting traditional ways of stingless bee keeping to adaptation of stingless bee in man-made box hive. The stingless bee honey production in farm land area was more decreasing than in forest area due to natural habitat losses. The traditional way of clearing forests for agricultural farm disposes the ground to degradation agents, hence an environmental threat. It is a well demonstrated fact that stingless bee domestication and conservation would significantly impact on food security, besides increased household income.

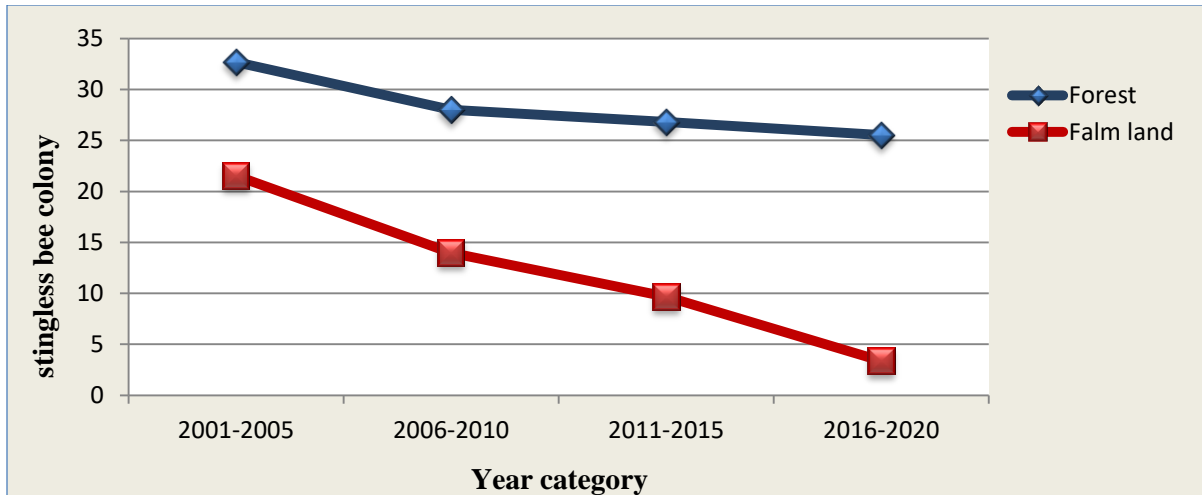


Figure 3. Stingless Bee colony Yield status

3.2.5. Stingless bee nesting area (ecology)

Based on respondents and visual observation, the stingless bee's colonies and nests were found in natural forests, in enclosure area, around the river, in communal grazing area and in farm area respectively. The more stingless bee colonies and its nest found in natural forest due to its availability of resources for survival or bets habitat.

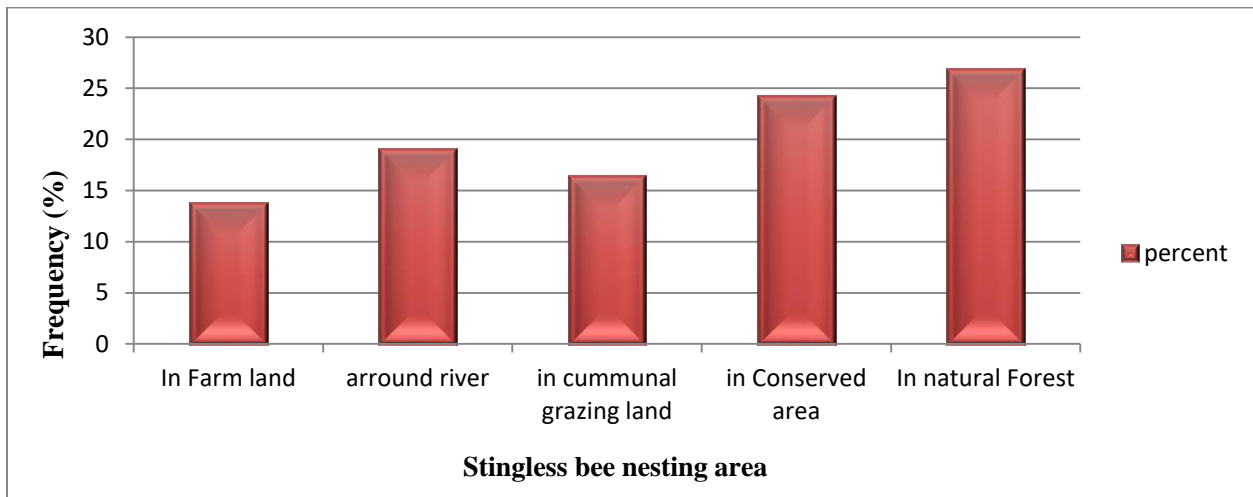


Figure 4. Stingless bee nesting area

Understanding the scale at which habitat influences species richness in ecosystems is central to ecology (Wettstein & Schmid, 1999) as both patch and landscape factors may contribute to the diversity of resident taxa (Collinge, *et al.*, 2003). Natural and anthropogenic disturbance affects the vegetation structure and composition. Due to anthropogenic ally mediated habitat, changes are taking place at multiple scales; science must distinguish between cover and landscape threats in order to develop effective conservation strategies. Community composition may be influenced by habitat variation from area to landscape-scale

depending on body size, home range area, and dispersal distance of the taxa of interest (Haskell, *et al.*, 2002).

3.3. Opportunities and Challenges of Meliponiculture

Ethiopia has immense natural resource for beekeeping and Meliponiculture activity. However, like any other livestock, this sub sector has been ceased by complicated constraints. The main production constraints in stingless bee productivity would vary depending on the agro ecology of the areas where the activities is carried out.

3.3.1. Opportunities

According to the respondents, the major opportunities for stingless bee keeping in in the study area include: non-aggressive and has no side effect of stingless bees, availability of bee forage and water, stingless bee honey as medical value, available of number stingless bee colonies and indigenous knowledge of farmers on stingless bee colonies Table 5.

Table 5. Opportunity of Meliponi culture

Opportunity	Relative degree of importance									Rank Index	Rank
	1st	2nd	3rd	4th	5th	6th	7th	8th	9th		
Non-aggressive and has no side effect	18	12	8	12	7	1	1	0	1	0.15	1
High demand and price value	15	12	8	12	7	1	1	0	1	0.14	2
Availability of bee forage and water	8	9	8	19	5	2	1	1	1	0.13	3
Hone as medical value	1	2	7	14	9	7	4	2	3	0.12	4
Available of number stingless bee colonies	1	2	7	18	4	2	3	0	5	0.11	5
Indigenous beekeepers knowledge	2	5	9	11	5	3	1	0	1	0.09	6
Existence of soil and water conservation and area enclosure	0	1	1	3	7	17	3	0	2	0.08	7
Meliponiculture experience of the farmer	0	1	1	1	9	5	12	2	1	0.07	8
Drought resistance	0	0	1	1	1	3		7	15	0.05	9

Stingless bees and honeybee colonies are also becoming important alternative pollinators to the honeybee, previously considered a universal pollinator, due to their abundance and adaptability, besides behavioral traits, which further enhance their suitability. Honeybees have emerged over the years, with great economic returns. Stingless bees do not sting humans or animals, making them easily acceptable to the bee farmer. They can be managed easily on pollen substitute and honey. Stingless bees honey has collected for medicine to alleviate various ailments and discomforts such as constipation.

3.3.2. Major Constraints of Meliponiculture

According to the results of the study, the major problems in Meliponiculture arise from bee characteristics or environmental factors that are beyond the control of the stingless beekeepers, the others problems mentioned by the respondents were; Habitat loss, lack of awareness and extension service, Poor indigenous knowledge sharing culture, agrochemical application (Table 6). Moreover it is also the lack of understanding on behalf of Meliponiculture combined with lack of regulations and enforcement that has enabled the increasingly rapid spread of pathogens during the past thirty years

Table 6. Challenges in Meliponiculture

Challenges	Relative degree of importance								Index	Rank
	1st	2nd	3rd	4th	5th	6th	7th	8th		
Habitat loss	17	13	9	11	5	3	1	0	0.25	1
Lack of awareness and extension service	10	21	6	4	2	0	0	0	0.21	2
Poor knowledge sharing culture	1	3	17	9	5	1	2	1	0.14	3
No attention and effort by government	1	2	7	18	4	2	3	0	0.12	4
Agrochemical application	0	5	3	7	15	2	0	1	0.11	5
Difficult to find nesting site	0	1	1	3	7	17	3	0	0.08	6
Lack of appropriate technology	0	0	1	1	9	5	13	2	0.06	7
Low productivity	0	0	1	1	2	5	9	10	0.04	8

The potential and levels of habitat loss due clearing forest for agricultural farm and grazing in the area has great effect on stingless colony. The rates of habitat degradation and destruction in tropical forests are almost greater than in any other biome in the world (Sala *et al.*, 2000;). Honey hunting for stingless bee honey had severe effects on stingless bees by killing bee colonies and leaving cavities unsuitable to be re-used by stingless bees.

4. Conclusion and Recommendation

From the result stingless bees prefer nest in forest and conserved land area that provide them with a suitable nest site. Landscapes and land uses can influence the nesting behavior of stingless bees due to their dependence on various substrates for nesting. From searching the stingless bee nest, lay on the ground was used as effective method with during farm practice as stingless bee nest finding. Harvesting the honey in the wild colony was only taking care for honey however from domesticated taking care for both colony and honey. According to respondents the amounts of stingless bee colony and their product was decreasing. Challenges in stingless bee include poor extension service, lack of awareness for farmers, low attention and effort by the government, lack of appropriate technology, and low productivity. The opportunity for stingless bee identified was high and price value of stingless bee honey, medical value of stingless honey, farmer's indigenous domestication and non-aggressive behavior of stingless bee. Awareness of stingless bee domestication and Meliponiculture equipment is important to support from agricultural Research, other experts and efforts should be made from concerning organizations as recommendation. These findings suggest that various landscapes and land uses can influence the nesting

behavior of stingless bees due to their dependence on various substrates for nesting. For the reason of time restraint in this study area, awareness creation and attention to driving force of challenge and opportunity of stingless bee is suggested by monitoring throughout the year.

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Adaptation trial of improved bee forages in West Hararghe Zone of Oromia Region, Ethiopia

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Abstract

The study was conducted at Mechara Agriculture Research station and in Gemechis district kunisekeria FTC for two consecutive cropping seasons of, 2018 and 2019, with the objective of identifying adaptable and potential bee forages for honey production. Five herbaceous bee forages were sown in 2mX2m plot size in Randomized Complete Block Design with three replications. The bee forages were evaluated based on germination efficiency days to flower, number of flower heads/plant and intensity of honeybees on flowers. The result indicated that, there were mean difference for germination date, blooming date, flowering length, number of flower heads/plant for all study plant species. Among the study bee forage plants, both black and white buckwheat's were take short day to flower with mean value of 20 and 24 days. Similarly *Sinapis alba* was take a short day to flowering at Kuni sakaria FTC while *Sinapis alba* at Kuni sakaria FTC has the highest mean value in duration of blooming (45) days followed by white buck wheat at both site (43) days. Coriander has highest flower per head/plant (92) at both site followed by *Sinapis* at Kuni sakaria FTC. There was mean difference in bee visit between each plant species at all study sites. Coriander was highly visited by honey bees at Kuni sakaria FTC followed by white buckwheat at both site. In general, both black and white type of buckwheat and Coriander had high performance in lowland and highland of the zone whereas *Phacelia tanacetifolia* and *Sinapis alba* showed good performance at highland part of the study area. Therefore, further demonstration and scaling-up is needed at recommended area for the respective similar agro ecology of the West Hararghe Zone.

Key word: Bee forage, Date to flowering, Duration of blooming, Flower perhead

Introduction

Beekeeping is one of the most important farming activities in Ethiopia (Workneh *et al.*, 2008). Ethiopia is gifted with diverse and unique flowering plants of 6000 to 7000 species. Thus making it highly suitable for large number of colonies (Admasu1996; Fitch and Admasu 1994; Gezahegn 2007; Gidey and Mekonen 2010). However, the success of beekeeping primarily depends on the availability of prevalent bee forages which is based on its population density, nectar and pollen potentiality and prolonged flowering periods (Baptist and Punchihewa, 1983). Availability of adequate perennial and annual sources of nectar and pollen is the most limiting factor in the survival, abundance and distribution of honeybees (Tura *et al.*, 2014).

There are different species of plants that are identified as major source of honey and tolerant to semiarid climate of Ethiopia. This includes, *Becium grandiflorum*, which provides white honey in northern parts of the country, *Melilotus alba*, *Fagophyrum esculentum*, are potential sources of golden honey in other parts of the world (Admassu Adi., *et al*, 2015). It was reported by the same authors *E. plantaginium*, *B. grandiflorum*, *M.alba* and *F. esculentum* performed were very well adapted under both rain fed and

irrigation condition in mid rift valley of east shoa zone. On the other hand, reports from Sinana Agricultural Research Center revealed that *Phacelia tanacetifolia*, *Sinaps alba*, *Corriandrum sativum* and black cumin are the major adaptable honeybee forages in Bale highland, being preferred by the foragers honeybees (Bekele T, et al, 2015). Depending on bee visits on forage flowers, duration on flowering and flower head production by the plants *Melilotus officinalis*, *Trifolium ruppellianum*, *Fagopyrum japonicum*, *Fagaophyrum esculentum*, *Sinapis alba*, , and *Medicago sativa* spp are found to be the most adaptable and major herbaceous bee forages under Haro sabu condition (Ofijan, 2015).

Herbaceous plants that grow as weed on cultivated field, neglected open lands, wastelands and grown as ornamentals are important source of bee forage (Edward, 1976) because they grow & flourish in short period of time and their seeds can be collected easily and sown for the next growing season. These herbaceous bee forages are seasonal and not available during the dearth period. As the result currently the scarcity of bee forage is becoming serious problem in West Hararghe zone due to drought and rapid population growth. Hence the objective of this study was to evaluate and identify the best performing honeybee forages that contribute to the production of honey and other bee products in West Hararghe Zone.

3. Materials and Methods

3.1 Description of study area

The experiment was conducted in field at Mechara Agricultural Research Center (McARC) on station, Daro Labu and kunisekaria FTC of Gemechis Districts West Hararghe Zone of Oromia National Regional State, Eastern Ethiopia. The study site located at 8⁰10'00N latitude, 40⁰30'00 E longitude and the altitude ranges from 1300 -2450m.a.s.l. The area constitutes different farming system (mixed farming, agro pastoral and pastoral) and predominantly known for cash crop production particularly coffee and chat (Dereje *et al.*, 2013).

3.2. Seeds Collection

For this study, five annual bee forages, *Sinapis alba*, *Phacelia tanacetifolia*, *black Fagopyrum esculentum*, *white Fagopyrum esculentum* and *coriander* were collected from HBRC, SARC and evaluated for their performance at Mechara Agricultural Research Center on station t. Each bee forage species was sown on 2m*2m plot sizes with three replications in Random Completed Block deign. To keep proper spacing and avoid nutrient competition, spacing between rows, plots and blocks were 20cm, 1m and 1.5m respectively. The necessary agronomic practices were carried out except no fertilizer application to keep its natural growing state. Then, data like germination date, bee visit frequency (intensity of honey bee), flowering date, duration of blooming and number of flower per head/plant were recorded. At 50% flowering, number of flower per heads were counted for each species by taking 1m² plot area as well as foraging intensity of honeybees on flowers was counted starting from 7:00 a.m. to 3:00 p.m. for ten minutes at every 2-hour interval. Moreover, time from blooming to shading was also recorded.

3.2 Statistical Analysis

The collected data were statistically analyzed by descriptive statistics using Statistical Package for the Social Sciences) (SPSS) version 20.

Results and Discussion

In this study black and white type *Fagopyrum esculentum*, *Sinapis alba*, *Coriandrum sativum* and *Phacelia tanacetifolia* were planted in two different agro-ecological zones and mean values for the investigated parameters were indicated in Table 1 and 2. For illustration of the performance of the bee forages, two years' data from each of the two locations is also presented in Table 1 and 2.

Table 1. Performance of selected annual bee forages at Mechara on Station

Plant Species	GD	DF	DB	FH
Black <i>Fagopyrum esculentum</i>	8.7±0.6 ^b	23.7±0.6 ^b	35.6±0.6 ^a	23.3±2 ^b
<i>Coriandrum sativum</i>	20.3±0.6 ^a	56±1 ^a	34±1 ^a	95.3±11 ^a
<i>Phacelia tanacetifolia</i>	8.3±0.6 ^b	0 ^b	0 ^b	0 ^b
White <i>Fagopyrum esculentum</i>	9.3±0.6 ^b	23.6±0.6 ^b	43.6±0.6 ^a	24.6±2 ^b
<i>Sinapis alba</i>	7.6±0.6 ^b	0 ^b	0 ^b	0 ^b

GD=germination date, DF=date to flowering, DB=duration of blooming, FH=flower per head

Table 2. Performance of selected annual bee forages at Kunisekaria FTC

Plant Species	GD	DF	DB	FH
Black <i>Fagopyrum esculentum</i>	8.3± ^b	20.6±0.6 ^b	33.3±1.5 ^{bc}	32.6±4 ^b
<i>Coriandrum sativum</i>	20±1 ^a	54±2 ^{ab}	32.6±1.5 ^{bc}	92.6±12.5 ^a
<i>Phacelia tanacetifolia</i>	7.6±0.6 ^b	68.3±1.2 ^a	25±1 ^c	15.6±1.5 ^b
White <i>Fagopyrum esculentum</i>	8.6±0.6 ^b	22.6±0.58 ^b	42.6±0.6 ^{ab}	19±1 ^b
<i>Sinapis alba</i>	7.7±0.6 ^b	22.6±0.59 ^{ab}	45.3±0.6 ^a	41.3±7 ^{ab}

GD=germination date, DF=date to flowering, DB=duration of blooming, FH=flower per head

Germination date

In germination date, the mean of all plants species has no significant difference except coriander. *Sinapis alba*, *Phacelia tanacetifolia*, *Fagopyrum esculentum* black and *Fagopyrum esculentum* white had the shortest germination date whereas *Coriandrum sativum* has long germination date at both site.

Date to flowering

Both black and white *Fagopyrum esculentum* had short date of flowering at both Mechara On station and Kuni sakaria FTC when compared to others. On the other hand *Phacelia tanacetifolia* has long date of flowering at Kuni sakaria FTC followed by *Coriandrum sativum* at Mechara On station. (Table 1 and 2).

Blooming to shedding (Flowering length)

There was mean difference between treatment species in flowering length. *Sinapis alba* has taken longer flowering time at Kuni sakaria FTC which is not survived at Mechara On station. Similarly, *Fagopyrum*

esculentum white has long flowering time at both sites. (Table 1 and 2). Bee forage plants which have long flowering period starting from blooming to shedding are highly preferred by honeybees for continues supply of nectar and pollen to boost honey production. Honeybees have a marked preference for one kind of plant species over the other, which may be equally abundant (Tura B *et al.*, 2018).

Number of flower heads/plant

There were mean difference for a number of flower heads per plants and *Coriandrum sativum* have higher number of flower head/plant at both sites. Similarly, *Sinapis alba* at Kuni sakaria has higher number of flower head/plant followed by white *Fagopyrum esculentum* at Mechara On station. Contrary *Phacelia tanacetifolia* had the shortest number of flower head/plant at kunisekeria FTC site which is not survive at Mechara. (Table1 and 2).

Foraging intensity of honeybees

The intensity of honey bee on different study plant species were evaluated at both site in different time of a day (hr.) for ten (10) minute at every two hour interval starting from one hour(1hr) up to nine hours(7a.m - 3p.m).There was mean difference in bee visit between each plant species at all study sites. *Coriandrum sativum* at (7am) and *Fagopyrum esculentum* white at (9am) were highly visited at kunai sakaria FTC whereas *Fagopyrum esculentum* white (9am) and *Fagopyrum esculentum* black at (9am) were highly visited at Machera On station. (Figure 1 and 2). On the other hand, *sinaps alba* and *Fagopyrum esculentum* black plant species were the least visited by honey bees at kuni sakaria FTC followed by *Coriandrum sativum* plant species at Machara On station. The study revealed that the foraging intensity of honey bees in the study areas were ranged from 7a.m to 3p.m and the honey bees more active in foraging from 7am to 9am for all study plants. Foraging intensity of the honeybees were few in the early morning and late in the evening due the cold weather condition affecting the flight of honeybees. The peak foraging time ranged from 10a.m to 2pm under rain fed and the foraging time of honeybee is varying from plant species to species based on nectar secretion time, volume and concentration and time of pollen release of plants. (Tura B, *et al.* 2018).

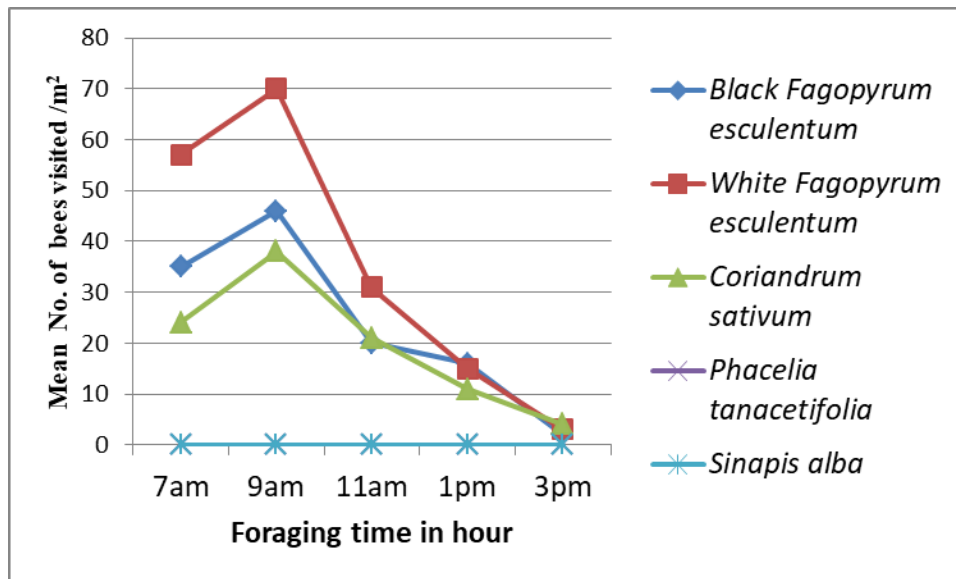


Figure1. Foraging time and intensity of honeybees at different time/hr. of days at Mechara Onstation

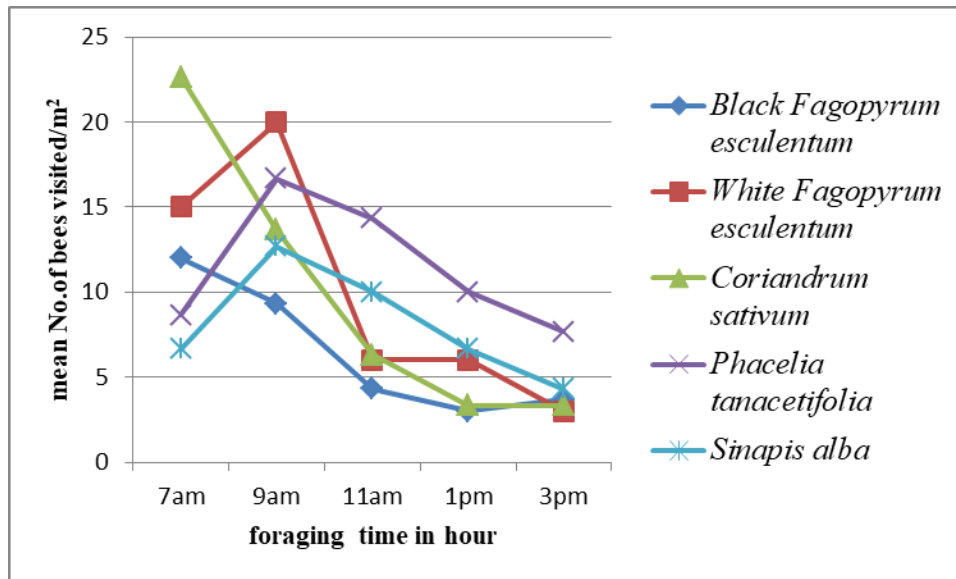


Figure 2. Foraging time and intensity of honeybees at different time/hr of days at Kunisekeria FTC

4. Conclusion and recommendations

Different herbaceous bee forages were evaluated for their performance at mechara Onstation and Kuni sakaria FTC of Gemechis district to select the best adaptable and more preferable bee forage by honey bees in order to solve the shortage of bee forage at study area. The study revealed that there were mean difference for study plants for germination date, blooming date, flowering length, number of flower heads/plant of different plant species. Additionally there were variation in foraging intensity of honeybee for the same plant species under different site due to the potential of bee forages availability at the peak flowering time and weather condition of the study plants in the study area. On the other hand, this variation was also seen for the same plant species at different time because of weather condition that exist at that time. Even though, the plants species used in this experiment showed better performance for honey bee forages at both study site (high land and lowland), the following recommendations were drawn,

- ❖ *Coriandrum sativum*, both black and white type of *Fagopyrum esculentum* were performed very well under both study areas
- ❖ On the other hand, *Phacelia tanacetifolia* and *Sinapis alba* were well performed at high land part of the zone
- ❖ Further demonstration and scaled-up is needed at high land and low land of the zone

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Nectar secretion pattern and characterizing physicochemical properties of *Coffea arabica* honey in western Oromia, Ethiopia

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Abstract:

*Coffee is an important export commodity for the Ethiopian economy and used for honey production. The study was designed with objective of characterizing Coffea arabica honey with respect to nectar secretion dynamics, concentration and volume, botanical composition, physicochemical parameters and antioxidant properties of honey. For nectar concentration and volume analysis, the study was conducted in Gera District of Jima. For this, five inflorescences were covered with fine mesh bags on a different part of the tree one day before nectar measuring commenced. From covered inflorescences, twenty flowers per tree were randomly selected and nectar volume was measured using micropipettes. For other parameters, 12 honey samples of Coffea arabica honey were collected from four districts of Jimma and Illubabore Zones. The honey pollen analysis and physicochemical properties of honey were determined using the method recommended by the International Commission for Bee Botany and Harmonized methods of the International Honey commission. The mean nectar concentration and volume of Coffee flowers were significantly different across times of the days ($p < 0.05$). The lowest mean nectar concentration was recorded at 7:00 pm and the highest nectar concentration recorded at 16:00 pm. The average nectar volume (μl) per flower in 24 hours, sugar amount per tree (g), expected honey yield per tree (kg) and honey (kg) production potential per hectare for *C. arabica* were 3.3 ± 0.2 , 0.040 ± 3 , 0.050 ± 4 and 125 kg (25-275 kg), respectively. The actual harvestable amount of honey is half of the potential (62.5 kg/ha). Pollen analysis of honey indicated that all honey samples collected from Gera, Gomma, Yayu and Manna were monofloral constituting 84%, 93%, 75% and 73 % of pollen count respectively. The mean moisture, Ash, HMF, electrical conductivity, Free Acidity, pH, Fructose, Glucose, Sucrose and Maltose content of the honey samples were, 22.48%, 0.21, 11.88, 0.49, 13.44, 3.32, 32.77, 32.9, 3.57, and 0.41 respectively. So, it can be concluded that the honey produced from area is monofloral since its pollen count exceeds more than 45% and the quality of coffee honey within range of Ethiopian and International standards. Therefore, beekeepers should focus on production of coffee monofloral honey following the calendar of the plant to exploit the honey potential of coffee and niche market opportunities for promotion and commercialization of coffee mono-floral honeys.*

Keywords: Nectar, honey quality, *Coffea arabica* and antioxidant

Introduction

Honey is complex substance and a source of nutrition that has been used by people since ancient time. It is the natural sweet substance produced by honey bees from the nectar of blossoms or from secretions of living parts of plants which honey bees collect, and combine with their own, store and left in the honey comb to ripen and mature (Gomes *et al.*, 2010). There are nearly 200 ingredients have been reported in honey which includes sugars, organic acids and secondary metabolites (Saxena *et al.* 2013). Apart from

this, honey encompasses protein, amino acids, enzymes, organic acids, minerals. (Alvarez-Suarez *et al.*, 2014). Moreover, honey is also a natural source of antioxidants which has therapeutic value against various diseases such as the anti-ageing, cancer, cardiovascular disease, reduced wound healing, gastrointestinal inflammatory diseases and Atherosclerosis (Halliwell & Gutteridge, 2015). The main cause for antioxidant effect of honey is due to presence of bioactive compounds such as phenols, flavonoids, carotenoids, vitamins, organic acids and other compounds (Johnston *et al.*, 2005; Geul & Pehlivan, 2018). The chemical composition of honey varies by its floral and geographical origin (Wang & Li, 2011). In this regards, Coffee honey is important product and contains some elements of nutritional and biological interest probably related to antioxidant effect (Kadri *et al.* 2016).

Coffee is perennial crop that belongs to the family Rubiaceae originated from Ethiopia (DaMatta 2004; Belitz *et al.* 2009). It is the most important cash crops for trading and exports since the 18th century, providing jobs for millions of families worldwide. In Ethiopia, Coffee grows in a wide range of agro-ecological conditions mainly from the range of altitudes 1200 to 3000 m (Muleta *et al.* 2011; Moat *et al.* 2017).

Coffee gives flowers profusely after rains but some coffee trees are found with flowers at any time of the year. It is a major honey source plant in south western and southeastern parts of Ethiopia (Bareke and Addi 2019a, Bareke and Addi 2018). However, the production of coffee honey is not adequately known in Ethiopia, due to lack of knowledge of the flowering calendar and management of colonies for coffee honey production. On the other hand, production of coffee honey is well known in Brazil and Indonesia where there is intensive growing of coffee plants (Kadri *et al.* 2016). Additionally, many authors have determined the honey production potential of bee plant species, based on their nectar secretion potential (Kim *et al.*, 2011; Adgaba *et al.*, 2012; Abdulaziz *et al.*, 2015; Adgaba *et al.*, 2016 and Bareke *et al.*, 2020). However, this important information is still lacking and its significance for honey production has not yet been documented for this the most economically important cash crops of Ethiopia. Therefore, this study was aimed to determine the nectar secretion dynamics, honey production potential, number of honeybee colonies required to be placed in a hectare of the coffee plantation as well as to identify the effect of temperature and humidity on the nectar secretion of *C. arabica*.

Currently there are different coffee production system in Ethiopia such as forest coffee production system, semi-forest coffee (SFC) and forest garden coffee production system (Tadesse and Feyera 2012). These production systems may provide a great opportunity of producing coffee honey by smallholder farmers and investors.

According to preliminary test using pollen analysis of honey from Yayu district in Illuabore zone of Oromia indicated that over 75% of the pollen count of Coffee pollen was found in honey samples analyzed for botanical origin. This indicated that it is possible to produce coffee monofloral honey by integrating beekeeping with Coffee production. Thus characterization of the monofloral honey from coffee plants may benefit beekeepers to exploit the niche market opportunities as organic monofloral for the commercialization of coffee honey. Therefore, it is important to authenticate botanical source of honey through pollen analysis and characterization of physiochemical properties for detection of adulteration and branding of honey. The study was aimed to characterize *Coffea Arabica* honey with respect to botanical composition, physicochemical parameters and antioxidant properties of honey.

Materials and methods

Decryption of the study area

This study was carried out in the main coffee-growing districts of Gera, Goma, Mana and Yayu in Jimma and Illuababora zones of Oromia regional states (Fig. 1). The landscape is dominated by small-scale agriculture, including coffee plantation and larger remnants of continuous forest dominated by wild forest coffee. Honey production is common practice with both modern and traditional beehives. The coffee normally starts to flower in January or February, after short rains, and it flowers one to four times before the main rainy season.

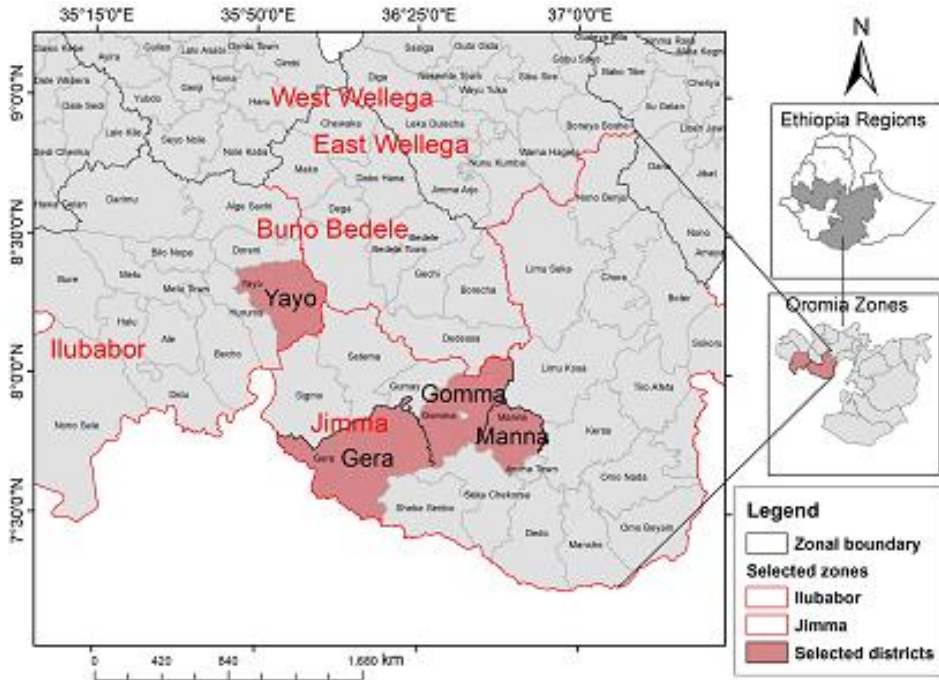


Figure 7. Map showing the study districts two zones (Jima and Buno Bedele) of Oromia.

Measurement of nectar volume and concentration

Flowering period of *Coffea arabica* was monitored from January to February during the study period. Nectar volume was measured when coffee plant produced flowers. Five (5) inflorescences were covered with fine mesh bags (40 x 40 cm) on different part of the plant, one day before nectar collection (Bareke *et al.* 2020). Flowers were marked at random from different inflorescence whorls (Wyatt *et al.* 1992). Fifteen (15) matured flower buds per plant were selected randomly bagged one day prior to anthesis using nylon mesh for each observation period. The accumulated nectar was sampled each hour, on newly opened flowers over the course of each day within the period from 8:00-18hours. To determine nectar production, the volume (μ l) of nectar secreted was measured using calibrated micropipettes. The nectar concentration as well as temperature and humidity were measured using digital refractometer.

Honey sample collection

Samples of coffee honey were obtained from established colonies of beekeepers kept in zander hives from Gera, Yayo and Goma districts during flowering period (January to February, 2019). The honey was

collected at the end of the coffee flowering (March). The samples of honey 500g were collected from each districts for analysis. All samples were kept in sealed glass jars and frozen at -20°C until analysis.

Melisso palynological analysis of honey

To determine the botanical origins, ten grams (10g) of honey sample was weighed in centrifuge tube and added with 20 ml of distilled water (20-40ml) to dissolve the honey. Subsequently, the samples were centrifuged and supernatant solution was decanted following the methods described by (Loveaux, 1978). The extracted samples of pollen grains were spread on a glass slide and then one drop of glycerin jelly was applied to the cover slip and examined under light stereomicroscope (Zeiss 2010). The pollen source plant was identified using reference pollen atlas (Adagaba, 2007) and frequency occurrences of pollen were determined by counting 500 pollen grains from a single slide (Belay *et al.* 2015).

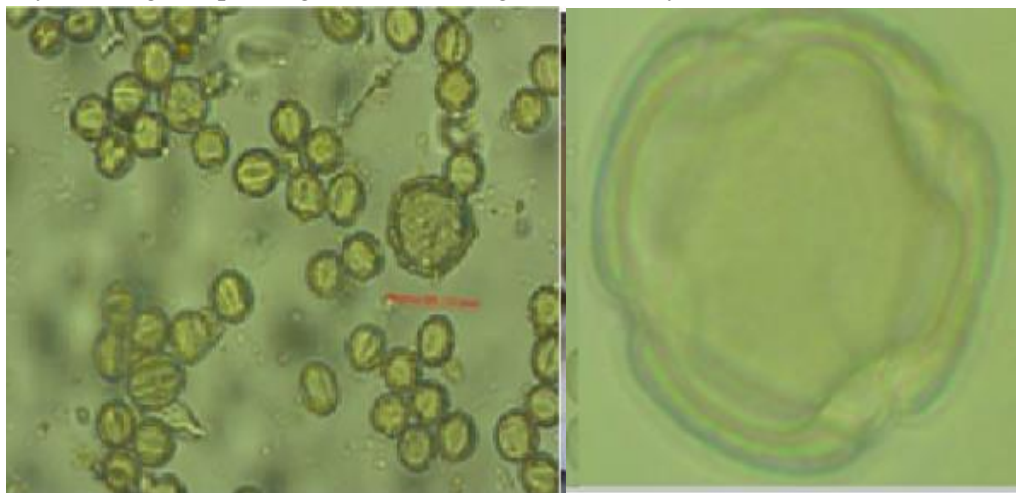


Figure 8. *Coffea Arabica* pollen grain morphology in a coffee honey sample

Physicochemical analysis

The honey quality was determined based on Harmonized methods of the International Honey commission (Bogdanov, 2009). The moisture content of honey samples was determined using Abbé refractometer that can be thermos stated at 20°C, regularly calibrated with distilled. Honey samples was homogenized and placed in water bath until all the sugar crystals are dissolved. After homogenization of the sample, the surface of prism of refractometer was covered with honey and after 2 minutes' refractive index for moisture was determined. Electrical conductivity of honey was determined based on (Bogdanov, 2009). The electrical conductivity of the honey was measured based on the electrical conductance of the sample using conductivity meter. For the determination of the pH of honey, a sample of 10gm of honey was dissolved in 75ml of distilled water in a 250 ml beaker. The solution was stirred using magnetic stirrer and the pH electrodes was immersed in the solution and pH of the honey was recorded. The Hydroxymethyl furfural was determined following the methods (Jeuring and koppers 1980). For the five gram of honey samples was weighed and dissolved into a 50ml of beaker in 25ml of distilled water. The solution was filtered using wattmann paper and the reading was made using HPLC equipped with UV detection.

Determination of mineral content (Ash)

Ash content of honey samples was determined according to the procedures of QSAE (2009). An ash dish was heated in an electric furnace at 600°C and subsequently cooled in a desiccators to room temperature and the dish was weighed (M_2). Five grams of honey sample was weighed to the nearest 0.001g and added to the ash dish (M_0). The dish was placed in the preheated furnace and heated for 1 and a half hours at a temperature of 600°C. The dish with the ash was then cooled in a desiccators and weighed. The ashing procedure was continued until constant weight was reached (M_1). Percent ash (g/100g) honey was calculated using the following formula..

$$WA = (M_3 - M_1) / M_2 * 100$$

Where: M_1 = weight of dish,

M_2 = weight honey taken

M_3 = weight of dish + ash

Determination of sugars by HPLC

The standard substances, for fructose, glucose, sucrose, and maltose were prepared using five level serial dilutions, based on International Honey Commission (IHC) and determination of sugars by HPLC (Bogdanov, 2009) with some modification. The standard was prepared and dissolved in 20 ml of HPLC grade water. About 5g of honey was weighed into beaker and dissolved in 40ml water. A 25ml of methanol was pipetted into 100ml of volumetric flask and the honey solution was transferred to the flask. The methanol mixture of honey solution was filtered using filter paper and solution is poured in vials and stored in standard solution. Peaks were identified on the basis of their retention times of the glucose, fructose and sucrose.

Determination of total Phenol

The phenolic compounds concentration in honey samples were estimated with Folin-Ciocalteu reagent according to the methods as described by (Woldegiorgis *et al.*, 2014). The solution of coffee honey sample was prepared by dissolving 2.5 g honey in 50 ml distilled water and filtered through Whatman no.1 filter paper. One ml of Folin-Ciocalteu phenol reagent was added to the mixture and shaken. After 3 minutes 1ml of saturated sodium carbonate (20%) solution was added to the mixture and adjusted to 10 ml with distilled water. The reaction was kept in dark room for 90 minutes after which the absorbance was read at 725 nm. The total phenolic content of the samples was expressed in milligram per Gallic acid equivalents (GAE). The total phenolic content was calculated as Gallic acid equivalent (GAE) using the calibration equation $Y = 0.0031x + 0.8095$ ($R^2 = 0.99$).

Total flavonoid content

The total flavonoid content of honey was estimated by aluminum chloride ($AlCl_3$); Quercetin was used as the reference which was expressed as QE. The total flavonoid content of each honey samples was determined (Chua *et al.*, 2013). The stock solution was prepared by diluting five grams of honey sample in fifty milliliters of distilled water and filtered through Whatman no.1 paper. Five milliliters from honey stock solution was pipetted and mixed in five milliliters of 2% aluminum chloride ($AlCl_3$) solution. After incubation for ten min, the absorbance of the reaction mixture was measured at 415 nm by using a spectrophotometer (Perkin Elmer Lambda 950 UV/VIS/NIR spectrophotometer). Quercetin (0-200 mg/L) was used as a standard chemical to produce a calibration curve and finally, the total flavonoid

content was reported as the mean value of triplicate assays and expressed as milligram of Quercetin equivalent (QE) per 100 g of honey from the mean value of triplicate data using the calibration equation.

Determination of Antioxidant

The antioxidant capacity of the honey samples was measured by dissolving 1.5 gm of honey with 25 ml distilled water and mixed with 25 ml methanol and placed at 25°C for 60 min maceration using temperature shaker incubator (ZHWHY103B) and then filtered through Whatman No. 4 paper. The residue was then extracted with two additional 25 ml portions of methanol as described above and the combined methanol extracts were evaporated at 40°C to dryness using a rota evaporator (Stuart R3300) and re-dissolved in methanol at the concentration of 50 mg/ml and stored at 4°C for further use. The antioxidant activity of methanol extracts was determined by 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging method as described by (Woldegiorgis *et al.*, 2014). A 0.004% solution of DPPH radical solution in methanol was prepared and then 2 ml of this solution was mixed with 1 ml of various concentrations (0.1–50 mg/ml) of the honey extracts in methanol. Finally, the samples were incubated for 30 min in the dark at room temperature. Scavenging capacity was read spectrophotometrically by monitoring the decrease in absorbance at 517 nm. Ascorbic acid was used as a standard and mixture without extract as the control. The capability of samples to scavenge DPPH was obtained by comparison of sample color reduction effect with the control using the following equation and expressed as percentage values:

$$\text{DPPH radical scavenging activity (\%)} = (A_0 - A_1) / A_0 * 100$$

:

Where A_0 = absorbance of the control

A_1 = absorbance of the sample

Statistical analysis:

Statistical analysis was accomplished on SPSS version 20 for windows and analysis of variance (ANOVA) was performed for significant difference using post hoc test ($p < 0.05$). Correlation among the different parameters was computed by Pearson's correlation coefficient (r) in bivariate linear correlation

Estimation of honey production potential

The honey production potential (HPP) of the plant was estimated by using the following formula: HPP = the average number of flower heads per plant * the average amount of sugar per flower head * nectar secretion length (days). This gives the average amount of sugar per tree/flowering season (Bareke *et al.*, 2020). The average amount of sugar per tree converted to honey was calculated.

At the international market, the average acceptable honey moisture content is 18% from 1 kg of honey whereas 82% is a total dissolved sugar. This is used to convert the mean mass of sugar produced by a single plant per flowering season to honey. The estimation of the number of plants per hectare was based on the recommended space required per this species.

Results and Discussions

Nectar secretion pattern

Nectar volume of *Coffea arabica* was significantly different between the start and end of the secretion date (Figure 1). As the age of the flower increased, the amount of nectar secreted was decreased. Accordingly, the peak nectar secretion was recorded on the day 2 while the lowest was recorded at the end of secretion (day 5). On the 6th day, it was difficult to measure nectar volume since the plant almost stopped nectar secretion. This suggests that *C. arabica* can be considered as a species with short flowering length.

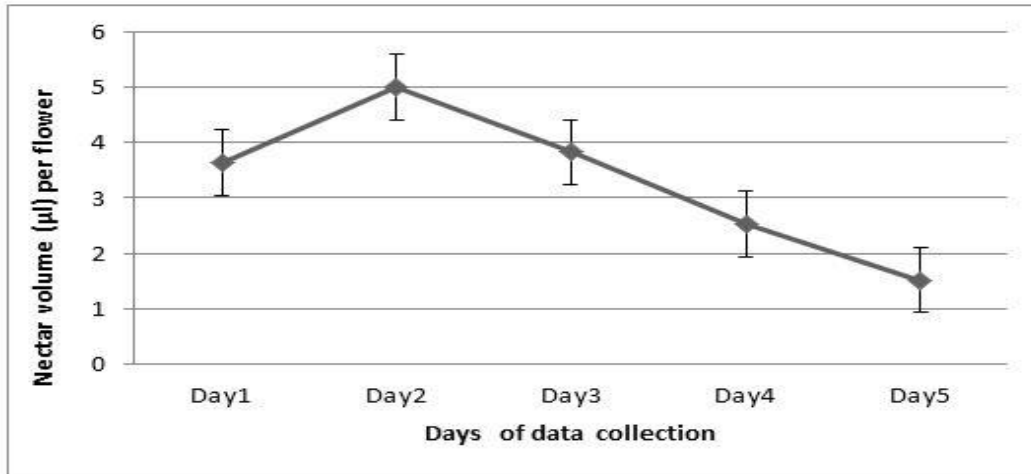


Figure 9. Nectar secretion length and volume of *Coffea arabica* flower from start of secretion to end (repeated collection daily) (N=15 flowers daily from the start to end)

The mean nectar concentration and volume were significantly different across times of the days ($p < 0.05$). The lowest mean nectar concentration was recorded at 7:00 pm and the highest nectar concentration recorded at 16:00 pm. On the other hand, the highest mean of nectar volume was recorded at 7:00 am while the lowest was at 17:00 pm. This is due to high humidity and low temperature in the morning (7:00) and vice versa in the study area. The mean amount of sugar present in nectar was not significantly different ($p < 0.05$) across times of the day (Table 1). This is due to coffee flowers provide nectar from 7:00 to 18:00. Similarly, study conducted by Adgaba *et al.* (2012) on *Ziziphus spina-christi* also indicates that this species provide nectar the whole day. In addition to this, *Lavender* species also secrete nectar the whole day (Adgaba *et al.* 2015). On the other hand, *Croton macrostachyus* secrete nectar from 8:00 to 15:00 (Bareke *et al.* 2020). The significant variations in the amount and patterns of nectar secreted by the different honey source plants could be due to the variations in biotic and abiotic factors associated with the different plant species in their respective environments (Al-Ghamdi *et al.* 2016). This indicates that nectar secretion time is varied from plant species to species. The causes of nectar secretion variability between flowers on the same plant are due to position on the flowering stem to microclimate of the area (Macukanovic *et al.* 2004; Jakobsen and Kristjansson 1994; Petanidou *et al.* 1996). In addition to this, day to day variation in weather may cause shifts in the pattern of nectar characteristics, morphological and phenological characteristics that have effect on nectar secretion.

Table 5. Mean nectar secretion dynamics: Mean nectar concentration (%), nectar volume (μ l) and amount sugar (mg) in nectar per flower in 1hour intervals per flower with \pm standard error (SE) of *C. arabica*

Time (hour)	Average nectar volume (μ l)	Average nectar concentration (%) \pm SE
7:00	6.6 \pm 1.1a	11.1 \pm 2.58d
8:00	4.0 \pm 0.74bc	14.1 \pm 2.01cd
9:00	4.7 \pm 0.71abc	18.6 \pm 2.17abcd
10:00	5.3 \pm 0.41ab	17.2 \pm 2.85bcd
11:00	3.7 \pm 0.7bc	19.9 \pm 1.82abc
12:00	3 \pm 0.7bc	18.8 \pm 1.84abcd
13:00	3.0 \pm 0.33bc	20.7 \pm 3.33abc
14:00	3.1 \pm 0.85bc	21.7 \pm 3.75abc
15:00	2.5 \pm 0.88c	24.3 \pm 1.71ab
16:00	3.2 \pm 0.7bc	25.9 \pm 1.64a
17:00	2.7 \pm 0.54c	23.4 \pm 2.14ab
18:00	3.2 \pm 0.96bc	25.4 \pm 2.69a

Note: Different letters show significant differences

Melissopalynological analysis of honey

Microscopic pollen analyses of honey samples indicated that all samples collected from four districts (Gera, Gomma, Yayu and Manna) were monofloral, since *Coffea Arabica* honey pollen count constituting 84%, 93%, 75% and 73% respectively (Figure 4). During honey pollen analysis, if the honey is considered to be monofloral and its pollen frequency in honey pollen sediments should constitute more than 45% or more in pollen count (Can *et al.*, 2015). The secondary pollens contributing for coffee honey from study districts were *Aspilia africana* (8.6%), *Bersama abyssinica* (8.86%), *Rumex nervosus* (21%), *Rubus studneri* (12.7%), *Vernonia amygdalina* (9.7%), The minor pollen source plants *Ceasalpina decaptella* (3%), *Eucalyptus spp* (3.8%) and *Hypoestes forskalii* (0.3%).

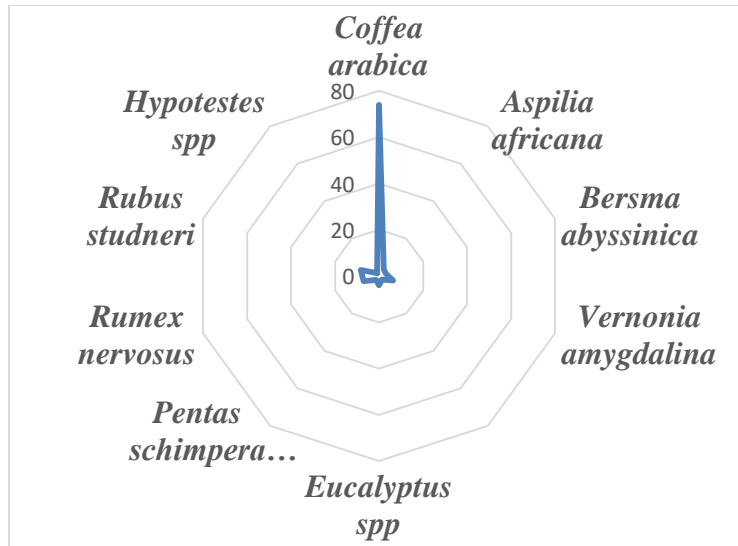


Figure 10. Contributing bee forages to *Coffea arabica* monofloral honeys in Gomma, Gera, Yayu and Manna districts

Physiochemical properties of honey

The results of the physico-chemical analysis of the mono floral honeys of *Coffea arabica* for different parameters were indicated and discussed in Table 2.

Moisture content

The moisture content of the Coffee honey samples of study area is ranging from 21. to 24.05 % with mean of 22.46 %. There is statistical significance difference ($P < 0.05$) of MC between the study districts. The moisture content of honey samples from Gera and Manna are significantly different from Gomma and Yayu (Table 2). The honey samples from *C. arabica* were higher than the country's average 20.6% reported by Nuru Adgaba *et al.*, (1999), Chala *et al.*, 2012 and Tewodros, (2010). The higher moisture content of honey in Gera and Manna is due to the prevailing atmospheric humidity and pre- and post-harvest management of honey of the area. Moreover, Gera and Manna districts are dominated by moist forest which might have resulted for increased moisture content of the honey in the area.

Ash

Ash content is one of a quality criterion for botanical and geographical origin of honey. In this result coffee honey samples showed the lowest ash content, ranging from 0.15 to 0.23/100g with mean value of 0.21 ± 0.3 g/100g). The ash content of honey samples significantly varies between honey sample ($p < 0.05$). The Ash content of the honey samples from Manna and Gomma is significantly different ($P < 0.05$) from Gera and Yayu. The variation of ash content of coffee honey might be due to the variability of soil types and concentration of minerals found in the nectar of the flowers at different location. Ash content of all samples was within the range (0.1 to 0.5 g/100 g) accepted by codex range (Codex, 2001).

Hydroxymethylfurfural (HMF)

The amount of hydroxymethyl furfural (HMF) in honey is one of the major important indicators of honey quality in that it indicates whether it is aged or overheated Marchini *et al.* (2007). In fresh honey, HMF is present only in small amounts and its concentration increases with storage time and prolonged heating of honey. The HMF values in the current study were found to be between 6.67 to 16.74 mg/ kg. There was no significant difference in HMF content of coffee honey ($p > 0.05$) between districts. The HMF of the *C. arabica* honey was relatively low as compared to National and international standards set by (Codex Alimentarius, 2001) indicating the freshness of honey. In Ethiopia, the acceptable HMF level is below 40 mg/kg honey and the HMF value of this study area was less than 40 mg/kg (Nuru, 1999). This is agreeing with (Chala *et al.*, 2012) in Gera district of Oromia region.

Free Acidity

Acidity is an essential quality parameter measure, for its antimicrobial property. Free acidity values indicate the fermentation of honey sugar by yeasts. During fermentation, glucose and fructose are converted into carbon dioxide and alcohol. Alcohol is further hydrolyzed in the presence of oxygen and converted into acetic acid. The acidity of tested honey samples varied from 32.96 to 38.90 mg/100 with the mean of 35.89 mg/100g of honey. There was no significant variation ($P > 0.05$) in free acidity among the honey samples from the four districts. Variation in free acidity among coffee honey samples could be due to the different floral origins or to the variation in harvesting seasons (Perez-Arquillue *et al.*, 1994).

pH

Honey pH has great importance during storage of honey, as they influence the texture, stability and shelf life of honey (Terrab *et al.*, 2004). There was no significant difference ($p > 0.05$) in pH of honey between honey samples obtained from four districts. pH of coffee honey samples in the current study ranged from 3.31 to 3.48 with an average value of (3.41 ± 0.09) . The mean value (3.4) of honey of the study area is in line within the report of Bogdanov (1997) who indicated that honey pH should be between 3.2 and 4.5 that ensures honey samples' freshness. The low pH of honey inhibits the presence and growth of microorganisms.

Electric Conductivity

The current results showed that the average electric conductivity of Coffee honey (0.44 mS.cm^{-1}) to 0.58 and with mean value of 0.49 ± 0.08 and there was significant difference between honey samples between the districts ($p < 0.05$). The honey sample from Gera district is significantly varied from three districts. The variation between honeys for EC value in four districts is due to existences of different flora species in area.

Sugar profile

The fructose content of coffee honey ranges from 31.46 ± 5.6 to 35.31 ± 6.08 with mean value of 32.77 ± 6.06 . The amount of sucrose in the honey samples obtained from different districts did not show significant differences ($p > 0.05$). The glucose content of coffee honey ranges 31.09 to 32.8 with mean value of 31.94. The fructose content of coffee honey is within range of National and International ranges nearly close to reports of Nuru *et al.* (2017). The mean, standard deviation and the range of the sucrose content of the coffee honey 2.72 to 4.75 with mean value of 3.75 ± 1.52 . The sucrose content of honey

between honey samples was significantly different ($p < 0.05$). The mean sucrose content of the coffee honey is less than the national average of 3.6%, which was reported by Nuru (1999) and lower than the maximum limits 10% set by QSAE (2005) and 5% set by IHC (2002). On the other hand, the maltose content of honey range from 0.27 to 0.8 with mean value of 0.41 ± 0.39 . The Maltose content between honey samples was significantly different ($p < 0.05$). The maltose content of honey fromYayu districts significantly vary from the rest of the honey.

Table 6. Physiochemical properties of Coffea arabica honey from different districts

Sample locality	MC	ASH	EC	FA	pH	HMF	Fructose	Glucose	maltose	Sucrose
Yayu	21±0.9b	0.2±0.07ab	0.5±0.1ab	35.9±66a	3.±0.18 a	17±8a	35±6.08a	33±2.31a	0.8±0.8a	3.6±1.3ab
Gommasa	22±0.3b	0.30±0.15a	0.63±0.21a	37.8±9a	3 ±0.1a	11±7ab	32±6.2a	32±2a	0.27±0.21 b	3±1.2b
Manna	23±0.1a	0.26±0.09a	0.59±0.16a	37±8a	3±0.12a	12±18ab	33±6 a	32±3a	0.28±0.11 b	2.72±1.3b
Gera	24.±1a	0.08±0.009 b	0.28±0.012 b	31.±4 a	3.±0.4a	6±2b	31±6a	31±2a	0.32±0.01 c	4.75±0.97 a
Mean	22.4	0.21	0.49	18	3	12	33	31.9	0.41	4

Correlation

The Pearson correlation coefficients between all parameters are presented in Table 3. There were significant and strong correlations between Ash content and electrical conductivity $r = 0.99$ ($p < 0.01$).The high correlation coefficient of the ash content and electrical conductivity indicates the possible influence of mineral component of honey on its electrical conductivity. The measurement of electrical conductivity depends on the ash and acid contents of the honey. The higher the ash and acid content, the higher the resulting conductivity. The correlation between electrical conductivity and total ash content found in this work is similar to the findings of Accorti *et al* (1987) and Belay et al 2015. There was also correlation between Free acidity and sugars in honey due to production of alcohol transforming in to organic acids from fermentation of sugar in honey.

Table 7. Pearson correlation coefficients among the analyzed parameters

	MC	Ash	EC	HMF	PH	FA	Fr	Glu	Sucr	maltose
MC	1									
Ash	-0.17 ^{ns}	1								
EC	-0.157	0.99**	1							
HMF	-0.197 ^{ns}	0.16 ^{ns}	-0.17	1						
PH	-0.16 ^{ns}	-0.104 ^{ns}	-0.065	0.22 ^{ns}	1					
FA	-0.07 ^{ns}	0.20	-0.182	0.021 ^{ns}	-0.054 ^{ns}	1				
Fr	-0.31 ^{ns}	-0.094	-0.09	-0.105 ^{ns}	0.10 ^{ns}	0.46*	1			
Glu	-0.34 ^{ns}	-0.38 ^{ns}	-0.36*	0.009 ^{ns}	-0.056	-0.09 ^{ns}	0.198 ^{ns}	1		
Sucr	0.01 ^{ns}	0.003 ^{ns}	-0.19	-0.22 ^{ns}	0.047 ^{ns}	0.036 ^{ns}	-0.039 ^{ns}	-0.062 ^{ns}	1	
Malt	-0.43 ^{ns}	0.026 ^{ns}	0.001	-0.23 ^{ns}	0.23 ^{ns}	0.099 ^{ns}	0.241	0.022 ^{ns}	-0.087	1

*, **, ns = significant at 5 and 1%, and non-significant at 5%, respectively. MC=moisture content, Ash=Ash content, HMF=hydroxyl methyl furfural aldehyde, pH=pH of honey, FA=Free Acidity, Fr=Fructose, Glu=Glucose, Malt=Maltose, Su =Sucrose.

The total content of Phenol and Flavonoid

There was significant difference in total phenol content of coffee honey in different districts. Total phenol content of honey samples from Manna is significantly different from Gera, Gomma and Yuyu. Total phenol in coffee honey ranged from 42.5±0.94 mg/100g to 82.1±6.48, The values were highest for Manna (82.1±6.48) and lowest for Gomma (69.3±3.8 mg/100g). On the other hand, total flavonoids in coffee honey ranges 21.57±0.90 to 53.9±4.4 and highest for Yuyu (42.5±0.94 mg/100g) and lowest for Mana (21.57±0.90mg/100g). There was strong significance difference (P < 0.05) between flavonoid content of Manna and Yuyu districts. The variation in the amount and type of polyphenolic and Flavonoids in honey is due to floral origin (Kucuk *et al.*, 2007).

The antioxidant content of honey

The DPPH free radical has been widely used as a tool to determine the free radical-scavenging activity in honey samples. Antioxidant content of coffee honey ranged from 57.9±5.86 to 6.4±0.32% inhibition (Table 4). There was no significant difference (P>0.05) between honey samples and the highest antioxidant level was found in Manna and Gera 66.3±4.84 and 66.4±0.32% and lowest for Yuyu and Gomma (60.9±6.9 and 60.2±2.27), respectively. The, higher correlations were observed between the DPPH radical scavenging activity and the total polyphenol (r = -0.755, p < 0.001), and total flavonoids (r = 0.167, p < 0.01), and between total flavonoids and total polyphenols (r = 0.81, p < 0.01). These results are in agreement with Alvarez-Suarez *et al.* (2010), Sant'Ana *et al.* (2012), and Ferreira *et al.* (2009) who found that there is a positive correlation between DPPH radical scavenging activity and total polyphenols, and total flavonoids.

Table 8. Total phenol and antioxidant content of coffee honey samples obtained from four districts from western Oromia

Honey sample location	TPC mg/100ml	T FC mg/100ml	Percentage of inhibition
Yayu	71.6 ±4.2ab	53.9±4.4a	60.9± 6.9a
Gomma	69.3±3.8b	35.9±0.99b	60.2±2.27a
Mana	82.1±6.4a	21.57±0.90c	66.3±4.84a
Gera	42.5±0.94c	33.05±5.05b	66.4±0.32a

Honey production potential

The average number of *C. arabica* flowers per plant was 1044 (Table 5). Each flower was observed to provide nectar for at least 5 days. The mean amount of sugar per flower was 3.6 ± 0.3 mg (ranges from 0.8 to 8.72 mg), and therefore, the average mass of sugar produced per plant is estimated to be 0.04 kg (range from 0.008-0.09 kg).

Table 9. Mean number flower heads/plant (N=18 plant), mean Nectar volume in 24 hours (μ l) (N=100 flowers) and mean sugar amount per flower life cycle (mg) (N=100 flowers) of *Coffea arabica* in Gera District

Treatments	Mean \pm SE	Minimum	Maximum
Mean number flower heads/plant	1044 \pm 135	485	2330
Mean Nectar volume in 24 hours (μ l)	3.3 \pm 0.20	0.70	7.30
Mean sugar amount per flower life cycle (mg)	3.6 \pm 0.30	0.80	8.72
Mean sugar amount per plant (kg)	0.04 + 0.003	0.008	0.09

Given that 1 kg of honey with 18% moisture content (wt/wt) contains 820 g of total dissolved sugar, the mean mass of sugar produced by a single tree of *C. arabica* (0.04 kg) per season is estimated to produce 0.05 kg of honey (range 0.01 – 0.11 kg). Space between coffee plants was 2 meters and therefore, the total number of Coffee plants per hectare of land is about 2500. Therefore, under ideal conditions, the average honey production potential per hectare of coffee plantation area per flowering season would be about 125 kg, which ranges from 25 to 275 kg. The actual amount of honey that can be harvested from the hive is half of the estimated potential which is 62.5 kg per hectare.

The average mass of sugar per coffee plant (0.04kg) is lower than the average amount of sugar of *Croton macrostachyus* per plant (Bareke *et al.*, 2020). Floral nectar is a reward offered by flowering plants to visiting pollinators, reflecting co-evolution between the plants and their pollinators (Ning-Na *et al.*, 2015; Power *et al.*, 2018). The common nectar variables relevant to pollination are its concentration, volume and sugar. Nectar is not part of the plant's sexual system but a reward offered to a foraging agent. Nectar collection method is primarily dictated by the flower size, nectar volume and solute concentration (Dafni, 1992). The common method is to extract the nectar with micropipettes for volumes above 0.5 μ l and concentration below 70%.

The actual amount of honey that can be harvested from the hive is half of the estimated potential of the plant. When bees collect and transport the nectar to the hives they definitely consume a certain amount of sugar for their flight energy. In addition to this, due to rapid crystallization, all the nectar secreted may not be available to honeybees (Adgaba *et al.*, 2012; Bareke *et al.*, 2020). The estimated honey production potential that can be obtained from *C. arabica* plantation per hectare was 125 kg. These results are comparable to the reports made for different annual plants and trees such as *C. macrostachyus* in range of 234 kg - 1770kg /hectare(Bareke *et al.*, 2020), Lime species (*Tilia* spp.) (90 to 1200 kg honey/ha) (Crane *et al.*, 1984), and *Ziziphus spina-christi* (550-1300 kg of honey/ha) (Adgaba *et al.*, 2012), *Brassica juncea* and *Sinapis alba* crops 65.5 kg and 71.2 kg/hectare, respectively (Masierowska, 2003). Monofloral honey of *C. arabica* is produced in some parts of Ethiopia. For example, from western Ethiopia it is produced in Gera District (Bareke and Addi, 2019). Since the flowering period of *C. arabica* is short, the beekeepers should apply seasonal colony management following flowering calendar to harvest the monofloral honey of this plant.

In general, trees were more productive in nectar secretion due to their larger biomass, dense flowers, deep roots and resistance to moisture stress (Adgaba *et al.*, 2017). Furthermore, in most trees, the flowers are not colorful and are expected to secrete more nectar to strongly attract sufficient pollinators (Schemske and Bradshaw, 1999). However, *C. arabica* has white color which can attract honeybees and other insect pollinators.

Conclusion and Recommendation

Coffea arabica is a good producer of nectar and significantly contributes to monofloral honey production since its pollen count is greater than 45% in most honey samples. The Melissopalynological analysis of honey indicated that the existence of different floral species contributing for *Coffea arabica* honey. From all identified honey plants *Aspilia africana*, *Vernonia amgdalina*, *Bersama abyssinica*, *Rumex nervosus*, *Justicia schimperna* and *Pentas schimperana* are the major contributing bee forages in the area. The physicochemical properties of *Coffea arabica* honey meets the basic honey quality standards both at the national and international honey quality specifications. Therefore, beekeepers should focus on production of coffee monofloral honey to exploit the niche market opportunities such as organic honey, promotion and commercialization mono-floral honeys from *Coffea arabica*.

Nectar secretion analysis indicated that the amount of nectar volume and concentration varied in different times of the day. One hectare of *C. arabica* plants has a potential to produce 125 kg of honey of which 62.5 kg is expected to be harvestable. The current study clearly indicates that coffee is not only economically valuable for its seeds, but also used for honey production. Therefore, integration of coffee orchard with beekeeping is recommended to produce honey, as well as to boost the seed yield of coffee. In order to achieve this, the producers should focus on appropriate management honey bee colony following the flowering period coffee plant since coffee has short flowering period.

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The impact of feeder type on the honeybee colonies (*apis mellifera* L.) And hive operation during colony feeding

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Abstract

Several types of feeders have been used by beekeepers for feeding their colonies with sugar syrup during dearth periods. However, each feeder type has its benefits and drawbacks both for the bees and the beekeepers. The effects of different feeder types on honeybee colonies well-being and their conveniences for feeding by the beekeepers were investigated. Time required to feed a colony, amount of feed consumed, number of dead bees during feeding, number of dead bees in/on the feeder when removed, disturbance and convenience based on technicians' opinion were compared. Accordingly, significantly ($p < 0.000$) shortest (40.45 sec) time of feeding was obtained for top feeder than the bucket (71.25 sec) and frame (137.80 sec) feeders. Likewise, significantly ($p < 0.001$) less number of dead bees (2.50) were observed while feeding a colonies using top feeder as compared to using bucket and frame feeders in which 5.45 and 11.00 dead bees were recorded under hive stand, respectively. Moreover, no dead bee was recorded on the top surface of top feeder compared to frame feeder in which 1.60 dead bees were counted. Feeder type did not affect the amount of sugar syrup consumed and colony survival during the experiment. Regarding bee technicians' opinion, top feeder is highly convenient to feed colonies with a minimum colony disturbance and reaction. Thus, from the current study, top feeder is recommended as it required less time for feeding, inflicted less damage to the bees, and found to be more convenient feeding method.

Keywords: bucket feeder, colony feeding, frame feeder, sugar syrup, top feeder

INTRODUCTION

In Ethiopia traditional beekeeping has been practiced following the flowering period which provide sufficient quantities of nectar and pollen to stimulate colonies build up and maintain optimum colony populations. However, nowadays, maintaining optimum colony population during dearth period with limited floral resources is becoming a major problem for the beekeepers. Shortage of floral resource availability leads to declining of colony's population eventually resulting in weaker colonies. Such colonies are also vulnerable to absconding, natural pests and predators of honeybees (Kibebew et al., unpubl. data). The first option to minimize dearth period colony starvation is providing supplementary feeds (Neupane and Thapa, 2005). Providing sugar syrup as supplementary feeding to honeybee colonies can be one of the management option during dearth period (Somerville, 2014). Sugar syrup feeding is popular and the most effective during dearth periods to maintain optimum colony population (Somerville, 2014) that ensures early colony build up to produce surplus honey during honey flow season (Sihag and Gupta, 2013). To this end, different types of feeders and feeding methods have been invented, tested and used for feeding honeybee colonies of different races with varying behaviors. However, the types of feeder

used depends on materials available at the beekeepers' level and possibly on the types of commercial feeders on the local markets. Thus, the primary choice of feeder type is the primary management decision to be made by the majority of beekeepers.

In Ethiopia, several types of feederlike top feeder, bucket feeder and frame feeder have been used to feed honeybee colonies, during dearth periods. However, the effects of these different feeder types on the well-being of honeybee colonies, time consumption during feeding and convenience in terms of hive operation and reaction of bees have not been evaluated under local honeybee colonies. Therefore, the aim of this study is to evaluate whether feeder types (top feeder, bucket feeder and frame feeder) affect time required to feed a colony, amount of feed consumed, cause death of bees during hive operation for feeding, and removing feeders, colony disturbance and absconding rate in relation to our local honeybee behavior. In addition to these, this study also aimed to collect and analyze bee technicians' opinion (preference) on the convenience of each feeding methods.

MATERIALS AND METHODS

Description of the study area

The study was conducted at two experimental apiaries (Holeta: 9° 30' N and 38° 30' E, Elevation 2450 m and 30 kilometers west of Addis Ababa; Bako: 9° 6' N and 37° 9' E, Elevation 1650 m and 258 kilometers west of Addis Ababa) of Holeta Bee Research Center in Oromia Regional State of Ethiopia.

Preparation of the feeders

A total of 10 bucket feeders, 10 plastic frame feeders and 10 top feeders were used for the experiment. Bucket feeders with volume of 2 L were purchased from local market. Plastic frame feeders that fit with the dimensions of the hives were obtained from bee equipment importer. Top feeder, an inner cover made up of plywood with the dimensions to perfectly fit with wooden rims dimensions, similar to the outer hive chambers were constructed at Holeta Bee Research Center's workshop (Fig. 1). The 2.5 cm high wooden rims (edges) on the outside provide sufficient room for supplementary feeding (pollen or sugar syrup) to bees on the top surface. The inner cover has a rectangular open space of about 5 cm x 8 cm at its center with a tight ledge of 1.5 cm high and 3 cm wide erected to facilitate provision of supplementary feeds. The open space allows bees' access to the space between the inner cover and the hive lid during feeding.

Establishing experimental colonies

For this study, well-established local honeybee (*Apis mellifera*) colonies were used at two experimental apiaries of Holeta Bee Research Center (Holeta and Bako). The colonies were kept in standard Zander hives with one additional box (super) and each box containing 10 frames. During flowering season, in last week of October 2018, colonies were subjectively estimated for their strength based on number of frames covered by adult bees, brood, nectar and honey, and pollen, as described elsewhere (Delaplane et al., 2013). Then, 15 honeybee colonies with approximately uniform strength were selected and randomly assigned to the three treatment groups (bucket feeder, frame feeder and top feeder) with five replications at each apiary.

Evaluation of feeder types

The feeding experiment was started in January 2019 and conducted during identified dearth periods using an objective mode which uses empirical measures. A subjective mode that relies on visual estimates by one or more observers was also employed to evaluate parameters like disturbance and convenience to a beekeeper based on the observers' opinion.

Bucket feeder:

Sugar syrup was prepared in 1:1 (sugar to water) ratio. One and half liter of the syrup was poured into each bucket feeder and some grass was placed on the top of the syrup to protect bees from drowning down while taking up the feed. A 1 m x 1 m plastic sheet was placed under each hive stand to collect data on the number of dead bees on the next morning after feeding. The hives were opened and at least four existing frames with empty combs were removed and bucket feeder with 1.5 L of sugar syrup was placed into the hive and the hive was closed. After three days, the colonies were checked for the dead bees and the feeders were removed. Time taken to pour the syrup, open the hive, remove the frames, close the hive and transport the frames taken away from the hives and remove the feeder was recorded at each step. The time recorded for hive opening and frame removal did not include the preparation, transportation and addition of supers (additional boxes) when needed. Similarly, time for feeder removal did not include removal of the supers (additional boxes) and their transportation, which required one additional working day. Moreover, number of dead bees under each hive stand, amount of feed over left, number of colony absconded (if any), number of dead bees in/on the feeder at a time of feeder removal, level of disturbance and bee technicians' opinion while feeding and feeder removal were recorded.

Frame feeder:

Plastic frame feeder with the capacity of 2L was used. Like in bucket feeder, sugar syrup was prepared in 1:1 (sugar to water) ratio. Unlike in bucket feeder, the hive was opened and one frame with empty comb was removed from the hive to place the frame feeder into one side of the hive and 1.5 L sugar syrup was poured into the frame feeder after placing the feeder in the hive. All other procedures, data recording and observations were made as described for bucket feeder.

Top feeder:

Here, an inner cover made up of plywood with wooden rims that serves both as inner cover and top feeder was used. Sugar syrup was prepared as describe under bucket feeder. The prepared syrup was taken closer to the hive. The lid of the hive was opened and 1.5 L sugar syrup was poured on the top surface of the inner cover/top feeder (Fig. 2). The hive was finally closed with the outer lid. Unlike the two feeders, there is no need to remove frames/add extra hive body (super) for the feeder and also no need of feeder removing at all. Otherwise, all other procedures, data recording and observations were made as for bucket feeder described above. The conveniences of the feeder types for the beekeeper in relation to bees' reaction were observed. For this purpose, five experienced bee technicians were allowed to run the feeding experiment every time and a technician given a chance to feed a colony using at least one feeder type. Their response on the convenience of each feeder type was recorded as less convenient, medium and highly convenient.



Fig.1. Top view of the inner cover/top feeder equipped with its wooden rims (Photo by ZewduArarsoHora).

Statistical analysis

Student's T test was used to compare effects of location on time required to feed a colony, and number of dead bees under hive stand and in/on a feeder a day after feeding bees with sugar syrup and at a time of feeder removal. The effect of feeder type on time required to feed a colony, number of dead bees under hive stand and in/on a feeder at a time of feeder removal were analyzed using ANOVA procedures of IBM SPSS Statistics version 20 (IBM Corp. 2017). Tukey Honest Significant Difference Test procedures were used to test for significant differences among the treatments at 95% confidence interval and $\alpha = 0.05$ level of significance. Moreover, crosstabs descriptive procedure was employed to determine if feeder type affects the convenience of honeybee colony feeding and Pearson's chi-square was used to test the relationship between the two variables.

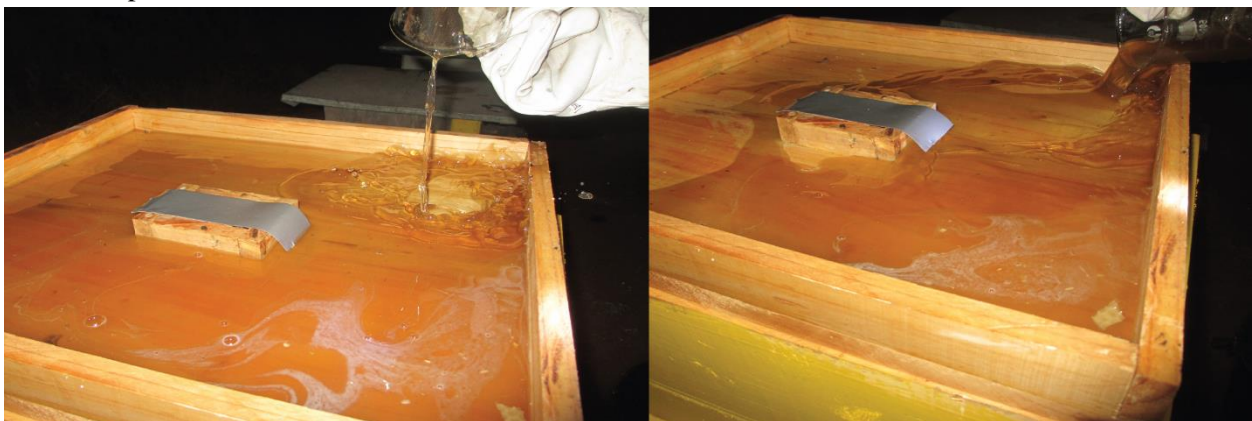


Fig. 2. Picture shows the process of sugar syrup pouring on the top surface of an inner cover (top feeder) to feed a colony (Photos by ZewduArarsoHora).

RESULTS

The effect of apiary location and feeder type on time required to feed a colony and number of dead bees

The average time required to feed a colony regardless of feeder type was 83.17 ± 6.17 sec with a range of 31.00 – 180.00 sec ($n = 60$). The average time required to feed a colony was not significantly varied between apiaries ($p > 0.65$) (Table 1). Similar results were obtained when comparing number of dead bees under the hive stand a day after feeding and at a time of feeder removal ($p > 0.12$ and $p > 1$, respectively). However, the results were significantly varied within each apiary. At Holeta, comparison among feeder types on time for placing the feeds and removing the feeder in each colony showed that significantly longest time was recorded for frame feeder, followed by bucket feeder, and the shortest time for the top feeder ($F = 19.62$, $df = (2,27)$, $p < 0.000$). At Bako sub-site, similar trend ($F = 185.38$, $df = (2,27)$, $p < 0.000$) (Table 2). As for the duration of hive operation, the number of dead worker bees was significantly higher when using frame feeder as compared with using top feeder and bucket feeder at both locations (Table 2).

Table 10. Mean \pm standard error of time required to feed a colony (sec), and number of dead bees under hive stand and in/on a feeder a day after feeding bees and at a time of feeder removal, across apiary sites

Variables	Locations	
	Holeta	Bako
Time require to feed a colony	80.33 ± 7.30	86.00 ± 10.04
Number of dead bees under hive stand	4.77 ± 0.98	7.87 ± 1.69
Number of dead bees in/on a feeder at a time of feeder removing	0.60 ± 0.16	0.60 ± 0.16

Analyses of time required to feed a honeybee colony, and number of dead bees under hive stand and in/on a feeder a day after feeding bees and at a time of feeder removal, were performed based on feeder types pooling the data from both apiaries. The time elapsed to feed a colony using different feeders was significantly different among feeder types ($p < 0.000$). The longest duration was recorded for frame feeder (137.80 ± 8.98 sec), while the shortest was for top feeder (40.45 ± 1.39 sec). Furthermore, the higher significant number of dead worker bees was counted while feeding colonies using frame feeder compared to top feeder and bucket feeder (Table 2 and 3).

Table 2. Effects of different feeder types on time required to feed a colony (sec), and number of dead bees under hive stand and in/on a feeder a day after feeding bees and at a time of feeder removal, when each apiary is analyzed separately. Values are mean \pm standard error of treatments

Feeder type	TRF		NDBUH		NDBIOF	
	Holeta	Bako	Holeta	Bako	Holeta	Bako
Top feeder	42.90 ± 1.86^C	38.00 ± 1.84^C	1.60 ± 0.58^B	3.40 ± 0.78^B	0.00 ± 0.00^B	0.00 ± 0.00^B
Bucket feeder	81.10 ± 2.46^B	61.40 ± 4.32^B	4.90 ± 0.89^B	6.00 ± 1.50^B	0.20 ± 0.13^B	0.20 ± 0.13^B
Frame feeder	117.00 ± 14.16^A	158.60 ± 6.64^A	7.80 ± 2.43^A	14.20 ± 4.22^A	1.60 ± 0.27^A	1.60 ± 0.27^A
p value	0.000	0.000	0.028	0.019	0.000	0.000

TRF = Time required to feed a colony, NDBUH = Number of dead bees under hive stand, NDBIOF = Number of dead bees in/on a feeder at a time of feeder removal

A, B, C = Within a column, means followed by different upper case superscript letters indicate significant differences among treatments

The effects of feeder type on amount of feed consumed, colony absconding, disturbance and convenience

To compare the effects of feeder types on the colonies feed consumption, an observation was made on the third day for the feed over left. Accordingly, there was no sugar syrup left in any of the feeder. Moreover, there was no absconding event occurred during the experiment. However, using top feeder allowed sugar syrup feeding with minimal disturbance to the colony when visually observed during the feeding processes (Fig. 3). Furthermore, the top feeder was also found highly convenient ($\chi^2 = 81.60$, $df= 4$, $p<0.000$) compared to bucket and frame feeders when subjectively evaluated by bee technicians involved in the feeding operation.



Fig. 3. Pictures show feeding of honeybee colonies using different types of feeder and the extent of colony disturbances (Photos by ZewduArarsoHora).

Table 3. Effects of three feeder types on time required to feed a colony (sec), and number of dead bees under hive stand and in/ona feeder a day after feeding bees and at a time of feeder removal, pooled data from both locations. Values are mean \pm standard error of feeder types.

Feeder type	TRF	NDBUH	NDBIOF
Top feeder	40.45 \pm 1.39 ^C	2.50 \pm 0.52 ^B	0.00 \pm 0.00 ^B
Bucket feeder	71.25 \pm 3.31 ^B	5.45 \pm 0.85 ^B	0.20 \pm 0.92 ^B
Frame feeder	137.80 \pm 8.98 ^A	11.00 \pm 2.48 ^A	1.60 \pm 0.18 ^A
<i>P</i> value	0.000	0.001	0.000

TRF =Time required to feed a colony, NDBUH = Number of dead bees under hive stand, NDBIOF = Number of dead bees in/ona feeder at a time of feeder removal

A,B,C= Within a column, means followed by different upper superscript letters indicate significant differences among feeder types

DISCUSSION

In different parts of the world many different methods and types of feeder have been used by beekeepers for feeding sugar syrup to honeybee colonies. Using each feeder type has its advantages and disadvantages (MacFawn, 2019). It is monotonous for the beekeeper having to open and operate each hive while feeding colonies internally, which is a time consuming and an extra work if there are many colonies (Abou-shaara, 2016). In this result, this is reflected by varying time of hive operation using three different feeder types. Colony feeding using top feeder relatively required less time as compared to using the bucket and frame feeders. This is consistent with the previously reported time used to feed a colony (Liseki, 1996). The shortest time to feed a colony may associate with using inner cover as top feeder, which did not require the removal of the existing frame or addition of extra super for a feeder as for frame and bucket feeders. In addition, when inner cover is used as a feeder, it makes opening of outer cover (hive lid) much easier than removing a propolized hive lid used without inner cover. Even removing a propolized inner cover is much easier because a hive tool can get easily in between the cover and the hive body than trying to open a propolized hive lid. As the feeder (inner cover) size is just right to fit to the hive body leaving no gap for bees and the time used to open the hive is so short, no bees would come out to sting and also bees could not fly out (see Fig. 3). Moreover, once top feeder is installed in position, it is easy to refill the syrup at any time. Thus, top feeder allows colony feeding with a minimum disturbance to the colony. Furthermore, such conditions give a chance for the beekeepers to feed their bees even without wearing beekeeping clothing (Liseki, 1996).

In general, Ethiopian honeybees are defensive, and defending the honeybee colony and its resources is crucial for maintaining the colony integrity (Andere et al., 2002; Patrice et al., 2018). The defensive behavior makes colony management difficult (Patrice et al., 2018), as hive operation can create a provocative situation to the colony (Alemu et al., 2014; Patrice et al., 2018). To this end, taking out a frame from a hive and inserting a frame feeder and bucket in the place is thought to provoke the bees to defend in mass (Blackiston, 2009), which might result in a significantly higher number of dead bees. This together with its demanding of long time hive operation suggests that using frame and bucket feeders to feed local honeybee colonies is less useful. Although a comparative comparison of the cost and benefits over several years of use by a diversity of beekeepers (with varying levels of experience) to fully assess the longevity of the equipment and productivity under beekeepers' condition is required for the future, top feeder is the best type from its ease of colony management point of view.

CONCLUSION

This study took the first step in evaluating the effects of feeder types on local honeybee behavior and convenience of the feeders to the beekeepers. The results suggested that top feeder is likely to have more place for Ethiopian beekeepers for its less time consuming, inflicting less damage to the bees, and being more convenient. Based on this finding, top feeder is recommended as a suitable feeder for local honeybees and beekeepers.

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Feed Resources and Rangeland Management Research Results

Adaptation Trial *Sesbania sesban* accessions in Highlands of East Hararghe Zone

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Abstract

*The experiment was conducted to evaluate the yield and adaptability of five accessions with one local check of *Sesbania sesban* in 2017/18 and 2018/19 G.C consecutive years at FTC of Burka Jalala kebele, Meta district of Eastern Hararghe Zone. The tested *Sesbania sesban* accessions were *S.sesban* 15019, *S.sesban* 10865, *S.sesban* 15036, *S.sesban* 10885, *S.sesban* 1238 and local check. The treatments were arranged in a Randomized Complete Block Design (RCBD) with three replications. The analysis result showed that there was significant ($P < 0.05$) variation among the accession in fresh leaf weight, fresh stem weight and leaf to stem ratio. The maximum fresh leaf biomass yield (7.91 ton ha⁻¹) was obtained from the accession *S.sesban* 1238, followed by accessions *S.sesban* 10885 (7.23 ton ha⁻¹). The highest leaf to stem ratio were recorded from accession *S.sesban* 10865(28.67%), followed by *S.sesban* 15019(27.66%) and *S.sesban* 1238(31.33%). The analysis of chemical composition indicated that significantly ($p < 0.05$) the maximum acid detergent lignin was obtained from *S.sesban* 15036(22.746%), and the minimum acid detergent lignin obtained from *S.sesban* 1238(14.875%) and *S.sesban* 10885(14.874%). Hence, accessions *S. sesban* 1238 and *S.sesban* 10885 were found to be the best promising to be demonstrated under the study area and similar agro-climatic conditions.*

Keywords: Forage Trees, quality forage, *Sesbania Sesban*, tree legumes

Introduction

Livestock and especially ruminants are an essential component of most of the agricultural production systems in sub-Saharan Africa. Livestock is an integral component for most of the agricultural activities in Ethiopia. The livestock sector has a share of 12-16% of the total Gross Domestic Product (GDP), and 30-35% of agricultural GDP (Ayeleet *al.*, 2002). Poor nutritive forage are the major causes of livestock productivity in smallholders' farms.

In the drier areas, the quantity of natural forages is often insufficient, whereas in the wetter areas, the feed supplies are usually ample but their protein and energy concentrations are low and they are poor quality. In both areas, feed shortages and nutrient deficiencies occur mainly in the dry season and similar to in the study area, livestock is greatly dependent on crop residues for feed and the farmers usually harvest fodder from thinned crop plants, weeds, and defoliated leaves (Kassa, 2003).

The increased utilization of leguminous and other tree and shrub species, fed as a high quality supplement to the low quality natural pastures and crop residues, has been recognized as one of the means of improving the forage supplies to ruminants in pastoral and crop-livestock systems. In the highland part of Ethiopia, *S. sesban* N-fixing and deep rooting shrub with good-quality foliage is one of the most

promising species for short-duration cover cropping (Desaeger and Rao, 2001) and serve as protein supplement to poor quality roughages or as substitute for commercial protein supplements (Mekoya *et al.*, 2009). Apart from this, its capacity to control soil erosion, hence restore, and maintain soil fertility makes it a useful component of traditional agroforestry (Degefu *et al.*, 2011). *S. sesban* tree has a high level of foliage nitrogen and is an excellent supplement to protein-poor roughage (Sabra *et al.*, 2010). The leaves and tender branches of this tree have high levels of protein (with 20 to 25% crude protein), and easily digestible when consumed by ruminants (Pravin *et al.*, 2012). It has a long history of use as a source of cut-and-carry forage (Naiket *et al.*, 2011). In Ethiopia, feeding *Sesbania* leaves and young twigs have become increasingly important as a protein rich supplement to a basal diet of either grass or poor quality forage for ruminants (Tessema and Baars, 2004).

Although *S. sesban* tree can improve soil fertility through nitrogen accumulation and help prevent soil erosion. As well as providing source of fuelwood, a commodity that is often in short supply especially in the areas with high population density. The perennial *Sesbania* species have considerable potential for use in agroforestry as they show a rapid early growth, grow under various ecological conditions and do not require difficult management procedures. *Sesbania sesban* found in areas with a semi-arid to sub humid climate, with a rainfall between 500-2000 mm per year. In the regions with low precipitation however, they occur primarily on poorly drained soils, which subjected to periodic waterlogging or flooding. Because of its good tolerance to low temperatures. *Sesban* is adapted to a wide variety of soil types, ranging from loose sandy soils to heavy clays. It has an excellent tolerance to waterlogging and flooding (Shelton, 1994). Therefore the activity was done for the objectives to select and recommend high yielding and adaptable *sesbania sesban* accessions for small farmer's households

MATERIALS AND METHODS

Description of the Study Area

The study was conducted in East Hararghe Zone, Meta district, Buraka Jalala Kebele FTC. The altitude of study site is 2130m.a.s.l and the annual rainfall of the area range from 650- 900mm. Randomized Complete Block Design (RCBD) with three replications was used. Plot size of 3x3 m² was used. A total of five (5) accessions of *Sesbania sesban* with one local check were evaluated. Seedling was raised and transplanted to well-prepared seed bed at spacing of 1 m between plants. Thinning was done two times during theseedling establishment and after transplanting.

Data Collection and Measurement

Survival rate: count all plants after transplanted for a month on total plot size that was survived

Plant height: it was measured at 5 months interval after the plants survived and take average per annuals from the ground level to the tip from five randomly taken plants and was averaged on per plant basis by using 5 m scaled meter.

Dry leaf weight: it was taken after chopping into 5 cm-8 cm length of 200g samples and then sun-dried until constant weight for dry biomass and then converted tone per hectare based

Dry stem weight: it was taken after chopping into 5 cm-8 cm length of 500g samples and then sun-dried until constant weight and then converted tone per hectare based

Leaf to stem ratio: the ratio of dry leaf weight to dry stem weight multiply by 100%

Chemical composition and quality parameters: Forage samples were taken randomly from three plants from the net plot for determination of dry biomass yield. The dry biomass weight of the sample taken after partial sun-dried of 150 g to determine the dry matters then the samples were oven-dried at 65 °C for 72 hours. The samples were analysis for Dry Matter (DM), Crude Protein (CP), fiber and Ash in Haramaya University nutritional laboratory.

Dry matter yield (DM): samples were prepared from the fresh samples and partial sun-dried then were oven-dried to a constant temperature at 105 °C for 16 hrs then re-weighted the dried samples. Laboratory dry matter % (Lab DM %) = $\{(W6 - W4) / (W5 - W4)\} \times 100\%$ Where: W4 = Empty weight of container in grams W5 = Initial weight of sample in grams W6 = Dry weight of sample and container in grams

Crude protein (CP): By micro-Kjeldahl method (AOAC, 1994) .The, crude protein content was determined by (Jackson, 1962).

Fiber: By Van Soest method (detergent Method) used to determine insoluble cell wall matrix such as; Neutral detergent fiber (NDF), Acid detergent fiber (ADF) and Acid detergent lignin (ADL)(Van Soest, 1967)

Ash: it was determined by igniting the dried sample in a muffle furnace at 500°C overnight, Cool in a desiccator, and take weigh.

Statistical Analysis

Data was analyzed using the Statistical Analysis Software to perform ANOVA (SAS 9.1) in a randomized complete block design. Means of all treatments were calculated and the difference was tested for significance using the least significant difference (LSD) test at $p < 0.05$ (Gomez and Gomez, 1984).

RESULTS AND DISCUSSION

Growth parameters, yield components and yield

Plant height

Mean performance of plant heights of different *Sesbania sesban* varieties are indicated in table 1. Mean plant height were none significantly ($p > 0.05$) differ among tested accession of *Sesbania sesban*. However, the grand mean of plant height (370.17 cm) obtained from *Sesbania sesban* accessions was in line with the plant heights reported by (Negasu and Gizahu, 2019).

Leaf biomass yield

Leaf dry biomass yield was significantly different ($P < 0.05$) among the treatments. The highest leaf dry biomass yield (7.91 t ha^{-1}) was obtained from *S.sesban*1238 whereas the minimum dry weight was recorded from accession of *S.sesban* 10865(4.64 t ha^{-1}). This result is similar to the finding of (Dutt et al., 1983; Gore SB & Joshi RN, 1976; Galang et al., 1990), where the yield of *S. sesban* ranged from 4 to 12 tonnes dry matter/ha/year. However, the result obtained was disagreed with the finding reported by

Negasu and Gizahu, 2019. The variation of dry matter yield might be due to frequency of harvesting and seasonal variations.

Stem biomass yield

The result indicated that significantly differences ($P < 0.05$) observed in stem dry biomass yield among the accession of *Sesbania msesban*. The maximum (25.35 t ha^{-1}) stem dry biomass was obtained from S.sesban 1238, followed by S.sesban1238 (24.86 t ha^{-1}) and S.msesban 15019(22.41 t ha^{-1}). Whereas the least stem biomass yield was recorded from accession of S.sesban 10865 (20.72 t ha^{-1}).

Table 1: The Mean agronomic data, yield and yield component obtained from S.sesban

Treatments	Survival rate (%)	PHt (cm/year)	LBY (t ha^{-1})	SBY (t ha^{-1})	LSR (%)	SY kg/ha
S.sesban 15019	100	354.67	6.21 ^{ab}	22.41 ^{ab}	27.66 ^{ab}	298.8
S.sesban 10865	96.33	384.00	4.64 ^b	20.72 ^b	22.39 ^b	330.4
S.sesban 15036	92.67	360.00	5.51 ^b	21.40 ^{ab}	25.75 ^b	317.2
S.sesban 10885	92.67	370.67	7.23 ^a	25.35 ^a	28.67 ^{ab}	360.2
S.sesban 1238	92.67	386.67	7.91 ^a	24.86 ^{ab}	31.33 ^a	380.9
Local Check	95.33	365.00	5.23 ^b	22.36 ^{ab}	23.39 ^b	319.1
Grand Mean	94.95	370.17	6.39	22.95	26.53	361.82
CV(%)	8.19	12.26	19.37	31.71	29.21	18.26
LSD(0.05)	NS	NS	2.4	4.21	5.65	NS

^{a,b,c} =Means with the same letter in the same column are not significantly ($p < 0.05$) different. PH=Plant Height, LBY=Leaf Biomass Yield, SBY=Stem Biomass Yield LSR=Leaf to Stem Ratio, SY=Seed Yield

Leaf to stem ratio

Leaf to stem ratio was a significantly different ($P < 0.05$) among the treatments. The highest value of leaf to stem ratio (31.67%) was obtained from S.sesban 1238 followed by S.sesban10885 (28.67%) and S.sesban 15019 (27.66%). Whereas the minimum leaf to stem ratio was obtained from accession of *S.sesban 10865* (22.39%). However, the tested S.sesban accession was not showed significant deferent ($p > 0.05$) in survival rate and seed yield among the accession of *Sesbania sesban*



Figures 1. Important Figures that showed during activity was implemented

Chemical compositions of *Sesbania sesban* accessions

The chemical composition indicated that there was a significant difference ($p < 0.05$) among the accessions in Ash and ADL. However, no significant difference in DM, CP, NDF and ADF of *Sesbania sesban* accessions (Table 2). The maximum (15.89%) Ash content was recorded by S.sesban 10865 while the least ash content was recorded from S.sesban 15036 (14.81 %).

The crude protein content was not significantly different ($P > 0.05$) among the treatments of *Sesbania sesban*. In general, CP obtained from tested sesban accession were classified under good quality animal feeds according to General Forage Quality Standards for Livestock Diets classification, > 19% prime (the best quality feeds), QS(1)17-19%, QS(2)14-16%, QS(3) 11-14%, QS(4) 8-10% and QS(5) < 8% of CP indicated that the lowest quality. However the obtained result disagreed with (Pravin *et al.*, 2012) finding; the leaves and tender branches of sesban have high levels of protein (with 20 to 25% CP). The variation might be due to stage of harvesting.

Table 2. The mean of nutritional quality parameters of *Sesbania sesban* accession

Treatments	DM (%)	ASH (%)	CP (%)	NDF (%)	ADF (%)	ADL (%)
S.sesban 15019	85.221	15.095 ^{ab}	18.371	37.324	16.759	15.87 ^b
S.sesban 10865	86.762	15.89 ^a	18.237	37.621	17.178	22.246 ^a
S.sesban 15036	85.291	14.81 ^b	18.187	36.824	16.259	22.746 ^a
S.sesban 10885	86.853	15.062 ^b	18.385	37.136	14.643	14.874 ^b
S.sesban 1238	86.259	15.889 ^a	18.585	36.812	16.678	14.875 ^b
Local Check	86.721	15.012 ^b	18.287	37.216	16.187	18.264 ^{ab}
Grand Mean	86.077	15.349	18.353	36.987	16.304	18.121
CV(%)	1.24	2.13	3.54	3.56	12.45	9.71
LSD(0.05)	NS	0.284	NS	NS	NS	1.523

a,b,c=Means within the same column followed by the same letter or by no letters of each factor do not differ significantly at 5% probability level. DM = dry matter, CP = crude protein, NDF= neutral detergent fiber; ADF = acid detergent fiber; ADL: Acid detergent lignin, NS = none significant. LSD = Least significant difference; CV = coefficient of variance.

On the other hand the maximum (22.746 %) ADL recorded from S.sesban 15036 while the minimum (14.874 %) ADL of *Sesbania sesban* was obtained of S.sesban 10885 accession. In general, this result indicates that the tested *Sesbania sesban* accession have high contents of CP and low contents of fiber fraction (NDF, ADF and ADL). This result indicated that *Sesbania sesban* used as concentrate animal feed resources and easily digestible.

Conclusion and Recommendation

The performance of *Sesbania sesban* was tested in the Highland of east Hararghe zone of Oromia regional state. Among the tested fodder tree (*Sesbania sesban*) S.sesban 1238 and S.sesban 10885 accession have better performances in terms of their biomass yield, leaf to stem ratio and better agronomic parameters and quality forage. Hence, accessions S.sesban 1238 and S.sesban 10885 were found to be the best promising to be demonstrated under the study area and similar agro-climatic conditions

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Evaluation of *Cajanus Cajan* (Pigeon Pea) Genotypes for Agronomic parameters, Yield and Nutritional Composition at Lowland areas of Eastern Hararghe Zone, Ethiopia

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Abstract

The study was conducted at lowland of Fedis district during 2018 and 2019 growing season to evaluate the performance of eleven improved pigeon pea genotypes for agronomic yield and nutritional composition. The study revealed that there were no significant differences among pigeon pea genotypes in terms of days to flowering and maturity. However, significantly higher dry biomass and grain yields were obtained. Grain yield ranged from 1,200 kg ha⁻¹ to 3,030 kg ha⁻¹ with 16524 having the highest and 11575 had the lowest grain yield. Herbage dry matter yield ranged from 2 to 4.4 t/ha. Genotype 16524 had the highest herbage dry matter yield (4.4 t/ha). Leaf to stem ratio of local check was ($p < 0.05$) lower than 11563 genotype but similar to the remaining tested genotypes and seed yield of 16528 genotype was lower than 16524 genotype but similar to the remaining tested genotypes. The crude protein (CP %), ASH%, neutral detergent fiber (NDF %), acid detergent fiber (ADF %) and Acid detergent lignin (ADL %) were significantly ($p < 0.05$) different among genotypes. However, dry matter (DM %) was not different ($p > 0.05$) among genotypes. It was concluded that genotype 16524 had performed best most agronomic parameters, biomass yield, seed yield and LSR as compared to the other tested genotypes and adapted check. Thus, it was found promising to be promoted or demonstrated in Eastern Oromia under climatic conditions similar to Fedis.

Keywords: Pigeon pea, Genotypes, Dry matter, nutritional composition

Introduction

The productivity and sustainability of the livestock industry is influenced by technical and socioeconomic factors. One of the major constraints to increasing livestock productivity in this region is the feed supply, followed by animal health and management practices. Poor animal nutrition and productivity arising from inadequate supply and low quality feed are among the major constraints facing livestock production in developing countries (Fekede *et al.*, 2015b). Similarly, feed shortage in terms of both quantity and quality is the leading problem affecting the livestock productivity in Ethiopia (Fekede *et al.*, 2015a). Nutritional factors are the binding constraint to sustaining livestock production in Ethiopia. According to the Country Feed Balance (FAO, 2018) “The difference between availability of feed resources as dry matter (DM), ME and CP and the requirements of all animal species (i.e. feed balance) showed that feed deficiency in Ethiopia is 9 per cent as DM, while ME and CP deficiencies are 45 per cent and 42 per cent deficient respectively”. These numbers clearly show the lack of quality feed. Thus integration of livestock and cropping systems is essential for sustainable natural resource management and improved livestock productivity (Alemayehu *et al.*, 2017). The production of adequate quantities of good quality forages,

better nutrition, genetics, and the combination of these strategies, are the only way to economically overcome the feed shortage and improve milk/meat production in Ethiopia (Mayberry *et al.*, 2017).

The increased utilization of leguminous and other tree and shrub species, fed as a high quality supplement to the low quality natural pastures and crop residues, has been recognized as one of the means of improving the forage supplies to ruminants in pastoral and crop-livestock systems. Pigeon pea (*Cajanus cajan* L.) is an important grain legume in Eastern and Southern Africa, Asia and Central America. In Africa it is mostly grown by subsistence farmers in the semi-arid areas due to its drought tolerance (Khaki, 2014). In Ethiopia, pigeon peas (*Cajanus cajan*) is known as lowland pulse crop which grows in areas where there is high temperature, erratic rainfall, and short growing season (Amsalu *et al.*, 2016). Pigeon pea is hardy, warm-season, drought tolerant, widely adaptable and tolerant to temperatures as high as 35°C (Vittal *et al.*, 2004). An average annual rainfall of between 600 and 1000 mm is most suitable for pigeonpea production (Green Harvest, 2013). The crop can be grown in a wide range of soil textures, from sandy soils to heavy clays; and is well suited for soil with pH range of between 4.5 and 8.4 (Singh and Oswalt, 1992).

The crop is a multi-purpose leguminous crop that plays important role in food security, maintenance of soil fertility through litter fall and nitrogen fixation, provision of fodder for livestock, source of protein, cash income and fuel for small-scale farmers in subsistence-agriculture (Wilson *et al.*, 2012). It is an important grain legume, particularly in rain-fed agricultural regions in the semi-arid tropics, as well as an excellent, high-protein cover/forage for livestock (Pal *et al.*, 2011) which can be intercropped or grown in mixed cropping systems with cereals or other short duration annuals (Joshi *et al.*, 2001). The main products of pigeon pea are dry grain, green pods and fodder (Mergeai *et al.*, 2001). The leaves and immature stems can be cut and used as a green manure (OAF, 2015). However, this fodder tree was not available in Eastern Oromia; where livestock is greatly dependent on crop residues for feed and the farmers usually harvest fodder from thinned crop plants, weeds, and defoliated leaves. Although this improved fodder tree showed high potential under different research centers, their performance in the study areas was not documented. Although pigeon pea can adapt well under drought conditions there is a great variability of different genotypes for yield under the drought conditions (Deshmukh and Mate, 2013). Therefore, the study was conducted to determine the performance of different pigeon pea genotypes and to select/identify the high yielding genotype (s) in dry matter yield and nutritional qualities suited at lowland agro-ecologies of Eastern Oromia.

MATERIALS AND METHODS

Description of the study area

The study was conducted under rain-fed conditions during 2018 and 2019 growing season at Fedis Agricultural Research Center, on Boko station. It is 550 km to the East of Addis Ababa and 24 km southeast of Harari city. The experimental site is situated at an altitude of 1500m above sea level, (Fuad *et al.*, 2018). The amount of rainfall varies between 650 and 750 mm, while the average temperature ranges between 25 and 30 °C (Zenna, 2016). Vertisols and *Afilsols* soil type are common to the area (FARC, 2013).

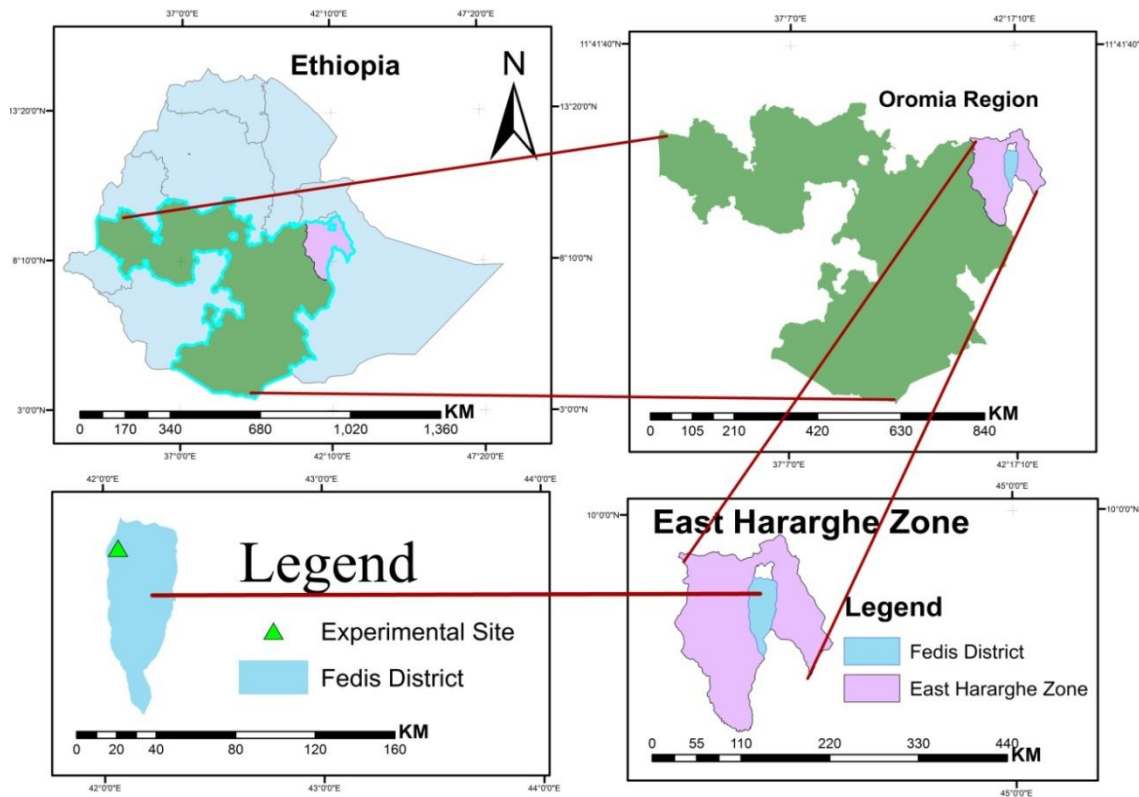


Fig. 1. Map of the experimental site

2. Experimental Design and Treatments

The experiment was conducted using randomized complete block design consisting of 3 replications. The plots were 3.75 m² each with a space of one meter from each plot and with a spacing of 0.75 m between rows. The plot size consisted of four rows of 3 m long and inter and intra row spacing of 0.75 x 0.5 m, respectively. The seeds of ten pigeon pea accessions (16555, 16526, 16527, 11575, 16524, 16528, 16520, 11566, 11563 and 16537) were obtained from international livestock research institute (ILRI) and one local check used. Land was ploughed and harrowed, and then two to three seeds of pigeon pea were planted per hole. Seedlings were thinned down to one plant per hole to ensure uniform plant stand per hole. Weeding was done when necessary. Neither fertilizer nor herbicide was applied to the plots.

2.3. Data collection

Days to flowering, plant height and biomass yield were collected when the plants attained 50% flowering. Data were collected from the two middle rows from each plot. The total harvest per two rows of leaf and stem fresh biomass was weight and about 500g of sub sample taken from each plot and chopped in short length for dry matter determination, and also for laboratory analysis. Sub-samples of air-dried harvested forage were bulked and taken to the Nutrition Laboratory of Haramaya University for analysis of dry matter (DM %), ASH%, crude protein (CP %), neutral detergent fiber (NDF %), acid detergent fiber (ADF %) and Acid detergent lignin (ADL %).

The height of plant at each plant was measured by measuring all the samples harvested for dry matter determination and the average height of all the plants taken as a height of plant at each plant. Seed yield and days to maturity were sampled when the pods attained physiological maturity. Seed yield and days to maturity were sampled when the pods attained physiological maturity.

Statistical Analysis

Data was analyzed using the Statistical Analysis Software to perform ANOVA (SAS 9.1) in a randomized complete block design. Means of all treatments were calculated and the difference was tested for significance using the least significant difference (LSD) test at $p < 0.05$ (Gomez and Gomez, 1984).

3. RESULTS AND DISCUSSION

Agronomic Yield Performance of Pigeon Pea Genotypes

The mean agronomic yield performance of pigeon pea genotypes evaluated was shown in table 1. Results showed that, there was significant difference ($P < 0.05$) in some of the genotypes evaluated. Analysis of variance showed significance difference ($P < 0.05$) in leaf and stem dry matter yield among the tested genotypes. The highest leaf dry matter (4.4 t/ha), stem dry matter (38.9 t/ha) and seed yield (3030 kg/ha) were recorded from 16524 genotype, whereas genotype 16528 resulted in the lowest leaf dry matter yield (2 t/ha). Increase in dry matter and seed yield might be attributed to more number of branches and number of pods per plant (Rani and Reddy, 2000; Lal and Raina, 2002). The significant differences in those yield components are further explained by Islam and Fakir (2007) that canopy structure, canopy spreading and degree of branching influences most of the yield components such as number of pods per plant. The significant differences were also might be due to reaction of different landraces in temperature and photoperiod. The photoperiod and temperature effects on flowering and plant canopy development in pigeon pea make agronomists to choose cultivars that adapt and perform well to specific climatic conditions (Silim *et al.*, 2005).

The study showed that there were no significant differences ($P > 0.05$) among pigeon pea genotypes in terms of days to flowering and maturity. Almost all the genotypes tested in this study were found to be short duration genotypes which took 10-150 days to reach physiological maturity. However, significant differences ($P < 0.05$) were observed among genotypes for plant height. Pigeon pea genotypes in this study were generally tall, probably due to influence of exposure to long-day conditions and environment (Egbe and Vange, 2008). The difference in plant heights might be also due to sensitivity to photoperiod among genotypes whereby they reacted differently and exposure to long day condition and temperature sensitivity among genotypes. This is evidenced by Robertson (2001), that genotypic differences are influenced by the temperature in terms of plant height. The results also indicated that leaf to stem ratio of local check was ($p < 0.05$) lower than 11563 genotype but similar to the remaining tested genotypes and seed yield of 16528 genotype was lower than 16524 genotype but similar to the remaining tested genotypes. Significantly lower seed yield was obtained by 16528 genotype, whereas the remaining genotypes not different ($p > 0.05$) in seed yield. In general, genotypes 16524, 11563 and 16537 performed better than local check in most agronomic and yield parameters. The highest herbage dry matter yield performance of 16524 genotype revealed that this genotype is better adapted and performed well as compared to the tested genotypes.

Table 1. Agronomic and yield performances of pigeon pea genotypes tested at Fedis during 2018 and 2019 growing season

Genotypes (treatments)	DF	PH(cm)	LDM t/ha	SDM t/ha	SY kg/ha	DM	LSR
16555	103.7	168cd	2.7b	25.6abc	2510ab	152.3	10.57ab
16526	98.3	169.7bcd	2.8b	26.9abc	2200ab	146	10.4ab
16527	98.3	193.4a	2.8b	34.3ab	1870ab	142.7	8.3ab
11575	108	181ab	2b	21.6bc	1200b	148	9.2ab
16524	111	180.8ab	4.4a	38.9a	3030a	151	12.1a
16528	109.3	143.4f	2b	26.2abc	1270b	142	8.8ab
16520	95.7	163.6df	2.7b	27.3abc	2260ab	144.7	10.1ab
11566	109.3	128.1g	2.3b	21.8bc	1730ab	144.3	10.43ab
11563	103.7	176.5bc	3.06ab	26.1abc	2290ab	148	12a
16537	95.3	152.8ef	2.96ab	27abc	2330ab	145.3	10.87ab
Local check	107	178.8bc	2.8b	38.92a	2280ab	184.3	7.9b
Grand mean	103.61	166.91	2.789	27.75	2.0885	146.67	10.309
CV (%)	12.32	4.46	32.38	31.84	31.99	4.75	21.95
LSD (0.05)	NS	12.681	0.737	7.15	1.49	NS	1.847

^{a,b,c,d,e,f} Means with the same letter in the same column are not significantly ($p < 0.05$) different. DF= days to flowering, PH=plant height, LDM= leaf dry matter, SDM=stem dry matter, SY=seed yield, DM= day to maturity, LSR=leaf to stem ratio, NS = none significant. LSD = Least significant difference; CV = coefficient of variance

Chemical composition of Pigeon Pea Genotypes

The chemical compositions of eleven pigeon pea genotypes are presented in table 2. The crude protein (CP), ash neutral detergent fiber, acid detergent fiber and acid detergent lignin were significantly ($p < 0.05$) different among genotypes. However, dry matter (DM %) was not different ($p > 0.05$) among genotypes. In the current study, genotypes 11566 and 16524 had significantly higher crude protein content than 16555, 16528 and 16520 genotypes. However, except with these three genotypes the crude protein content of the remaining genotypes was almost similar to 11566 and 16524 genotypes. Significantly the higher ash content was obtained by 16528 and 16527 genotypes whereas the lower content was observed by 11575 and 16555 genotypes.

Contents of NDF was also significantly different ($P < 0.05$) among the evaluated pigeon pea genotypes. The higher NDF was recorded for 16520 and 16528 followed by 16555 genotypes while the lower value was observed by 16524 and 11563 genotypes. Similarly, significantly the higher ADF was obtained by 16528 and 16520 genotypes where as the lower value was observed from 11563 genotype. Content of ADL also significantly varied ($P < 0.05$) among genotypes, where significantly higher value was recorded for 11575, 16520, 12.936 and 16528 genotypes and significantly lower value was obtained by the remaining tested pigeon pea genotypes.

In general, genotype 16524 showed significantly higher herbage dry matter and seed yield performance. Moreover this genotype had higher nutritional qualities as compared to the tested pigeon pea genotypes.

Table 2. Herbage Chemical composition of Pigeon pea genotypes

Treatments	DM%	Ash %	CP%	NDF%	ADF%	ADL%
16555	92.422	9.075d	19.234b	63.182ab	36.712c	10.842b
16526	92.33	10.164b	21.78ab	58.606cd	33.22e	11.576b
16527	92.882	10.425ab	21.015ab	57.285d	36.215cd	11.326b
11575	92.212	8.954d	21.94ab	56.643de	41.351b	13.076a
16524	92.45	10.232b	23.282a	48.955f	33.96de	10.705b
16528	92.011	10.706a	19.136b	64.972a	44.411a	12.784a
16520	92.223	9.656c	19.113b	64.985a	43.865a	13.214a
11566	92.324	9.456c	23.578a	61.013bc	33.747e	11.361b
11563	92.427	10.335b	22.01ab	45.612f	30.343f	11.453b
16537	92.625	9.419c	21.799ab	57.629cd	40.115b	11.231b
Local check	91.739	9.454c	21.675ab	53.745e	40.208b	12.936a
Grand mean	92.331	9.807	21.324	57.51	37.65	11.844
CV (%)	0.69	1.41	6.96	2.69	2.86	3.98
LSD (0.05)	NS	0.138	1.48	1.55	1.0765	0.472

^{a,b,c,d,e,f} Means within the same column followed by the same letter or by no letters of each factor do not differ significantly at 5% probability level. DM = dry matter, CP = Crude protein, NDF= neutral detergent fiber; ADF = acid detergent fiber; NS = none significant. LSD = Least significant difference; CV = coefficient of variance

Conclusion and recommendation

The study shows that genotype 16524 performed best in biomass and seed yield and had high nutritional qualities among the tested genotypes. These results suggested that pigeon pea has the potential to provide forage of high quality and adequate quantity for livestock when other summer forages are unproductive. Hence, genotype 16524 is recommended for promotion in lowland environments of East Hararghe zone. Moreover, future research should be focused on breeding programs to identify extra short duration pigeon pea and evaluate its yield potential in lowland areas of Eastern Hararghe zone.

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Herbage Yield Evaluation of Desho Grass (*Pennisetum Pedicellatum L.*) Lines under Rain Fed Condition at Sinana Agricultural Research Center, Ethiopia

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ABSTRACT

*This experiment was undertaken at Sinana Agricultural Research center. The aim of the research was to select the best herbage yielder Desho grass (*Pennisetum pedicellatum L.*) among the ecotypes. Randomized Complete Block Design (RCBD) with four replications was used. The result revealed that, the height and dry matter yield in ton per hectare were not significantly varied ($p>0.05$). Other parameters such as number of tillers per plant and stem thickness were differ significantly ($P<0.05$) between four Desho grass ecotypes. Despite, the dry matter yield per hectare was not differ significantly ($P>0.05$) large amount of dry matter yield in ton per hectare of 11.88 ± 1.77 was yielded by Areka DZF# 590 ecotype. Moreover, the highest plant high ($45.75\pm 6\text{cm}$) was obtained on Areka DZF# 590 and the highest number of tillers per plant (206.30 ± 17.68) was observed on Kindu Kosha DZF# 589. Therefore, all lines of Desho grasses were well adapted and performed good under Sinana Condition south east Ethiopia.*

Key words: Ecotypes, Grass lines, Desho

INTRODUCTION

The availability and nutritional quality of feed resources are the most important factors that determine the productivity of livestock. In Bale highland contribution of grazing land to livestock production is declining from time to time due to poor management systems and continued advance of crop farming into native grazing lands. The continued expansion of crop farming in Bale high land is resulting in the increasing share of crop residues as livestock feed resource. This has resulted high dependency of livestock production on low quality feed such crop straw and high slop land for grazing purpose. Utilization of high slop land and over stocking of communal and private grazing land has resulted serious land degradation in the area. As reported by the Ethiopian Highland Reclamation Study (EHRS FAO, 1984) 27 % (over 14 million hectare) of the highland area of Ethiopia were seriously eroded and some 6 million hectare should be completely withdrawn from agricultural use to be re-afforested.

To rehabilitate and improve the productivity of degraded area replacement of local grass with perennial improved grasses was sated as best option in high land rain fall sufficient area and it is very palatable species to cattle and sheep (FAO, 2010). Multipurpose grasses and trees such as Desho grass, Vetiver grass and different forage trees are among recommended materials.

Desho is an indigenous grass of Ethiopia belonging to the family of Poaceae (Smith, G., 2010 and Welle S. *et al.*, 2006). Under its Agro-ecological zone this grass has a *high* potential of biomass production and control water loss effectively and recovers rapidly after watering even under severe drought conditions (Smith, G., 2010). Desho grass adapts best to high-rainfall areas and has highest dry matter yield

compared to other grasses. It is a benched type reproduces through vegetative propagation in the altitude of 1500-2800 ma.sl. This technology can also respond to cropland encroachment onto communal grazing areas and overstocking of livestock that has led to overgrazing, causing further land degradation and serious pasture shortages. It has a high biomass production capacity 30–109 t/ha (Ecocrop, 2010) under irrigation and grows upright with the potential of reaching 90–120 cm based on soil fertility (SLM Ethiopia; Shiferaw et al. 2011). However; adaptation study of this valuable grass was not yet studied. Therefore, this study was aimed to select the best herbage yielding Desho grass among the four ecotypes so as to recommend for livestock producer in the area.

MATERIALS AND METHODS

Description of the Study Area: The experiment was undertaken at Sinana Agricultural Research center (SARC). SARC is located in Bale Administrative Zone, Oromiya Regional State, at (7°N latitude and 40°E longitudes; and 2400 m.a.s.l) and 463km south east of Addis Ababa and east of Robe, the capital of Bale zone in bimodal rain fall area.

Establishment: Four Desho grass ecotypes Kulumsa DZF# 592, Areka DZF# 590, Kindu Kosha DZF# 591 and Kindu Kosha DZF# 589 with Randomized complete block design with four replications were employed. A total of sixteen experimental plots each with 12m² (3m*4m) were used. Each treatment groups were assigned randomly and independently to each experimental block. The root split were planted 0.5m space between and within rows. DAP and Urea fertilizer was applied at the rate of 100 kg/ha and 50kg/ha respectively during establishment as well as to enhance sward consolidation. Management practices (weeding, pest and disease monitoring/ control) were done uniformly.

Data Collection: The collected data were includes plot cover, stand vigor, herbage yield using quadrat, plant height, number of tiller per plant and stem thickness. Incidence of disease, insect and weed infestation were observed and recorded. Plant Height: The height of harvested plant was taken from ground to the tip of the plant. The average of six plant heights was taken randomly from each plot at the 90 days after establishment and 90 days after urea top dressing for re-growth data. Estimation of Biomass Yield: The biomass yield of different Desho grass lines were harvested at 10cm above the ground. Weight of the total fresh biomass yield was measured from each plot in the field and a subsample was taken from each plot to the laboratory, upon arrival at laboratory it was oven dried for 72hours at temperature of 65°C. The oven dried samples were weighed to determine the total dry matter yield. Then the result was converted in to dry matter ton per hectare for comparison (Aklilu, M., 2007). Sampled leaf was separated from stem to determine leaf to stem ratio. Since SARC is located in bimodal rain fall area the required data was collected twice a year for three Consecutive years (2018-2020). The total of six harvesting season's data were used.

Data Analysis: Quantitative data sets were analyzed using general linear model of GenStat Discovery Edition 4. Least significant difference (LSD) test was employed difference (P<0.05). The statistical model for data analysis was

$$Y = \mu + t + bj + e ,$$

Where: Y is the response variable under examination, μ is the overall mean, t is the treatment effect, b is the block effect/ random effect of experimental plots, ij. (j = 1, 2, 3) and e_{ijk} is the random error associated with the observation

RESULTS AND DISCUSSIONS

The Dry Matter Yield (DM) and plant height (PH) were not significantly different ($P>0.05$) (Table 1). The Dry matter yield variation of the four grass lines agree with the result by Tekalegn Y. *et al.*, (2017), but the largest dry matter yield at Sinana was much lower than the result at Wondogenet which was 28.83 ± 2.66 and 11.57 ± 1.77 ton/ha respectively. This high variation was related to the agro-ecology variation of the places where the grass ecotypes were collected in which the Desho grass adapts best to high-rainfall (hot and Humid) areas and has highest dry matter yield compared to other grasses. However, the Average stem circumference (ASC) and Average Number of Tillers per plant (ANT) were significantly different ($P<0.05$). Highest number of tiller per plant (206.30 ± 17.68) were recorded by DZF# 589 ecotype were as the lowest (194.70 ± 17.68) were observed on PZF# 592

Table 1: Over all Agronomic performance of Desho grass lines

No.	Treatment Accessions	Major Agronomic parameters			
		PH (cm) \pm SE	ASC \pm SE	ANT \pm SE	DM \pm SE
1	Kulumsa DZF# 592	43.17 \pm 6.00	2.54 \pm 0.20	194.70 \pm 17.68	11.57 \pm 1.77
2	Areka DZF# 590	45.15 \pm 6.00	2.78 \pm 0.20	198.40 \pm 17.68	11.88 \pm 1.77
3	Kindu Kosha DZF# 591	44.67 \pm 6.00	2.71 \pm 0.20	201.3 \pm 17.68.	10.69 \pm 1.77
4	Kindu Kosha DZF# 589	45.75 \pm 6.00	2.18 \pm 0.20	206.30 \pm 17.68	11.63 \pm 1.77
	Mean	45.18 \pm 6.00	2.38 \pm 0.20	201.10 \pm 17.68	11.44 \pm 1.77
	CV%	13.30	8.60	8.80	18.10
	Sig	ns	*	**	Ns

PH: Plant height, **ASC:** Average stem circumference in cm, **DM:** Dry Matter, **ANT:** Average Number of Tillers per plant, **Sig:** Significant level, ****:** Significant at 0.01 level, ***:** Significant at 0.05, **ns:** non-significant, **CV:** Coefficient of Variation, **SE:** Standard Error of Mean

Desho grass is perennial grass; once the mother plants were established the bio-mass harvesting from re-growth were carried out with optimum management such as fertilizer application applied during rainy season. Accordingly, the re-growth data results were indicated some changes on major parameters.

During the six consecutive harvesting seasons of the three years of the experiment the trends of Average number of tiller per plant was shown increment up to 4th harvesting season (Fig.1). However; the flattened trend were observed on the graph at 5th to 6th harvesting season. The result was related to plant population climax attained at 5th harvesting season which creates high computation on nutrient, water and sun light.

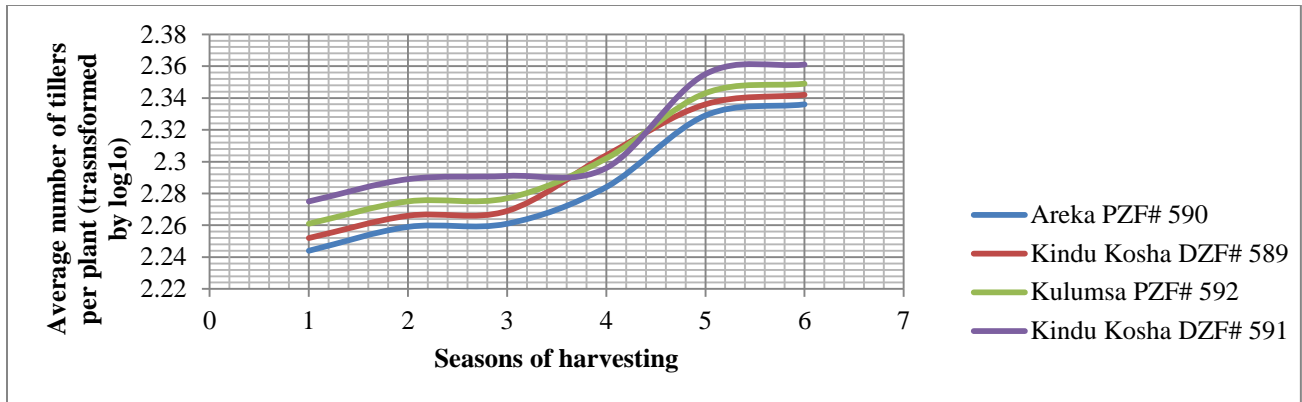


Fig. 1: Trends of number of tillers per plant of Desho Grass lines

The trends of dry bio-mass yield were indicated increment up to 4th harvesting season (Fig. 2). However; the bio-mass yield turning down were observed during 5th and 6th harvesting season.

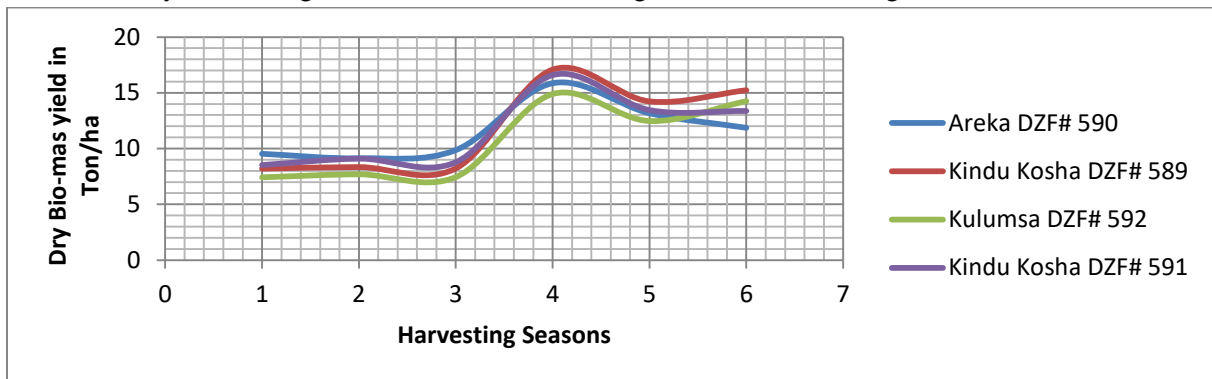


Fig. 2: Trends of Dry matter yield (ton/ha) of Desho grass lines

Thickness of the stem is one of the palatability indicator of the forage plants. The undulated and decreasing patterns were observed on graph showing the trend of plant stem thickness (Fig. 3). Both thickness and Bio-mass yield falling were related to climax attained as the plant population increases which creates high computation on nutrient, water and sun light.

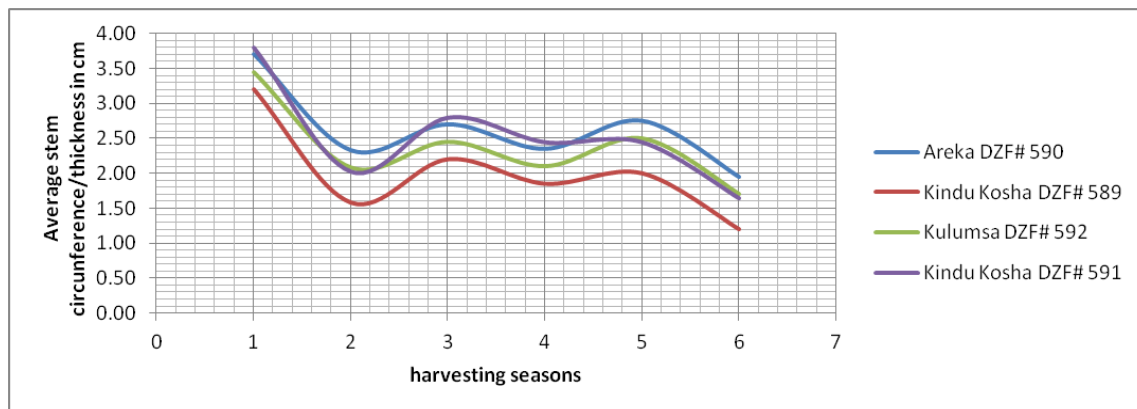


Fig. 3: Trends of average Stem Circumference/Thickness (cm) of Desho grass lines

CONCLUSIONS AND RECOMMENDATIONS

The results revealed non-significant differences in plant height and dry matter yield and the average maximum and Minimum Dry matter yield were 11.88 and 10.69 ton/ha respectively. However, the Average stem circumference (ASC), Average Number of tillers per plant (ANT) were significantly different ($P<0.05$) between four Desho grass lines. Therefore, all tested lines of Desho grasses were well adapted and performed under Sinana environmental conditions. Desho grass is heavy feeder and high biomass producer forage plant. For sustainability of production; continues soil fertility management and adequate moisture availability maintenance is crucial. Further research is needed to exploit its potential under a range of livestock production performances and how to determine the fertilizer requirement in amount and types and water requirement of this grass.

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Evaluation of Alfalfa Cultivars for their Agronomic Performance and Nutritive value in Bore highland and Adola midland Districts of East Guji Zone, Oromia, Ethiopia.

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Abstract

The study was conducted with the objective to identify adaptability, high biomass and seed yield of alfalfa cultivars. Eight alfalfa cultivars; Hunter river, Magna-801-FG, Pioneer(1995) DZF (406), Segule (1396)-408, Peruvian DZF-406, F-G-9-09, F-L-L-77 (406) and Hunter river (4010) were evaluated in randomized complete block design (RCBD) with three replications. At highland area, the result revealed that disease and plant height were significantly ($P < 0.05$) differ among the treatments. The highest value of plant height was measured from Segule (1396)-408 (76.05cm) cultivars followed by Pioneer (1995) DZF (406) (67.9cm), while the shortest plant height was recorded from Hunter river (4010) (49.1cm). On the other hand leaf to stem ratio, biomass yield and seed yield were not show significant ($P > 0.05$) different between treatments. Chemical composition showed that F-G-90-9 was highest in Dry matter (DM) (91.29) and the lowest dry matter (DM) was obtain from Hunter river (4010) (85.3) cultivar. At Midland area the revealed result that disease and plant height were significantly ($P < 0.05$) differ among the treatments. The highest plant height was measured from F-L-L-77 (406) (99.22cm) followed by Peruvian DZF-406 (97.58cm) while the lowest plant height was measured from Magna-801-FG (69.5cm). The highest biomass yield was measured from Segule (1396)-408 (8.33). The highest in organic matter (OM) and lowest in total ash (TASH) was measured from F-L-L-77 (406) cultivar. The highest crude protein (CP) was measured from Magna-801-FG (28.4) followed by Hunter river (4010) (25.9). The result implies that Segule (1396)-408, F-G-9-09, F-L-L-77 (406) and Peruvian DZF-406 cultivars were well performed in agronomic, yield and quality parameters. Thus it could be possible to conclude that the alfalfa cultivars should be recommended for improving the constraint of feed shortage in highland and midland of Guji zone and similar agro ecologies.

Keywords: Medicago Sativa, Cultivar, Adola, Chemical Composition

Introduction

Alfalfa (*Medicago sativa* L.) is often recognized as one of the most important perennial forage legumes worldwide and is widely known as the “queen of the forages” due to its ability to consistently produce high forage yield and forage quality and adaptability to different climatic conditions (Kamalaket *al.*, 2005; Turanet *al.*, 2009).

Alfalfa is a drought tolerant forage crop because it has a deep root system that reaches down to 4 m and to 7-9 m in well drained soils. The plant survive long periods of water stress by impeding its vegetative

growth (Annicchiarico *et al.*, 2010) and accessing water from deep layers through its long root system (Volaire, 2008). The optimum growing air and soil temperatures for alfalfa are 27°C and 12°C respectively, but it is tolerant of air temperatures above and below 27°C (McKenzie *et al.*, 1988). This forage legume is also known as an effective source of biological nitrogen fixation, an energy-efficient crop to grow and an important source of protein yield. The optimum soil pH range for alfalfa is 6.5 to 7.5 and tolerates relative salinity. Alfalfa grows best on well-drained, deep soils but it thrives on sandy soils with adequate moisture and fertility (Barnhart, 1997).

Alfalfa does not grow well on soils where root growth is limited such as shallow hardpans, high water tables, bedrock or acidic sub soils (Lacefield *et al.*, 1987). Poorly drained or waterlogged soils are strongly discouraged for alfalfa because root and crown diseases reduce stand longevity under these conditions. The crop needs very frequent irrigation during its early growth period at an interval of about one week but once the plants are established, subsequent irrigations are provided at an interval of 10-15 days during dry season. Alfalfa is one of the few cultivated forage crops that can produce high level of biomass with minimum inputs. Low quality crop residues need nitrogen supplementation, often provided by forage legumes to become productive diets (Anderson, 1985). Climate, cultivation practices, feed technologies and genetic variations are the main factors affecting the nutritional value of feed for livestock. Forage legumes contribute significantly to livestock production in crop livestock systems. Legume forages generally lead to higher intakes and animal production than grass silages of comparable digestibility (Dewhurst *et al.*, 2003).

Alfalfa nutritive value is identified with protein content which depends on the share of leaves in dry matter yield which in turn is positively correlated with protein content. The proportion of leaves and stems in alfalfa hay can vary greatly, depending on maturity at harvest, cultivars, handling and rain damage (Katic *et al.*, 2006). Protein content in alfalfa dry matter varies from 18 to 25% depending on the growth stage, cultivar difference and other factors. Alfalfa is one of the most important forage legumes of the world as major source of protein for livestock and it is a basic component in rations for all classes of domestic animals (Barnes *et al.*, 1988).

Where alfalfa can easily be grown, it is regarded as key forage for high-producing ruminants because of its richness in protein, palatability, high calcium and vitamin content. In many cases animals feeding on alfalfa do not require supplements. Alemayehu (2002) noted that because of its very high feed value, alfalfa should be used as a supplement for crop residues and natural hay in mixture of 30 percent alfalfa and 70 percent other roughages. Alfalfa produces more protein per hectare than other legume and grasses; therefore, it is widely used for hay production and as pasture for livestock, especially to ruminants (Monteros and Bouton, 2009). Ruminants fed on alfalfa have higher nutrient intake and digestibility than when fed on other forage legumes and grasses (Frame, 2005). To improve availability of livestock feed in terms of quantity and quality, it is better to cultivate alfalfa 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, leaf to stem ratio and nutritional quality of alfalfa cultivars grown at high land and mid land parts of Guji zone.

Materials and Methods

Description of the study area

The experiment was carried out at Bore Agricultural Research Center, Bore district, 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 types of the site is mostly black soil. Bore Agricultural Research Center station is located at Songo which is 7 km from Bore district which is geographically located at 62°4'37" N latitude and 38°34'76" 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 and 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 activities take place, which are the major livelihood of the local people. The total area of District is 1254.56 km². 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 type of the district is nitosols (red basaltic soils) and orthicacrosols (Yazachew E. and Kasahun D., 2011).

Experimental treatments and design

The experiment was conducted at 2018 and 2019 cropping season. The experiment was conducted using eight varieties Hunter river, Magna-801-FG, Pioneer (1995) DZF-406, Segule 1396 (408), Peruvian DZF (406), F-G-9-09, F-L-L-77-406- and Hunter river (4010).

The experiment was conducted in randomized complete block design (RCBD) with three replications. Seeds were sown in rows spaced 20cm and 1m, 1.5m between plot and block respectively on plot size of 2m x 2m (4 m²). Seed & fertilizer was applied and other agronomic crop protection practice was adopted uniformly as per recommendation for production.

Data Collection.

All relevant data like days to 50% emergency, days to forage harvest (maturity), resistance to disease, resistance to insect, growth habit, plant height, Biomass yield, seed yield, leaf to stem ratio and nutritive value were recorded.

Chemical Analysis

Chemical analysis was conducted at Haramaya University Animal nutrition Laboratory workers staff. Samples of feeds were taken in green stage of 50% flowering and dried in an oven for 72 hr at 65°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 5500C and N content of feeds was determined according to Kjeldhal 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). Hemicellulose (HC) and cellulose (C) contents were calculated as NDF minus ADF and ADF minus ADL, respectively.

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, μ = 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

Yield and yield components

Mean value of agronomic and yield parameter of alfalfa cultivars are show in (table 1). The analyzed result show that plant height and disease was significantly ($P < 0.05$) different among the treatments. The highest value of disease was measured from Hunter river cultivar (2.5) followed by Hunter river (4010) (2.3) cultivar. The highest value of plant height was measured from F-L-L-77(406) (99.22cm) followed by Peruvian DZF (406) (97.58 cm) at Adola sub site. The shortest plant height was obtained from Hunter river (4010) (49.1 cm) at Bore on station. On the other hands, leaf to steam ratio, Biomass yield and seed yield were not show significant ($P > 0.05$) different between treatments. However, numerically the highest biomass during conducted experiment at Bore on station and Adola sub site district were recorded (8.13t/ha) and (8.33 t/ha) respectively from Segule-1396 (408) cultivar. The lowest value of biomass was produced from Hunter river (4010) (7 t/ha).

The highest seed yield was measured from F-L-L-77(406) (0.1qu/ha) at Bore on station, followed by Pioneer (1995) DZF-406 and Hunter river (0.09qu/ha) cultivars at Bore on station.

Table 1. The mean value of traits of agronomic and yield component of Alfalfa cultivars at highland areas of Guji zone.

C u l t i v a r s	B o r e o n S t a t i o n					A d o l a S u b - S i t e				
	PH (cm)	D _s	LSR	BM _Y t ⁻¹	SY qt ⁻¹	PH (cm)	D _s	LSR	BM _Y t ⁻¹	SY qt ⁻¹
H u n t e r r i v e r	66.7 ^{ab}	2.5 ^a	0.57	7.4	0.09	88.6 ^{ab}	0.3 ^{ab}	0.56	7.22	0.051
M a g n a - 8 0 1 - F G	63 ^{ab}	0.0 ^c	0.61	7.6	0.08	69.5 ^c	2 ^a	0.61	7.03	0.052
Pioneer(1995) DZF-406	67.9 ^{ab}	2 ^{a b}	0.53	8	0.09	97.24 ^{ab}	1.5 ^{ab}	0.54	8.01	0.066
S e g u l e - 1 3 9 6 (4 0 8)	76.05 ^a	0.0 ^c	0.61	8.13	0.088	88.58 ^{ab}	0.0 ^b	0.69	8.33	0.078
P e r u v i a n D Z F - 4 0 6	66.75 ^{ab}	0.5 ^{bc}	0.66	7.8	0.085	97.58 ^{ab}	1 ^{a b}	0.72	8	0.075
F - G - 9 - 0 9	61.12 ^{ab}	0.83 ^{abc}	0.64	8.11	0.082	82.8 ^{bc}	0.5 ^{ab}	0.58	7.25	0.062
F - L - L - 7 7 _ (4 0 6)	63.2 ^{ab}	2 ^{a b}	0.62	8	0.1	99.22 ^a	0.8 ^{ab}	0.63	8.06	0.056
H u n t e r r i v e r (4 0 1 0)	49.1 ^b	2.3 ^{ab}	0.72	7	0.085	93.8 ^{ab}	2 ^a	0.69	7.2	0.059
M e a n	64.2	1.2	0.62	7.78	0.09	89.7	1	0.63	7.64	0.062
C V	15.9	128	27.3	20.5	17	8.6	156	31.1	22.55	26.05
L S D (5 %)	*	*	n s	N s	N s	*	*	n s	n s	n s

^{a,b,c} Mean in a column within the same category having different superscripts differ significantly ($p < 0.05$) PH (cm)=plant height in centimeter, D_s=Disease resistance, LSR=leaf to stem ratio, SYqt⁻¹=seed yield cunatal per hectare, BM_Yt⁻¹= biomass yield tone per hectare, CV=Coefficient of variation, LSD= Least significant difference, *= significantly different, NS= None significant different.

Plant height at forage harvesting stage

Plant height was significantly differ ($P < 0.05$) for all cultivars at forage harvesting stage (table 1). The highest mean value of plant height was measured from F-L-L-77(406)(99.2cm), followed by Peruvian DZF-406 (97.58cm) cultivar (97.58cm). This is higher than the result reported by Geletiet *al.*, 2014 (78.78 cm).

Disease occurred at different stages

Disease was significantly differ ($P < 0.05$) for cultivars at different of forage stages. At cold temperature environmental conditions favoring disease like leaf spot which reduce leading to leaf deaths on plants. Therefore, primary managing of alfalfa disease making varieties selection is very important component for successful alfalfa production.

Leaf to stem ratio

Leaf to stem ratio at forage harvesting stage which, ranged from (0.53 to 0.72) was not significantly different ($P > 0.05$) among alfalfa cultivars at forage harvesting stage. The highest leaf to stem ratio was measured Hunter river (4010) and Peruvian DZF-406 (0.72). The least leaf to stem ratio was measured from for Pioneer (1995) DZF-406 (0.53) among the cultivars. This is lower than the result reported by Geshawet *al.*, (2015) (0.7-0.91).

Biomass yield

The mean average of biomass yield, at study area were not show significant ($P > 0.05$) different between treatments.

The highest mean value of herbage dry matter yield (DMY) was measured from Segule-1396 (408) (8.33 t/ha) followed by F-L-L-77(406) (8.06t/ha) at midland area. The lowest biomass yield was measured from Magna-801-FG (7.03 t/ha). This is higher than the result reported by Gezahagnet *et al.*, (2017)

Seed yield

The mean average of seed yield, at study area were not show significant ($P>0.05$) different between treatments. The highest mean value of seed yield was measured from F-L-L-77(406) (0.1qu/ha), followed by Pioneer (1995) DZF-406 and Hunter River (0.009qu/t) cultivar at highland area. This is due to long rain fall and soil moisture.

Nutritive value at forage harvesting stage

From alfalfa cultivars (Table 2) F-G-9-09cultivar was higher in %DM (91.29), less in Total ash (TASH) and highest in Organic matter (OM) (80.44). The highest CP was measured from Pioneer (1995) DZF-406(24.9). The mean result of current study were (24.9) crude protein percentage of the cultivars were higher than the mean result reported by Geletiet *et al.*, (2014) (18.87%).

From alfalfa cultivars (table3) the highest dry matter was measured from Segule-1396 (408) (91.8) and Organic matter (81.12). The highest crude protein was measured from Magna-801-FG (28.46), flowed F-L-L-77(406) (25.69) cultivars. The ADL content all of the cultivars ranging from (7.7 – 32.5 %) was higher than that reported by Markovicet *et al.*, (2007) (4.64 %).

Table 2: Mean chemical composition of alfalfa cultivars at highland area.

C u l t i v a r s	DM %	TASH%	OM %	NDF%	ADF%	ADL %	CP%
H u n t e r r i v e r	90.77	11.30	79.46	57.31	48.76	28.66	24.14
M a g n a - 8 0 1 - F G	90.52	13.51	77.01	72.27	46.54	24.27	22.28
Pioneer (1995) DZF-406	91.78	12.51	79.26	54.87	34.54	10.79	24.91
S e g u l e - 1 3 9 6 (4 0 8)	90.97	13.3	77.61	53.41	46.97	8.93	20.02
P e r u v i a n D Z F - 4 0 6	90.94	13.09	77.84	66.15	35.6	11.09	24.49
F - G - 9 - 0 9	91.29	10.8	80.44	58.0	40.82	32.5	11.28
F - L - L - 7 7 _ (4 0 6)	89.96	15.5	74.43	62.73	41.98	11.09	21.3
H u n t e r r i v e r (4 0 1 0)	83.34	14.13	71.21	52.74	46.39	10.79	24.91

^{a,b,c} Mean in a column within the same category having different superscripts differ significantly ($p<0.05$) DM=dry matter, TASH=total ash, OM=organic matter, NDF=neutral detergent fiber, ADF=acid detergent fiber, ADL=acid detergent lignin, CP= crude protein, CV=Coefficient of variation, LSD= Least significant difference, *= significantly different, NS= None significant different.

Table 3: Mean chemical composition of alfalfa cultivars at midland area.

C u l t i v a r s	DM%	TASH%	OM %	NDF %	ADF %	ADL %	CP %
H u n t e r r i v e r	88.34	12.7	75.60	50.73	39.36	14.76	24.72
M a g n a - 8 0 1 - F G	90.87	13.7	77.15	56.70	38.74	14.72	28.4
Pioneer (1995) DZF-406	90.98	12.6	78.38	39.22	35.15	7.95	22.81
Segule-1396 (408)	91.83	10.7	81.12	55.47	40.64	7.78	20.12
Peruvian DZF-406	91.04	11.36	79.68	69.31	35.43	16.37	22.23
F - G - 9 - 0 9	91.29	10.85	80.44	58.0	40.82	32.58	11.28
F - L - L - 7 7 _ (4 0 6)	90.79	9.06	81.73	66.47	47.54	20.56	25.69
Hunter river (4010)	87.85	14.03	73.8	39.3	40.14	14.22	25.9

^{a,b,c} Mean in a column within the same category having different superscripts differ significantly ($p < 0.05$) DM=dry matter, TASH=total ash, OM=organic matter, NDF=neutral detergent fiber, ADF=acid detergent fiber, ADL=acid detergent lignin, CP= crude protein, CV=Coefficient of variation, LSD= Least significant difference, *= significantly different, NS= None significant different.

CONCLUSIONS AND RECOMMENDATION

Segule-1396 (408), F-L-L-77_(406 for midland areas and F-G-9-09, Peruvian DZF-406 for highland areas alfalfa cultivars were well adapted and being productive regarding the plant height, biomass yield and seed yield of alfalfa which is hopeful to fill the gap of low quality and quantity ruminant feed supply of the community. In current study Segule-1396 (408) cultivar was not exposed by any disease sign at both study area. In addition the nutritional values (chemical composition) were promising particularly the dry matter percentage (DM %) and Organic Matter (OM) and less in total ash (TASH) content in Segule-1396 (408) and F-G-9-09. Thus it could be possible to conclude that the Alfalfa cultivars especially Segule-1396 (408) and F-G-9-09 used as a protein supplement for highland and midland of Guji, which were suffering from poor quality roughage and low protein and digestible crop residues which were the major livestock feed sources particularly in Guji. Based on its adaptability, plant height, biomass yield and seed yield, good DM and OM Segule-1396 (408), F-L-L-77_(406, Peruvian DZF-406 and F-G-9-09 is recommended for further promotion in the highland and midland of East Guji zone.

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Adaptability Study of Desho Grass (*Pennisetum glaucifolium*) Varieties in West and Kellem Wollega Zones

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Abstract

The study was conducted with the objectives to identify and select better adaptable, higher herbage yielding forage variety. Four desho grass varieties (Kindu kosha1-DZF-589, Areka-DZF-590, Kindu kosha2-DZF-591, and Kulumsa -DZF-592) 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 plant height was not showed statistically significant variation ($P>0.05$) among Desho grass varieties. However, number of tiller per plant, number of node per plant, length between node per plant, total green fresh yield, and dry matter yield were significantly difference ($P<0.05$) among desho grass varieties. The heighest herbage dry matter yield was recorded from Kulumsa-DZF-592, Kindu kosha2-DZF-591 and Areka-DZF-590 desho grass varieties. These varieties are well adapted and suitable for use as animal feeds under the study areas. As a result, these three desho grass varieties were recommended for livestock producers as feed resources to enhance animal production and productivity in the study sitses and other areas with similar agro-ecologies.

Keywords: *Desho grass, herbage yield; variety*

INTRODUCTION

In Ethiopian agriculture, livestock production plays a fundamental role in the livelihood of the people (Shiferaw *et al.*, 2011). Despite the large livestock population in Ethiopia (CSA, 2015), its contribution to the national economy is below potential, owing to a range of factors including availability and quality of feed, the poor genetic potential of animals for productive traits, poor health care, and poor management practices (Mengistu, 2006; Legesse, 2008). Of these factors, the most limiting is the low quantity and quality of feed (Shapiro *et al.* 2015). Due to rapid population growth in the Ethiopian highlands, traditional communal grazing areas are increasingly being fragmented into cropland to meet growing demand. In turn, massive pressure is placed on the remaining grazing land as overstocking cow and oxen leads to overgrazing and land degradation (Danano, 2007). This pattern negatively affects agricultural productivity and places a direct threat to the livelihoods of local farmers (Smith, 2010).

Adaptable indigenous fodder species like desho grass is used as mitigation strategies of feed shortage in the country. This perennial grass is native to tropical Africa and widespread from West to East Africa (Leta *et al.*, 2013). Though often considered to be a noxious weed (ISC, 2015), in Ethiopia the grass was first used in Southern Nations Nationalities and Peoples' Region and is currently used for both soil conservation practices and animal feed in other regions of the country (Welle *et al.*, 2006; Yakob *et al.*, 2015). The grass has the ability to control water loss effectively and recovers rapidly after watering even under severe drought conditions (Noitsakis *et al.*, 1996; Welle *et al.*, 2006). It has an extensive root

system that anchors well with the soil and it grows upright with the potential of reaching 90–120 cm based on soil fertility. It can grow anywhere from 1500–2800 masl with an optimum elevation over 1700 m.a.s.l on medium to low soil fertility (SLM Ethiopia). Desho grass has many different uses such as a year round livestock fodder (SLM Ethiopia), for erosion control through strip planting (Welle *et al.*, 2006), to rehabilitate degraded land (Smith, 2010), to improve grazing land management (Danano, 2007). Moreover, desho grass provides a small business opportunity for Ethiopian farmers (sale of cut forage and planting material) (Shiferaw *et al.*, 2011).

Desho grass can provide large amounts of green herbage per unit area (30–109 t/ha/year; Heuzé and Hassoun, 2015) and can be a year-round fodder for livestock (Leta *et al.*, 2013). Therefore the study was conducted with the objectives to identify better adaptable and high herbage yielding desho grass varieties.

MATERIALS AND METHODS

Description of the study area

The study was conducted at Mata, Hawagalan in Kelem Wollega, and Nedjo in West Wollega during 2018 and 2019 the main cropping seasons. These sites are located in western Oromia, Ethiopia. The main rainy season covers from April to October. The area is characterized by coffee based farming and a crop-livestock mixed farming system (HSARC, 2012).

Treatments and Experimental design

Four Desho grass varieties (Kulumsa-DZF-592, Kindu kosha1-DZF-589, Kindukosha 2-DZF-591, Areka-DZF-590) were evaluated using randomized complete block design (RCBD) with three replications. These varieties were introduced from Debrezeit Agricultural Research center. The gross plot comprised of eight rows of 3 m length ($8 \times 0.5 \text{ m} \times 3 \text{ m} = 12 \text{ m}^2$). The spacing between plots and blocks was maintained 1 m and 1.5 m, respectively. Fertilizer was applied at the rate of 100 kg ha⁻¹ DAP and 25 kg ha⁻¹ UREA at planting time. Weeding was done as early as possible to eliminate re-growth of undesirable plants and to promote fodder grass growth by increasing soil aeration, the plots were kept weed-free throughout the growth period (Orodho, 2006).

Data collected

The morphological parameters such as plant height and node length per plant were measured with measuring tape and the number of tillers and nodes per plant were computed as the mean of counts taken from five plants that were randomly selected from the middle rows of each plot at 120 days after planting in all locations. Harvesting was done by hand using sickle leaving stubble height of 8 cm and optimum harvesting stage (120 days) according to recommendations made by (Leta *et al.*, 2013; MoALR, 2017). A fresh herbage yield of Desho grass was measured immediately after each harvest using a portable sensitive balance. Subsamples were taken from each plot at each site to determine the fresh yield and dried in the air until constant weight for dry matter yield determination.

Data Analysis

All the agronomic data collected were analyzed using SAS statistical software version 9.3 (2011). The treatments were compared for their significance using the calculated least significant difference (LSD) values at a 5% level of probability. The following model was used for combined analysis:

$$Y_{ij} = \mu + B_i + T_j + \epsilon_{ij};$$

where, Y_{ij} = measured response of variety i in block j ; μ = overall mean; B_i = i^{th} effect of block; T_j = j^{th} effect of treatment; ϵ_{ij} = random error effect of variety i in block j .

RESULTS AND DISCUSSION

Agronomic parameters of desho grass

Number of plant survival

The number of Desho grass survived in all locations are presented in figure 1. Even though, desho grass planted at Nedjo location was more survived than the two locations might be due to incremental day interval of vegetative desho plant took to plant as well as due to variations in moisture, temperature and soil characteristics of three locations. Amongst the varieties tested across locations and years, the Higher number of plant survived/plot was obtained from Kidu kosha1-DZF-589 followed by Kulumsa-DZF-592 and Areka-DZF-590 whereas the lower number of plant survived was obtained from Kindu kosha2-DZF-591.

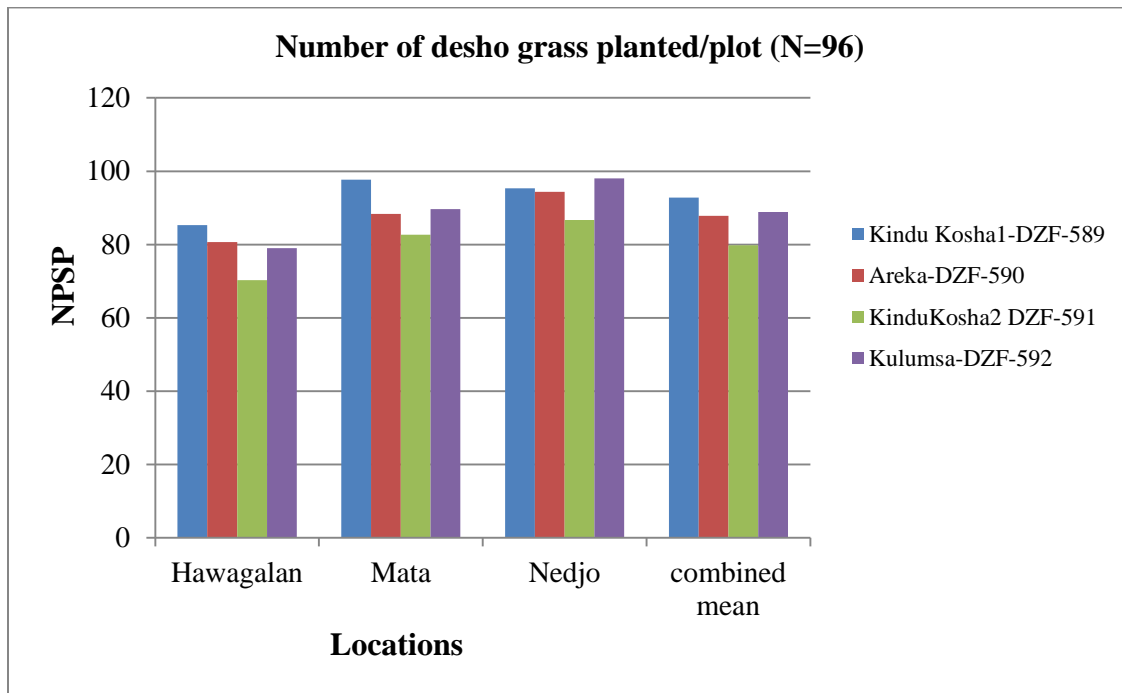


Figure 1. Average number of plant survived per plot ($NPSP = \text{number of plant survived per plant}$)

Plant height at forage harvest

Mean performance of plant height of desho grass varieties were presented in Table 1. The mean of a current results of desho grass varieties across two years showed that plant height was significantly different ($P < 0.05$) among the tested desho grass varieties at Mata. The highest plant height was recorded from Kulumsa-DZF-592 (143.67 cm) followed by Kindu kosha1-DZF-591 and Kindu kosha2-DZF-589 while the lowest plant height was recorded from Areka-DZF-590 (119.80 cm). This significance

difference observed among desho grass varieties might be attributed to the variations in the genetic makeup of the species, soil and environmental adaptability (Zaman, 2006). On the other hand, plant height was not significantly different ($P>0.05$) at Hawa Galan and Nedjo locations among the evaluated desho grass varieties. The absence of plant height different among the desho grass varieties of the current finding was similar with the findings reported by Tekalegn *et al.* (2017) for similar desho grass varieties tested at Wondogenet Agricultural Research Center. Plant height was varied among the experimental locations and such variation might be due to many factors likes, season, weather, soil type and fertility, soil moisture, agroecology, and other factors (Kilcher, 1981).

Table 1. Mean plant height (cm) and number of tiller per plant of Desho grass varieties tested across locations and years (2018 and 2019)

Varieties	Hawagalan		Mata		Nedjo		Combined mean	
	PH (cm)	NTPP	PH (cm)	NTPP	PH (cm)	NTPP	PH (cm)	NTPP
KK1-DZF-589	117.3	54.86	129.87 ^{ab}	36.40	75.66	37.00	107.62	42.76 ^b
Areka-DZF-590	110.53	64.13	119.80 ^b	48.66	74.80	43.60	98.07	52.13 ^a
KK2-DZF-591	100.33	60.06	140.33 ^{ab}	53.20	80.77	47.73	106.9	53.67 ^a
Kulumsa-DZF-592	99.6	61.73	143.67 ^a	50.33	71.66	35.53	108.58	49.20 ^{ab}
Mean	106.95	60.20	133.42	47.15	76.61	40.96	105.29	49.43
LSD _(5%)	17.95	18.84	23.59	19.75	20.26	16.42	10.65	8.319
CV(%)	8.40	15.66	8.85	20.96	13.23	20.07	10.35	17.21
SL	ns	ns	*	ns	ns	ns	ns	*

^{a-b} Means with different letters in a column significantly different ($P<0.05$). KK1= Kindukosha 1; Kindu-Kosha 2; PH= plant height; NTPP= number of tiller per plant; cm= centimeter; LSD=least significant difference; CV= coefficient variation; SL = significant level; * = significant at $P<0.05$; ** = significant at $P<0.01$; ns = non significant.

Number of tiller per plant

There was no significant different ($P>0.05$) for the Numbers of tiller per plant (NTPP) of desho grass at forage harvesting stage among the tested varieties at all locations. The mean number of tiller per plant of the present finding at each location was lower than the findings of Tilahun *et al.* (2017) who reported that the average number of tiller per plant (78.6) of desho grass at Northern highland of Ethiopia. Similarly, it was lower than the findings reported by Heliso *et al.* (2019). This difference might be due to differences in agroecology like altitude, temperature, rainfall, soil, harvesting stage, plant spacing and other factors. Results from the analysis of variance for number of tiller per plant revealed significant effect of location and year ($P<0.05$) among Desho grass varieties (Table 1). The highest NTPP (53.67) was recorded for Kindu kosha2-DZF-591 which was comparable with Areka-DZF-590 and Kulumsa-DZF-592, while the lowest was recorded for Kindu kosha1-DZF-589 variety. The reason for higher NTPP was probably due to the lower plant survival rate per plot which resulted in low resource competition among the plants. The over all mean for tiller number per plant observed in the present study (49.43) was comparable with (48.57) reported by Bimrew (2016), but lower than the findings (78.6, 214, 66.74) reported by different authors (Tilahun *et al.*, 2017; Heliso *et al.*, 2019; Worku *et al.*, 2017).

Number of node and Length between node per plant

There was significantly different results ($P < 0.05$) were observed for number of node per plant (NNPP) and length between node per plant (LBNPP) among the desho grass varieties at Mata while no statistically significant difference ($P > 0.05$) was observed between the varieties at Hawagalalan and Nedjo locations over the study years. Pooled over the four varieties, the highest number of node per plant (NNPP) and length between node per plant (LBNPP) values (15.18 and 6.20 cm) were obtained for Kindu kosha1-DZF-589 and Kulumsa-DZF-592, respectively while the lowest values of number of node per plant (NNPP) (13.16) and length between node per plant (LBNPP) (5.26 cm), for Kulumsa-DZF-592 and Kindu kosha1-DZF-589 in that order.

Table 2: Mean number of nodes and length between node per plant of desho grass varieties tested across locations and years (2018 and 2019)

Varieties	Hawagalalan		Mata		Nedjo		Combined mean	
	NNPP	LBNPP (cm)	NNPP	LBNPP (cm)	NNPP	LBNPP (cm)	NNPP	LBNPP (cm)
KK1-DZF-589	14.86	6.05	15.60 ^a	6.44 ^b	15.07	3.28	15.178 ^a	5.26 ^b
Areka-DZF-590	13.26	6.04	12.80 ^b	7.940 ^a	13.87	3.39	13.31 ^b	5.79 ^{ab}
KK2-DZF-591	12.93	6.18	15.53 ^a	7.54 ^{ab}	13.20	3.77	13.78 ^b	5.83 ^{ab}
Kulumsa-DZF-592	12.60	6.57	13.93 ^{ab}	8.45 ^a	12.60	3.56	13.156 ^b	6.2 ^a
Mean	13.41	6.21	14.47	7.59	13.68	3.50	13.85	5.76
LSD _(5%)	2.51	1.30	2.72	1.18	3.20	0.78	1.29	0.65
CV(%)	9.36	10.48	9.43	7.80	11.71	10.72	9.49	11.6
SL	ns	ns	*	*	ns	ns	*	*

^{a-b} Means with different letters in a column significantly different ($P < 0.05$). KK1= Kindukosha 1; Kindu-Kosha 2; PH= plant height; NNPP= number of node per plant; LBNPP= length between node per plant; cm= centimeter; LSD=least significant difference; CV= coefficient variation; SL = significant level; * = significant at $P < 0.05$; ** = significant at $P < 0.01$; ns = non significant.

Forage Yield

Green forage yield

Results for analysis of variance for green forage yield indicated significant differences observed ($P < 0.05$) between the tested Desho grass varieties at the study sites. The combined mean green forage yield of desho grass varieties was 42.31 t/ha with values ranging from 39.15 t/ha for Areka-DZF-590 to 47.58 t/ha for Kulumsa-DZF-592. The green forage yield among the Desho grass varieties in the present study was contrary with the findings of Gadisa *et al.* (2019) who reported for similar desho grass varieties at Mechara, Eastern Ethiopia. O'Connor (1982) suggested that differences in response among grass species result largely from climatic conditions under different conditions grasses have different growth rates.

Table 3: Combined mean values of forage yield (in both fresh and DM bases) of Desho grass varieties tested across locations and years (2018 and 2019)

Varieties	Forage yield (ton/ha)							
	Hawagalan		Mata		Nedjo		Combined mean	
	GFY	DMY	GFY	DMY	GFY	DMY	GFY	DMY
KK 1-DZF-589	51.41 ^a	6.689 ^{ab}	48.34 ^b	7.05 ^b	25.52 ^b	4.48	41.76 ^b	6.07 ^b
Areka-DZF-590	47.11 ^a	7.102 ^a	44.33 ^b	6.90 ^b	27.70 ^{ab}	4.51	39.71 ^b	6.17 ^{ab}
KK2-DZF-591	40.87 ^b	5.622 ^b	49.10 ^b	7.83 ^{ab}	30.65 ^a	5.11	40.21 ^b	6.19 ^{ab}
Kulumsa-DZF-592	50.52 ^a	7.101 ^a	59.80 ^a	9.20 ^a	32.43 ^a	5.33	47.58 ^a	7.21 ^a
Mean	47.48	6.63	50.39	7.74	29.07	4.86	42.31	6.41
LSD (5%)	5.40	1.09	6.27	1.75	4.79	0.90	3.29	1.08
CV (%)	9.19	13.35	10.05	18.23	13.29	14.97	11.58	15.74
SL	**	*	**	*	*	ns	*	*

^{a-b}Means with different letters in a column significantly different ($P < 0.05$). KK1= Kindukosha1; Kindu-Kosha2; GFY= green forage yield; DMY=dry matter yield; LSD=least significant difference; CV= coefficient variation; ha =hectare; SL = significant level; * = significant at $P < 0.05$; ** = significant at $P < 0.01$; ns = non significant.

Herbage Dry matter yield

Results for analysis of variance for herbage Dry matter yields (DMY) revealed that significance differences observed ($P < 0.05$) between the varieties at Hawagalan and Mata, and mean values were recorded 6.63 t/ha and 7.74 t/ha, respectively, while non significant results obtained at Nedjo site with mean value of 4.86 t/ha. The combined mean DM yields of Desho grass varieties was 6.41 t/ha with values ranging from 6.07 t/ha for Kindu kosha1-DZF-589 to 7.21 t/ha for Kulumsa-DZF-592 (Table 3).

Similarity in herbage DM yields at Nedjo site among the desho grass varieties in the present study was in line with the findings of Tekalegn *et al.* (2017) who reported that similar values in DM yields of desho grass varieties at Wondogenet, Southern Ethiopia. Gadisa *et al.* (2019) also reported significant differences in herbage DM yields for similar desho grass varieties at Mechara, Eastern Ethiopia which supports the result obtained at Hawagalan and Mata locations in the present study.

In contrary to the current study, Bimrew (2016) reported higher herbage DMY (14.65 - 16.84 t/ha) for desho grass produced at different altitudes of northern Ethiopia. Similarly, forage DMY of desho grass in this study was lower than the results reported by different Authors (Gadisa *et al.*, 2019 (24.69 t/ha); Tekalegn *et al.*, 2017 (25.05 t/ha); Tilahun *et al.*, 2017 (16.1 t/ha); Heliso *et al.*, 2019 (19.06 t/ha); Worku *et al.*, 2017 (11.4 t/ha)). However, the pooled mean herbage dry matter yield (6.41 t/ha) of desho grass in this experiment was higher than the finding of Yegrem *et al.* (2019) who reported lower mean herbage DM yield (3.51 t/ha) of desho grass at East Gojjam, Northwest Ethiopia. The significant differences observed were probably due to different agro-ecology, agronomic activities like harvesting stage, spacing, cutting cycle, fertilizer, various soil and climate conditions.

Conclusions and Recommendations

The adaptation trial of four desho grass varieties (Kulumsa-DZF-592, Kindu kosha1-DZF-589, Kindu kosha2-DZF-591, Areka-DZF-590) were conducted at Mata, Hawagalan in Kellem Wollega, and Nedjo in West Wollega during 20018 and 2019 main cropping seasons. In this study several parameters number of tiller, node per plant, node length and dry matter yield were shown significant differences among the tested desho grass varieties while plant height was not brought any change between desho grass varieties. Based on the high herbage dry matter production potential, three varieties- Kulumsa-DZF-592, Kindukosha2-DZF-591 and Areka-DZF-590 were selected as adapted improved forage varieties used for animal feeds in livestock industry. Therefore, these three varieties were recommended and further be demonstrated and scaled up in the study sites and other areas with similar agro-ecologies.

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Adaptation Trial of Alfalfa (*Medicago Sativa* L.) Varieties for Midland Agro-Ecology of Kellem Wollega Zone, Oromia, Ethiopia

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Abstract

Six alfalfa varieties were evaluated to identify adaptable and high biomass yielder under rain fed condition of mid-altitude of Kellem Wollega Zone, Oromia, Ethiopia. The experiment was conducted in Randomized Complete Block Design (RCBD) with three replications. DAP fertilizer at the rate of 100 kg/ha was uniformly applied at sowing. Agronomic parameters and biomass yield were determined. Results revealed that the tested varieties were varied significantly ($P < 0.05$) for days to emergence, plant coverage, days to 50% flowering (forage harvest), plant height, and biomass yield. Accordingly, variety Hunter river was early emerged (8.33 days) and had maximum (68.33%) percentage of plot coverage though DZF-552 (9.08 days) was late emerged and had minimum (56.33%) percentage of plot coverage. Variety DZF-552 was late flowering while F-L-L-77 was early flowering. Hairy Peruvian (70.92 cm) gave the maximum plant height though DZF-552 (55.08 cm) gave a minimum height. The highest mean fresh biomass (6.46 t/ha) and dry matter yield (1.28 t/ha) were recorded from Segules-1359 variety. On the other hand, the lowest fresh biomass (4.79 t/ha) and dry matter yield (0.97 t/ha) were recorded from variety DZF-552. Based on the results, it could be concluded that variety segules-1359, F-L-L-77, Hunter river, and Hairy Peruvian could be recommended to grow under the midland ecological condition of Kellem Wollega zone and similar agro ecology for livestock producers as feed resources.

Key words: Alfalfa; Biomass yield; Variety

INTRODUCTION

Feed scarcity both in terms of quantity and quality is a major bottleneck for livestock production in Ethiopia. Improved forage crops play an important role in sustaining the livelihoods of small and medium scale farmers, mainly as a result of their positive effects on livestock production and contribution to economic and environmental sustainability. Many indigenous forage species in Ethiopia have low productivity, low nutritive value, and digestibility, which reduce their usefulness for livestock nutrition (Alemayehu, 2002). On the other hand, one way of improving livestock production and productivity is to introduce improved forage development and proper supplementation with leguminous forages (Poppi and McLennan, 1995). Forage legumes have several advantages to mixed crop-livestock production systems especially in being protein source, which is usually the most limiting nutrients in tropical animal diets and can be grazed, harvested, and fed fresh or stored as hay or silage (Harricharan *et al.*, 1988). One of such potential legume species for integration into existing livestock feeding system is alfalfa (*Medicago sativa* L.).

Alfalfa is one of the most important forage crops worldwide due to its high forage quality, yield, and adaptability to different climatic conditions (Turan *et al.*, 2009) and it is a basic component in rations for all classes of domestic animals. It can be used directly for grazing or conserved as silage or hay and is a reliable forage species that could represent a significant contribution to the livestock sector (Borreani and Tabacco, 2006). As a perennial legume, alfalfa may be used as a cover crop; its roots improve soil texture and its leaves add organic matter and nitrogen to the soil. The herbage DM yield and chemical composition of alfalfa depend on cutting cycles and cultivars, among others. Crude protein tends to be lower in aged alfalfa plants while the content of crude fibers increases (Stancheva *et al.*, 2008).

In the western part of Oromia like anywhere in the country, livestock depends on natural pasture and crop residues. These feedstuffs are grossly low in quantity and quality to sustain production. Most legume forages are protein source in livestock nutrition and of which, those some grown feeds make farmers less dependent on the purchase of the other protein source. Research has identified high yielding and better quality forages adaptable to various agro-ecologies and production systems, improved forages are not yet adopted and developed by the farming community due to inadequate knowledge, poor extension service, and shortage of land and policy issues. So, to address the feed shortage problem in the areas, using available feeds efficiently, improving the nutritional quality of existing feeds by planting legume fodder crops like alfalfa. Therefore, the study was aimed to identify and select better adaptable and herbage yield performance of alfalfa variety grown at Kellem Wollega zone under rain fed condition.

MATERIALS AND METHODS

Description of the study area

The study was conducted at Haro Sabu Agricultural Research Center (HSARC) in midland (altitude) agro-ecology of Kellem Wollega zone of Haro sabu (On station), Mata, Hawagalan and Kombo sub sites (locations) during 2018 and 2019 of the main cropping seasons. The center was located in Western Ethiopia in the Oromia region at 550 km from Addis Ababa. It has an altitude of 1515 m above sea level. It has a warm humid climate with an average annual minimum and maximum temperature of 14 and 30°C, respectively. The area receives an average annual rainfall of 1000 mm and its distribution pattern is uni-modal. The area is characterized by coffee based farming system and crop-livestock mixed farming system (HSARC, 2012).

Treatments and Experimental design

The six Alfalfa varieties (materials) were collected from Debre Ziet, Adami Tulu and Melka Werer Agricultural research centers and evaluated at Haro sabu Agricultural Research Center for the mid altitude of Kellem Wollega zone on four locations. The treatments used were DZF-552, F-L-L-77, Hairy Peruvian, Hunter river, Pioneer-1995, and Segules-1359. The design used was a randomized complete block design (RCBD) with three replications with a plot size of 4m x 3m with 1.5 m, 1 m, and 0.2 m between replication, plot, and row respectively. The varieties were planted by seeds and the seed was sown drilling in a row with a seed rate of 10 kg/ha and fertilizer application of Di-ammonium phosphate (DAP) fertilizer at a rate of 100 kg/ha. Recommended management practices for herbaceous legumes were applied during the experimental periods.

Data collected

Days to emergence, percentage of plot coverage and days to 50% flowering (forage harvest) of each alfalfa variety was recorded. Five plants were taken randomly from the middle rows and measured at the time of forage harvest for plant height parameter. Forage samples were taken from middle rows during 50% flowering and then dried in air until constant weight for dry matter yield determination.

Data analysis

Differences among varieties were tested by using analysis of variance (ANOVA) procedures of SAS (Version 9.0) general linear model (GLM) to compare treatment means. The least significance difference at 5% significance level was used for comparison of means. The following model was used for combined analysis:

$$Y_{ij} = \mu + B_i + T_j + \epsilon_{ij}$$

Where Y_{ij} = measured response of variety i in block j ; μ = overall mean; B_i = i^{th} effect of block; T_j = j^{th} effect of treatment; ϵ_{ij} = random error effect of variety i in block j .

RESULTS AND DISCUSSION

Agronomic Characteristics and Yield Performance

Days to Emergence and Plot Coverage

Days to emergence and percentage of plot coverage of alfalfa was presented in Table 1. The combined analysis of variance for days to emergence were significantly different ($p < 0.05$) among alfalfa varieties. Variety DZF-552 was had late days to emerge which was comparable with other varieties except Pioneer-1995 and Hunter river varieties (early emerged). This difference of days to emergence was attributed to variety, seed depth, and environmental condition (temperature, rainfall, moisture) disparities. The combined analysis of variance for plot coverage percentage at the age of eight week were significantly different ($P < 0.05$) among alfalfa varieties. The combined analysis of variance showed that higher percentage of plot coverage was obtained from Hunter river which was insignificantly differed as compared to the rest varieties except DZF-552 that had the lowest percentage of plot coverage. This indicates that dues to forage genetic makeup difference.

Table 1. Mean days to emergence and plot coverage at eight weeks of alfalfa varieties

Varieties	DE				Combine d Mean	Plot coverage (%)				Combine d Mean
	Harosa bu	kombo	Hawag alan	Mata		Harosab u	Komb o	Hawaga lan	Mata	
DZF- 552	8	8	8.33	12 ^{ab}	9.08 ^a	46.67	79.33	47.67	51.67 ^b	56.33 ^b
F-L-L-77	8	7.33	8.33	12.33 ^a	9 ^{ab}	51.67	82	51.67	79 ^a	66.08 ^a
Hairy Peruvian	7.67	8.67	8	11.67 ^{ab}	8.75 ^{abc}	51.67	81.67	50.67	69.33 ^a	63.33 ^a
Hunter river	7.33	7.33	7.67	11 ^b	8.33 ^c	55	83.33	56.67	78.33 ^a	68.33 ^a
Pioneer – 1995	7.33	7.33	7.67	11.33 ^{ab}	8.42 ^{bc}	53	82	53	76 ^a	66 ^a
Segules- 1359	7.33	7.33	7	12 ^{ab}	8.50 ^{abc}	54	84	52.33	79.33 ^a	67.42 ^a
Mean	7.61	7.50	7.89	11.72	8.68	52	82.05	52	72.28	64.58
LSD (5%)	0.71	1.44	2.04	1.31	0.64	9.34	4.87	10.80	10.21	5.10
CV %	10.63	6.44	14.20	6.16	9.03	8.86	4.45	11.41	7.76	9.61
SEM	0.41	0.33	0.59	0.35	0.56	3.60	1.84	4.72	3.03	4.37
SL	Ns	ns	ns	*	*	Ns	ns	ns	**	**

^{a, b, c} Means with different letters in a column significantly different at ($P > 0.05$). DE = days to emergence; LSD = least significance difference; CV = coefficient of variance; SE = standard error of mean; SL = significant level; * = significant at $P < 0.05$; ** = significant at $P < 0.01$; ns = non significant.

Days to 50% flowering (forage harvesting stage)

Days to 50% flowering (forage harvest) for the tested varieties of alfalfa showed significant ($P < 0.05$) differences at four locations (Table 2). Hairy Peruvian and Pioneer-1995 were early reached for forage harvesting (50% flowering) at Haro sabu and Hawagalan locations respectively, whereas F-L-L-77 was early reached for forage harvest at both Kombo and Mata locations and had comparable 50% flowering with other varieties, while DZF- 552 was significantly ($P < 0.05$) late for forage harvesting stage for all locations of the study area. The result indicated that the overall mean of days to 50% flowering of the tested alfalfa varieties across locations were ranged from 106.83 to 112.33 with the mean of 108.45 days which was required after sowing of seedlings. Amongst the alfalfa varieties tested across locations, the DZF-552 variety was significantly required more days for the forage harvesting stage (112.33 days) followed by segules-1359 (108.17 days), while F-L-L-77 had the early days for 50% flowering (forage harvesting stage). The differences of days for 50 flowerings might be attributed to the genetic variability of the tested varieties. The significance of variety difference for days to 50% flowering of this finding is in agreement with finding that reported by Hidosa (2015), the mean 50% heading of different alfalfa accessions tested was observed significance difference grown on station of Jinka Agricultural Research Center under rainfed condition.

Table 2. Mean days to 50% flowering (forage harvest) Alfalfa Varieties tested per locations

Varieties	Locations				Combined mean
	Haro sabu	Kombo	Hawagalan	Mata	
DZF- 552	116.33 ^a	104.33 ^a	112.67 ^a	116 ^a	112.33 ^a
F-L-L-77	114 ^{ab}	98.67 ^b	107.33 ^{ab}	107.33 ^b	106.83 ^c
Hairy Peruvian	112.67 ^b	100.33 ^b	108.33 ^{bc}	109.33 ^b	107.67 ^{bc}
Hunter river	113 ^b	101 ^b	108.33 ^{bc}	109.67 ^b	108 ^{bc}
Pioneer – 1995	114 ^{ab}	100.33 ^b	106.67 ^c	110 ^b	107.75 ^{bc}
Segules-1359	115 ^{ab}	99.67 ^b	109.67 ^b	108.33 ^b	108.17 ^b
Mean	114.17	100.72	108.83	110.11	108.45
LSD (5%)	2.57	2.65	2.51	2.77	1.19
CV (%)	1.24	1.45	1.25	1.38	1.33
SEM	0.82	0.84	0.79	0.88	0.42
SL	*	*	**	**	**

^{a-c} Means with different letters in a column significantly different at ($P>0.05$). LSD = least significance difference; CV = coefficient of variance; SE = standard error of mean; SL = significant level; * = significant at $P<0.05$; ** = significant at $P<0.01$.

Plant height at forage harvesting stage

Plant height was significantly differed ($P<0.05$) among the alfalfa varieties at the four locations (Table 3). The highest plant height was recorded from hairy Peruvian (67.33 and 69 cm) at Harosabu and Hawagalan, and Pioneer-1995 (81.40 cm) and F-L-L-77 (70.66 cm) at Kombo and Mata locations respectively. Whereas, the lowest plant height at forage harvest was recorded from the variety DZF-552 at all tested locations. The mean plant height of tested alfalfa varieties over locations was ranged from 55.08 to 70.92 cm with a mean of 66.36 cm. The highest mean plant height was recorded from Hairy Peruvian followed by Hunter river, F-L-L-77, and pioneer-1995 respectively, while DZF-552 had the lowest plant height over locations. The differences in plant height among the tested variety were due to the fact that the genetic variability. The significance variety differences for plant height concur with that reported by different authors (Geleti *et al.*, 2014; Hidosa, 2015; Gezahagn *et al.*, 2017). In addition to genetic variability, soil fertility could also contribute to the difference in height over locations. Generally, the presence of genetic variation among the tested varieties, responses of varieties to environmental factors, and their interactions are the major reason for plant height variation in alfalfa. Ullah *et al.* (2009) also reported differences in plant height to be linked to genotypic differences and explained this trait to be influenced by the differential response of genotypes to prevailing sites and crop management conditions. However, the current result disagrees with the report of Turan *et al.* (2017) who noted that the plant height of six alfalfa varieties tested in Eastern Turkey was not significantly different at the forage harvesting stage. The difference between our results could be attributed to such factors as the type of soil, climatic conditions, variety considered, date of flowering/heading for forage harvesting, and other management conditions.

Table 3. Mean plant height (cm) at forage harvesting of Alfalfa Varieties tested per locations

Varieties	Locations				Combined mean
	Haro sabu	Kombo	Hawagalan	Mata	
DZF- 552	47.93 ^c	70.33 ^b	48.73 ^c	53.33 ^b	55.08 ^c
F-L-L-77	62.20 ^{ab}	79.26 ^a	62.33 ^{ab}	70.66 ^a	68.62 ^{ab}
Hairy Peruvian	67.33 ^a	79.73 ^a	69 ^a	67.60 ^a	70.92 ^a
Hunter river	63.27 ^{ab}	79.73 ^a	65.26 ^{ab}	68.93 ^a	69.30 ^{ab}
Pioneer - 1995	61.60 ^{ab}	81.40 ^a	62.26 ^{ab}	66.13 ^a	67.85 ^{ab}
Segules-1359	59.33 ^b	78.20 ^a	60.33 ^b	67.60 ^a	66.37 ^b
Mean	60.28	78.11	61.32	65.71	66.36
LSD _(5%)	6.49	4.77	7.72	9.21	3.26
CV (%)	5.92	3.54	6.93	7.70	5.97
SEM	2.06	1.51	2.45	2.92	1.14
SL	**	**	**	*	**

^{a-c} Means with different letters in a column significantly different at ($P>0.05$). LSD = least significance difference; CV = coefficient of variance; SE = standard error of mean; SL = significant level; * = significant at $P<0.05$; ** = significant at $P<0.01$.

Biomass Yield

Analysis of variance of mean values for biomass yields (fresh biomass and dry matter yield) of six alfalfa varieties at forage harvesting stage are presented in Table 4. The biomass yields (fresh biomass and dry matter yield) and the overall mean were showed significantly ($P<0.05$) vary among the alfalfa varieties over locations across years. The highest fresh biomass yield was obtained from Segules-1359, Hunter river, Hairy Peruvian, F-L-L-77 at Haro sabu, Kombo, Hawagalan and Mata locations, respectively though DZF-552 variety had recorded lowest fresh biomass yield at all tested locations. The present result indicated that the fresh biomass yield of the evaluated varieties of alfalfa was significantly different across the tested locations. This might be due to the variations of genetic inheritance, soil and climate differences, and ages of cutting stages. The significant variety differences observed for fresh biomass yield in the present study in agreement with Hidosa, (2015) who stated that the fresh herbage yield of different alfalfa accessions tested at Jinka Agricultural Research Center was significantly ($P<0.05$) varied.

The overall mean of fresh biomass yield showed that fresh biomass yield was significantly ($P<0.05$) vary among the tested alfalfa varieties. The overall mean fresh biomass yield ranged from 4.79 to 6.46 t/ha with a mean of 5.88 t/ha. The mean indicated that the fresh biomass yield was significantly higher ($P<0.05$) in Segules-1359 and non significance difference with the rest varieties except DZF-552, whereas the lower was obtained from DZF-552. The alfalfa varieties in the present study had the lowest fresh biomass yield when compared with various reported literature values. For instance, the fresh biomass yield values ranging from 16 to 44 t/ha (Hidosa, 2015) and from 29.22 to 43.57 t/ha was reported (Turan *et al.*, 2017), which were indeed much higher than those observed in the present study. The wide differences of the fresh biomass yield values observed in different research findings and our result could be attributed to varietal, environmental differences, harvesting stage, and other management factors.

Table 4. Mean biomass yield (t/ha) of Alfalfa varieties tested per locations across years (2018 and 2019)

Varieties	FBY					DMY				
	Haro sabu	Kombo	Hawa galan	Mata	Combin ed mean	Haro sabu	Komb o	Hawag alan	Mata	Combined mean
DZF-552	3.51 ^{bc}	9.07 ^b	3.06 ^c	3.51 ^d	4.79 ^b	0.66 ^b	1.91	0.63 ^{bc}	0.70 ^c	0.97 ^c
F-L-L-77	3.47 ^{bc}	10.91 ^{ab}	3 ^c	6.54 ^a	5.98 ^a	0.73 ^b	2.28	0.56 ^c	1.18 ^a	1.19 ^{ab}
Hairy	3.10 ^{bc}	10.87 ^{ab}	4.79 ^a	5.40 ^c	6.04 ^a	0.69 ^b	2.245	0.87 ^a	0.91 ^b	1.18 ^{ab}
Peruvian										
Hunter river	3.04 ^c	11.57 ^a	4.42 ^{ab}	6.16 ^{ab}	6.30 ^a	0.59 ^b	2.25	0.86 ^a	1.04 ^{ab}	1.18 ^{ab}
Pioneer-1995	3.62 ^b	9.80 ^{ab}	3.99 ^b	5.56 ^{bc}	5.74 ^a	0.75 ^b	2.08	0.85 ^{ab}	0.96 ^b	1.13 ^b
Segules-1359	4.65 ^a	11.07 ^{ab}	3.88 ^b	6.22 ^a	6.46 ^a	1.05 ^a	2.247	0.69 ^{bc}	1.15 ^a	1.28 ^a
Mean	3.57	10.54	3.86	5.56	5.88	0.74	2.17	0.72	0.99	1.16
LSD _(5%)	0.57	2.45	0.65	0.62	0.78	0.17	0.50	0.15	0.17	0.15
CV (%)	13.37	19.37	14.12	9.28	22.98	18.68	19.23	17.01	14.58	22.44
SE	0.19	0.83	0.22	0.21	0.276	0.056	0.17	0.05	0.06	0.053
SL	**	*	**	**	**	**	ns	**	**	**

^{a-c} Means with different letters in a column significantly different at ($P > 0.05$). FBY = fresh biomass yield; DMY = dry matter yield; LSD = least significance difference; CV = coefficient of variance; SE = standard error of mean; SL = significant level; * = significant at $P < 0.05$; ** = significant at $P < 0.01$; ns = non significant.

Forage dry matter yield of alfalfa varieties were determined by environmental and genetic variability. Results from analysis of variance for DM yield of the tested alfalfa revealed that significant differences observed ($P < 0.05$) between the varieties at Haro sabu, Hawagan and Mata, and mean values were recorded 0.74 t/ha, 0.72 t/ha and 0.99 t/ha, respectively, while non significant results obtained at Kombo location with mean value of 2.17 t/ha. Accordingly, the higher dry matter yield was obtained from Kombo than other locations for the reason that might be due to soil fertility differences among the locations.

The overall mean of DM yield of Alfalfa varieties was 1.16 t/ha with values ranging from 0.97 t/ha for DZF-552 to 1.28 t/ha for Segules-1359. The present result indicated that DM yield was insignificantly ($P < 0.05$) higher in segules-1359 as compared to F-L-L-77, hairy Peruvian, and hunter river but significantly higher than pioneer-1995 and DZF-552. The presence of significant varietal differences for DM yield of alfalfa in this study concurs with reports of other researchers (Walie *et al.*, 2016; Gashaw *et al.*, 2015). Similarly, in evaluating five alfalfa genotypes, Gezahagn *et al.* (2017) observed significant differences between genotypes for dry matter yield with an overall mean of around 6.46 t/ha. Unlike to this finding, Solomon and Tesfay, (2019) reported that the DM yield of selected alfalfa tested under the lowland of Raya valley, northern Ethiopia doesn't showed significant difference among cultivars. The result revealed that mean dry matter yield of alfalfa varieties tested in the present study was found within a range of dry matter yield values of alfalfa from 0.6 to 2.16 t/ha reported by Awad and Bakeri, (2009). However, other researchers reported respective DM yield values ranging from 4.22 to 4.77 t/ha (Geleti *et al.*, 2014) and from 1.78 to 3.23 t/ha (Afsharmanesh, 2009) which was higher than those recorded for the alfalfa varieties in the present work. The wide range of DM yield values observed in different reports could be attributed to varietal differences, various soil fertility, and climate conditions, and agronomic activities of the experimental sites.

Conclusions and Recommendations

The tested alfalfa varieties were significantly ($P < 0.05$) varied in terms of days to forage harvest (50% flowering), plant height, fresh biomass, and dry matter yield. The mean result revealed that DZF-552 had late to reach 50% flowering while F-L-L-77 early reached. The highest mean plant height was recorded from Hair Peruvian though DZF-552 had a lower height. It was also concluded that from the tested alfalfa varieties, Segules-1359 was produced more fresh biomass and DM yield followed by F-L-L-77, Hunter-river and Hairy Peruvian. However, DZF-552 was less yielded than others which showed that low adaptability to the study area. Therefore, based on high biomass yield potential, four varieties: Segules-1359, F-L-L-77, Hunter-river and Hairy Peruvian were recommended to study area. Moreover, the information obtained would benefit for promotion of forage species for demonstration and wider scaling out through scaling up program to the area and similar locations.

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Farmer Participatory Evaluation and Demonstration of Brachiaria Grass Cultivars for Forage Production in Kofale District of West Arsi Zone, Ethiopia

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Abstract

The experiment was conducted with the objective of identifying and selecting Brachiaria grass cultivars with high forage dry matter yield and recommend the best performing ones for the farmers. It was conducted at Farmers Training Center (FTC) of Hulabera rural kebele of Kofale district of West Arsi Zone under rain fed conditions in 2019. Farmers Research Extension group that comprises 20 members (15 male and 5 female) was organized. Training on improved forage production and utilization system was given for farmers and development agents. The experiment was arranged in Randomized Complete Block Design (RCBD) with three replications using three Brachiaria grass cultivars. Prior to harvesting the grass cultivars, mini field day was organized to evaluate and select the cultivars. Farmers determined their own selection criteria. Agronomic and yield data including plant height, number of tiller per plant and biomass yield were collected and analyzed. The results of agronomic and biomass yield of the grass cultivars indicated a significant ($p < 0.05$) differences among the cultivars with respect to the plant height, number of tiller per plant and biomass yield. Maximum plant height (68.23cm), number of tiller per plant (178.36) and biomass yield (1.43 t ha^{-1}) were recorded from Mulato-II. The highest biomass yield of Mulato-II as compared to the other cultivars could be due to the highest number of tiller per plant, which contributes to biomass yield. Variations in biomass yield among the tested cultivars could be due to the differences in growth rate and growth habit, which were mediated through the genotypic and phenotypic differences. According to the farmer's evaluation criteria, Mulato-II scored the highest average point and selected as first mainly due to its better performance in coverage, uniformity, tillering ability, and biomass yield as compared to the other cultivars. Therefore, this cultivar was selected as superior grass due to its better performance and hence, it is recommended for further promotion in Kofale districts and areas with similar agro-ecologies.

Key words: Biomass yield, Brachiaria, cultivars, Forage evaluation, Mulato-II

Introduction

Ethiopia endowed with high livestock population that could enable the country to gain from the growing global markets for livestock products if production and productivity is improved. The estimate of livestock population in the country stands at about 59.5 million cattle, 30.70 million sheep, 30.20 million goats, 2.16 million horses, 8.44 million donkey, 0.41 million mule, 1.21 million camels, 56.53 million poultry and 5.92 million beehives (CSA, 2017). Despite high livestock population and the existing favorable environmental conditions, the current livestock contribution is below its potential due to various reasons associated with a number of complex and inter-related factors such as feed shortage, disease, and drought (Yadessa *et al.*, 2016; Abule, 2015; Berhanu, 2009). According to Gelayenew *et al.* (2016) and Ulfina *et al.* (2013), seasonal feed shortage and inefficient utilization have been identified as the major problems

affecting livestock production and productivity. This creates a challenge to provide the ruminant with good quality forages over extended periods and often animals have to cope with poor quality feeds (Sampaio *et al.*, 2010).

One approach of alleviating the feed problem is the identification and development of forage grass species suitable for the existing climatic condition. Production of good quality fodder is of a great importance for the economical ruminant production. Both quality and quantity of fodder are influenced by plant species (Kaiser and Piltz, 2002), stage of growth (Ghanbari and Lee, 2003) and agronomic practices (Ghanbari and Lee, 2003, Rehman and Khan, 2003). In the study area, livestock feed is based on natural pastures, fallow and stubble grazing and crop residues which are poor in quantity and quality. Thus, the existing feed resources do not meet the nutrient requirements for growth and reproduction of animals. Hence, evaluation and selection of improved forage species like *Brachiaria* grass cultivars with high herbage yield and adaptability to the areas are very important in tackling feed shortage through cultivating such improved forage species and then rehabilitating degraded natural pasture/grazing lands.

Brachiaria is a perennial forage grass of high forage yield potential and adaptability to different agro-ecologies. It also produces more dry matter yield than most tropical grasses during the dry season and has been widely used as quality pastures for animal production and for grazing land improvement. This grass is very important because of its high productivity under intensive use, its tolerance to low fertility, relative freedom from pests and disease and in remaining green long into the dry season. *Brachiaria* species produce high yields, show excellent response to fertilizer, and are persistent. Data on nutritive value indicate that forage from *Brachiaria* is highly palatable to stock, leading to high intake, whether fed fresh or grazed *in situ* (Ndikumana and Leeuw de, 1996). There is a considerable literature showing that *Brachiaria* species can give high yields of forage under good climate and management. Yields range between 5 and 36 t DM/ha/year depending on soil fertility, moisture and fertilizer application (Bogdan, 1977). Studies also indicated that animals that consume *Brachiaria* grass produce higher yields of milk and their manure emits smaller amounts of nitrous oxide. The deep-rooted, productive *Brachiaria* grasses can capture atmospheric carbon on a scale similar to that of tropical forests thus having a further advantage in mitigation of climate change.

Regardless of the importance of this grass, widely adaptable and high yielding cultivars have not been identified for forage production in the study areas. Hence, the experiment was conducted to evaluate and select *Brachiaria* grass cultivars for high dry matter yield under the farmer management conditions and recommend the best performing ones for smallholder farmers in the area.

Materials and Methods

Site and farmers selection

The experiment was conducted at Hulabera rural kebele of Kofale district of west Arsi Zone under rain fed conditions. Kofale districts is located at 280 km south of Addis Ababa and located at 7° 19'N to 7° 40'N and 38° 30'E to 38° 53'E. The area is about 1187 km² with a mean monthly rainfall of 102.6 mm. The mean monthly minimum and maximum temperatures are about 5.40°C and 19.80°C, respectively (Umer Seid *et al.*, 2015). The experimental site was selected in collaboration with district office experts as well as development agents. Livestock potential and feed shortage were considered as criteria for

selection of the kebele. From selected rural kebele, a farmers' group, which comprises 20 members (15 male and 5 female) was selected, and organized as a farmers' research and extension group (FREG).

Farmers training

Prior to conducting the experiment, training on improved forage production and utilization system was organized for the farmers and development agents. The training was focused on routine activities in improved forage production including site selection, land preparation, forage establishment, forage management, harvesting and feeding to animals. Relevant information on Brachiaria grass production and utilization system was also given for the participants. Farmers also raised questions regarding on the production, utilization and advantage of improved forage in general and Brachiaria grass in particular and responses were given accordingly. Challenges and opportunities of improved forage production were raised, and discussed with the farmers group.

Experimental procedure

The trial was established at FTC using three Brachiaria grass cultivars collected from Bako and Melkasa Agricultural Research Centers. The Brachiaria cultivars were *Brachiaria decumbens*, Mulato-I (*Brachiaria brizantha* x *Brachiaria ruziziensis*) and Mulato-II (*Brachiaria ruziziensis* x *Brachiaria decumbens* x *Brachiaria brizantha*). The experiment was laid out in Randomized Complete Block Design (RCBD) with three replications. The grasses were established by splitting on plot size of 5m*3 m with spacing between plots and rows of 1.5m and 50cm respectively. All other cultural practices including weeding were kept at normal and uniform for all plots.

Forage evaluation and selection

Prior to the harvesting, all Farmers Research and Extension Group (FREG) members carried out participatory variety evaluation and selection. Farmers determined their own selection criteria. Accordingly farmers considered plot coverage and uniformity, tillering ability of the grass, plant height, biomass yield, tolerance to disease/ frost, regenerating/re-growth ability and greenness and leafiness of the grasses, as their evaluation and selection criteria.

Data collection and analysis

All relevant agronomic and yield data including plant height, number of tiller per plant and biomass yield were collected. Three plants were randomly selected from each plot and plant height was measured from the base of the plant to the flag leaf. The mean plant height was then calculated. Number of tiller per plant was determined by direct counting of the tillers from three plants that were randomly selected and the average was taken. Forage biomass yield was estimated by harvesting the forage at 50% flowering stage. Plants in the middle row of the plots were harvested and weighed immediately to obtain fresh yield. The harvested forage samples were manually chopped into small pieces using sickle and a sub-sample of 300 gm fresh weight were taken and oven dried at 65°C for 72 hrs for determination of herbage dry matter yield. Forage DM yield (t/ha) was obtained by using James *et al.* (2008) formula which is $DM\ yield\ (t/ha) = (10 \times TFW \times SSDW) / (HA \times SSFW)$. Where; 10 = constant for conversion of yields in kg/m² to tone/ha, TFW = total fresh weight from harvesting area (kg), SSDW = sub-sample dry weight (g), SSFW = sub-sample fresh weight (g). The collected data including agronomic parameters and biomass yield were subjected to SAS 9.0 software while data on farmers evaluation of the technology were organized and summarized by excel sheet.

Results and Discussion

Plant height, number of tiller per plant and biomass yield

The result of agronomic and biomass yield of *Brachiaria* grass cultivars are indicated in (Table 1). There were significant ($P<0.05$) differences among the cultivars with respect to the plant height, number of tiller per plant and biomass yield of the grass. Significantly maximum plant height (68.23cm), number of tiller per plant (178.36) and biomass yield (1.43t/ha) were recorded for Mulato II, followed by Mulato I, with plant height of 53.46cm, number of tiller per plant of 121.93 and biomass yield of 0.87t/ha. On the other hand, *Brachiariadecambus* had showed the least plant height (35.86cm), number of tiller per plant (73.43) and biomass yield (0.46t/ha). Plant height and number of tillers per plant are the major factors that can influence the herbage yield of forage plants. Variations in biomass yield across the cultivars can also be attributed to the differences in growth rate and growth habit, which are mediated through the genotypic and phenotypic differences. This is a common phenomenon in grasses (Mganga K., 2009; Ogillo, 2010). In this study, the highest biomass yield of Mulato II as compared to the other cultivars could be due to the highest number of tiller per plant, which contributes to biomass yield. Tiller density is an important attribute of grasses as it increases the chances of survival and amount of available forage (Laidlaw, 2005).

On the other hand, the agronomic and biomass yield results indicated that all cultivars tested recorded lower performances as compared to other studies (Mutimura and Everson 2012; Clara M. 2013). The low performance of this grass was mainly due to the water logging problem that occurred in the experimental site. Other studies also indicated that *Brachiaria* grasses are not able to tolerate waterlogging (FAO, 2016). Moreover, the variation in agronomic and biomass performances could be due to the temperature, rainfall, soil type, fertilization level, and stage of harvesting (Huhtanen *et al.*, 2006). Most of the grasses perform more at altitudes ranging from sea level up to 1750 m.a.s.l. and at temperature ranges between 30-35°C (FAO, 2016).

Table 1: Agronomic and biomass yield of *Brachiaria* grasses

Cultivars	Plant height (cm)	Number of tiller per plant	Biomass yield (t/ha)
<i>Brachiariadecambus</i>	35.86 ^b	73.43 ^c	0.46 ^c
Mulato-I	53.46 ^a	121.93 ^b	0.87 ^b
Mulato-II	68.23 ^a	178.36 ^a	1.43 ^a
Mean	52.52	124.57	0.92
CV	14.49	5.23	18.54
LSD (0.05)	15.21	13.03	0.34
Sig.level	**	***	**

Means with different superscripts within a column and ** in rows are significantly different ($P<0.05$)

Training

Theoretical and practical training on improved forage production and utilization in general and on *Brachiaria* grass in particular was given for FREGs and development agents at FTCs. Accordingly a total of 20 FREG member farmers (15 male and 5 female) and 2 development agents participated on the training. Practical training was given for the group at the spot (on-farm sites) during forage establishment

and harvesting time. Farmers have raised their opinions and questions regarding the production and utilization system of improved forages including Brachiaria grass. They were very much interested to use the improved forages and get benefits from the forages. However, they raised their worries as to from where they can get adequate planting materials/seeds.

Participatory evaluation and selection of Brachiaria grass

As indicated in Table 2, farmer's research and extension groups (FREG) identified their own criteria for evaluation and selection among the Brachiaria grasses. Accordingly, farmers considered plot coverage and uniformity, tillering ability, plant height, biomass yield, tolerance to disease/ frost, regenerating/re-growth ability, greenness and leafiness of the grasses as their evaluation and selection criteria. Studies conducted by Tewodros and Meseret (2013) also indicated that the major forage species selection criteria were based on its biomass yield, tillering capacity and the like.

According to the farmer's evaluation, Mulato-II scored the highest average point (4.14) and selected as first. Followed by Mulato-I with average score of 3.71 while *Brachiaria decumbens* obtained the least score for the evaluated parameters. Most of the participant farmers appreciated the performances of Mulato-II and given the highest point mainly for parameters such as coverage and uniformity, ability of tiller and tolerance to disease/frost. The performance of *Brachiaria decumbens* was the lowest in most farmers' selection criteria including plot coverage and uniformity, tillering ability, plant height, biomass yield and tolerance to disease/ frost. The performance of Mulato-II and Mulato-I in terms of plant height and biomass yield was similar. On the other hand, all tested Brachiaria grasses did not differ in regenerating/re-growth ability, their greenness, and leafiness according to the farmer's evaluation.

Consequently, Mulato-II was selected as superior grass due to its adaptability and best agronomic and yield performance in the study area. Hence, this Brachiaria grass cultivar (Mulato-II) was recommended for further demonstration and promotion in Kofele districts and areas with similar agro-ecologies.



Picture 1: Farmers group while evaluating and ranking Brachiaria grass cultivars at Hulabera Kebele of Kofale district.

Table 2: Farmers group preference for the Brachiaria grasses

Selection criteria's	Score given for Brachiaria grasses (1-5)		
	<i>Brachiariadecambus</i>	Mulato-I	Mulato-II
Coverage and uniformity	2	3	4
Tillering-ability	3	4	5
Plant height	3	4	4
Biomass yield	3	4	4
Tolerance to disease/ frost	3	3	4
Regenerating/re-growth ability	4	4	4
Greenness and leafiness	4	4	4
Overall score	22	26	29
Average score	3.14	3.71	4.14
Rank	3	2	1

* Ranking of Brachiaria grasses was based on a scale of 1-5, 1 being very poor and 5 being very good

Conclusion and Recommendation

The result of agronomic and biomass yield of Brachiaria grass cultivars indicated variation among the cultivars with respect to the plant height, number of tiller per plant and biomass yield of the grass. The maximum plant height, number of tiller per plant and biomass yield were recorded from Mulato-II followed by Mulato-I. The least agronomic and yield performance were recorded from *Brachiariadecambus*. Theoretical and practical training on improved forage production and utilization in general and on Brachiaria grass in particular was given for a total of 20 member of FREG farmers (15 male and 5 female), 2 development agents. Practical training was given for the group at the spot (on-farm sites) during forage establishment and harvesting time. According to the farmer's evaluation, Mulato-II scored the highest average point and selected as the first. As compared to the performance of other cultivars, most of the participant farmers appreciated the performance of Mulato-II cultivar and given it the highest point with regard to plan coverage and uniformity, tillering ability and biomass yield. Generally, Mulato-II was selected as superior grass due to its adaptability, best agronomic and yield performance. Hence, it recommended for further demonstration and promotion in Kofele districts and areas with similar agro-ecologies.

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Evaluation of Desho (*Pennisetum pedicellatum*) grasses for adaptability and yield performance in different agro-ecologies of East Showa and West Arsi Zone of Oromia, Ethiopia

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Abstract

This experiment was undertaken at Adami Tulu Agricultural Research center (ATARC), Shashemene and Kofele districts for two consecutive years of 2019 and 2020 cropping season with the objective to evaluate the adaptability and yield performance of Desho grass varieties for forage production. Four Desho grass varieties (Areka-DZF # 590, KK2-DZF # 589, KK1-DZF # 591 and Kulumsa-DZF #592) were used. Randomized Complete Block Design (RCBD) with three replications was employed. The result revealed that, the agronomic performance of plant height was none significance ($P>0.05$) at Kofele and Shashamane experiential sites except ATARC. The highest plant height was recorded at Shashamane (105.92cm). Total dry biomass yield in ton per hectare was showed significance difference ($P<0.05$) at all experiential sites. High total dry biomass yield was recorded at Kofele study site from Areka-DZF # 590 and KK2-DZF # 589 varieties (23.71 and 23.02 t/ha, respectively), second was at Shashamane from Areka-DZF # 590 (21.02t/ha) and the last was at ATARC from KK1-DZF # 591 (15.12t/ha). This indicated the total biomass yield production increased with the increment of altitude (from low land to high land). Areka-DZF # 590 variety is well performed in total biomass yield production in ton per hectare at Kofele and Shashamanethan the other tested varieties. Leaf length was none significance different ($P>0.05$) at all study sites. Leaf to stem ratio was significance difference ($P<0.05$) at Kofele and Shashamane experiential sites, but none significance ($P>0.05$) at ATARC. Crude protein (CP) contents of the grass was none significance difference between all varieties at all study sites. The obtained CP value was sufficient for normal rumen function. Therefore, all varieties of Desho grasses were well adapted and performed good under at all study sites. An Areka-DZF # 590 variety is recommended for both Kofele and Shashamanestudy sites, but KK1-DZF # 591 was recommended for low land area. Further research is needed to exploit its potential by application of different agronomic practices by irrigation especially at low moisture areas

Key words: - Adaptability, Biomass yield and Desho grass

Introduction

Shortages of animal feed resources have been identified as one of the major factor limiting the production and productivity of livestock. In West Arsi and East Showa Zones, livestock feed is based on natural pastures, fallow and stubble grazing and crop residues. However, natural pasture and crop residues are poor in quantity and quality (Derejeet *al.*, 2010). Thus, the existing feed resources do not meet the nutrient requirements for growth and reproduction of animals. One approach for alleviating the problem is identification and development of forage species suitable for the existing climatic condition. Hence,

production of adaptable perennial forage species with high herbage yield and quality are very important for tackling feed shortage and rehabilitating degraded natural pasture/grazing lands.

Desho (*Pennisetum pedicellatum*) is one of adaptable multipurpose perennial grass which has an extensive root system that anchors well in the soil. It grows in mid and high altitudes (1500-2800 masl) with a wide adaptation to wide ranges of well-drained soils and topographies, with optimum elevation over 1700 masl on medium to low soil fertility (Welleet *al.*, 2006; Smith, 2010). It has vigorous vegetative growth and a high biomass production capacity 30-109 of green herbage/ha/year and crude protein of 5.4% (Ecocrop, 2010). The grass is convenient for smallholder farmers as a backyard enterprise for cut and carries feeding systems. It can be preserved as hay and silage for use as dry season feed. It also provides good soil cover and used as erosion control and grazing land improvement (Welleet *al.*, 2006; Ecocrop, 2010).

Regardless of the importance of this grass, adaptable and high yielding varieties of Desho grass have not been identified for forage production in the study areas. Hence, this study is designed to evaluate the adaptability and yield performance of Desho grass varieties under West Arsi and East Showa conditions with the objective to evaluate the adaptability and yield performance of Desho grass varieties for forage production.

Materials and methods

Description of study areas

The study was conducted at ATARC, Shashemene (KararuFilacha) and KofeleFarmers Training Center (Hula bara and Girmichu) under rain fed conditions. ATARC represents low land, Shashemenemid, while Kofele is found at highland agro-ecology. This was based on Traditional agro-ecological zones classification of Ethiopia (as cited in Alemayehu, 2006).

Adami Tulu Agricultural Research Center is located in the central rift valley (CRV), 167km south of Addis Ababa on Hawassa road. It lies at a latitude of 7°9'N and 38°7'E longitude. It has an altitude of 1650 meter above sea level and a bimodal unevenly distributed average annual rainfall of 760 mm. Rainfall extends from February to September with a dry period in May to June, which separates the preceding 'short' rains from the following "long" rains. The pH of soil is 7.88 fine sandy loams with sandy clay having sand, silt, and clay in proportion of 34%, 48% and 18% respectively.

Shashamane is one of the district in the Oromia Regional State of Ethiopia. Part of the WestArsi Zone located in the Great Rift Valley, Shashamane is bordered on the south by the Sidamo Region, on the West by Shala, on the north by NegeleArsi, on the East by the Kore, and on the Southeast by Kofele. It's located at south 240 km from the Addis Ababa. It has latitude of 7° 12' north and a longitude of 38° 36' east.

Kofele is one of the district in the Oromia Regional State of Ethiopia. It is named after the administrative center of the woreda, Kofele. Part of the WestArsi Zone, Kofele is bordered on the south by the Kokosa, on the West by the SidamoRegion, on the north by Kore, on the East by GedebAsasa, and on the South east by Dodola. Its latitude and longitude of 7°00'N 38°45'E with an elevation of 2695 meters above sea level.

Forage establishments

About four Desho grass varieties (Areka-DZF # 590, KK2-DZF # 589, KK1-DZF # 591 and Kulumsa-DZF #592)

were collected from different research centers. The experiment was laid out in Randomized Complete Block Design (RCBD) with three replications. Plot size with 3m*3m and spacing of 50cm, 1m and 1.5m respectively between rows, plots and block were used. The plant was established by root splitting. NPS and urea were applied at planting and after establishment at the rate of 100 and 25 kg per ha (Bimrewet *al.*, 2018). 25kg of urea was applied after each harvesting cycle for maintenance (Danano, 2007). Management practices were done uniformly for all experimental plots.

Collected Data

Both Destructive (Herbage yield and leaf to stem ratio) and non-destructive (plant height and number of tillers per plant) sampling techniques were applied.

Dry matter yield determination and Chemical analyses

The biomass yield of different Desho grass varieties were harvested at 50% flowering at 10cm above the ground. Weight of the total fresh biomass yield was measured from each plot in the field and a subsample was taken from each plot to the laboratory, upon arrival at laboratory it was oven dried for 72hours at temperature of 60°C for partial DM determination. The oven dried samples were weighed to determine the total dry matter yield. Then the result was converted in to dry matter ton per hectare for comparison as:

$$(10 \times \text{TotFW} \times (\text{DWss} / \text{HA} \times \text{FWss})) \text{ (Tarawali } et \text{ al., 1995).}$$

Where, TotFW = Total fresh weight, DWss = reweight subsample, FWss = Fresh weight subsamples and HA = Harvesting area.

Sampled leaf was separated from stem to determine leaf to stem ratio. Crude protein was calculated as $N \times 6.25$ (Kjeldahl methods).

Statistical Analysis

Collected data were organized, summarized and analyzed by using SAS. LSD test at 0.05 probability levels to compare the treatment means (Steel and Torrie, 1984).

Results and Discussion

Performance of Desho grass at Adami Tulu Agricultural Research Center on station (ATARC)

The results of agronomic performance of Desho grass varieties tested for adaptability at ATARC were present in table 1. The plant height was none significance among the used varieties ($P > 0.05$) except KK2-DZF # 589 variety. The tallest mean plant height was recorded from KK1-DZF # 591, while the shortest was from KK2-DZF # 589 (Table 1). The average mean value of the current result of plant height is higher than the report of (Birmaduma *et al.*, 2019), which recorded 87.6cm. This may be due different agro ecology, soil fertility, time of harvesting and management practices. The total biomass yield was none significant among the used varieties ($P > 0.05$) except Areka-DZF # 590. This variety was lower in total

biomass yield (11.66t/ha) as compared with other variety. The present result was a line with the report of (Bimrewet *et al.*, 2018). This variety is also the highest in plant height (104.38cm) from the tested variety. Plant height recorded in the current work was higher than the work of (Bimrew *et al.*, 2018)(39.40 cm). This may be due different agro ecology, soil, variety, time of harvesting and management aspects. At ATARC condition KK1-DZF # 591, KK2-DZF # 589 and Kulumsa-DZF #592 are the best in total biomass yield production.

Number of tillers per plant (NTPP) was none significant ($P>0.05$) in the all study area. The NTPP recorded in the current work was higher than report of (Bimrew *et al.*, 2018). Leaf length per plant (LLPP) in the present work was higher than the finding of (Bimrew *et al.*, 2017), but lower than the work of (Genet *et al.*, 2017). This result was might have been vary due to harvesting stage, soil fertility, agronomic practices and other management. Leaf steam ratio (LST) shows none significance difference. The current result (0.78) was lower than the finding of (Bimrewet *et al.*, 2017) at harvesting of 90 and 120days (1.24 and 1.17), but agree with at harvesting of 150days(0.82). This might have been due to reduction in leaf proportion and an increase in the stem fraction of the grass at the advanced stage of harvesting (Buttet *et al.*, 1993)

Crude protein (CP) contents of the tested varieties were none significance difference ($P>0.05$). However, numerically better CP (13.5%) was obtained from KK1-DZF # 591 variety. The mean of the current result (12.5%) was higher than the work of (Ecocrop 2010; Bimrewet *et al.*, 2018; Birmadumaet *et al.*, 2019), which records 5.4%, 7.3% and 9.5% CP, respectively. The mean value of the CP content of Desho grass in the current study was higher than the critical value of 7% required for normal rumen microbial function (Van Soest, 1994). Pasture and other roughage feeds are classified as high, medium and low quality according to their CP contents. Accordingly, roughage feeds with CP content of 9.92 to 15.2%, 6.6 to 9.1% and 3 to 6.5% were classified as high, medium and low quality roughage feeds, respectively (Nsahlai *et al.*, 1996). Based on this report, the mean value of CP contents of Desho grass in the current study can be classified as high.

Table 1. Agronomic performance of Desho grass at ATARC

Variety	Parameters						
	PH(cm)	LL(cm)	TDMY(t/h)	CP%	NTPP(counts)	LSR	Cover
KK1-DZF # 591	104.38 ^a	49.98	15.12 ^a	13.5	90.84	0.79	89.77
KK2-DZF # 589	88.49 ^b	45.57	14.04 ^{ab}	13	89.09	0.78	89.735
Kulumsa-DZF #592	97.96 ^{ab}	47.01	13.80 ^{ab}	10.4	87.04	0.78	89.35
Areka-DZF # 590	102.92 ^{ab}	48.13	11.66 ^b	13.1	86.42	0.77	89.98
mean	98.44	47.67	13.66	12.5	88.35	0.78	89.73
CV (%)	15.36	13.29	21.54	25.7	28.84	13.60	0.705
LSD(0.05)	14.51	ns	2.82	ns	ns	ns	ns

^{a,b} = Means with the same letter in the same row are not significantly different, PH=plant height.

LL=leaf length, TDMY = total dry matter yield, NTPP = Number of tiller per plant LSR=leaf to stem ratio, CV= coefficient of variation kk= kindokosha, LSD= Least significance different and ns= non-significant

Performance of Desho grass at Shashamane (KararuFilicha)study site

The results of agronomic performance of Desho grass varieties tested for adaptability at Shashamane were present in table 2. The agronomic performance of plant height, leaf length and number of tiller per plant were not differ significantly ($P>0.05$) between the four desho grass varieties tested at mid agro ecology. However, total dry matter yield ton per hectare was significant ($P<0.05$) highfor Areka-DZF # 590 variety. Areka-DZF # 590 variety produced 21.02 t/ha dry matter yield. The current result obtained was lower the finding of Tekalegnet *al.* 2017which recorded 28.35 t/ha.The former result was recorded may due to application of irrigation. KK2-DZF # 589 variety was the second highest in total biomass yield production (18.02 t/ha) than the other left varieties. Leaf to stem ratio recorded from Areka-DZF # 590 variety also the highest as compared with other varieties. This variety is well performed by all recorded agronomic performance in this agro ecology. Crude protein (CP) contents of the tested varieties were none significance difference ($P>0.05$) at study site. The mean value of the current result (8.3%) was a line with a work of (Bimrewet *al.*, 2018; Birmadumaet *al.*, 2019), which records 7.3% and 9.5% CP, respectively, but higher than the report of (Ecocrop, 2010). The mean value of the CP content of Desho grass in the currentstudy was higher than the criticalvalue required for normal rumen microbial function (Van Soest, 1994).According to the classification of (Nsahlaiet *al.*, 1996), the CP contents feeds, the current result can be classified as medium in CP contents.

Table 2. Agronomic performance of Desho grass at Shashamane trial site

Parameters							
Variety	PH(cm)	LL(cm)	TDMY(t/ha)	CP%	NTPP(counts)	LSR	Cover
KK1-DZF # 591	103.67	43.88	14.76 ^c	6.9	102.17	0.64 ^{bc}	91.02
KK2-DZF # 589	104.83	43.68	18.02 ^b	8.1	110.00	0.72 ^{ab}	89.95
Kulumsa-DZF							
#592	105.50	39.08	15.79 ^c	8.8	102.67	0.62 ^c	89.435
Areka-DZF # 590	109.67	40.62	21.02 ^a	9.7	117.17	0.74 ^a	89.285
mean	105.92	41.82	17.40	8.3	108.00	0.68	89.92
CV (%)	14.59	12.51	8.09	15.5	13.53	11.72	1.35
LSD(0.05)	ns	ns	1.70	ns	ns	0.1	ns

^{a,b,c} = Means with the same letter in the same row are not significantly different, PH=plant height.

LL=leaf length, TDMY = total dry matter yield, NTPP = Number of tiller per plant LSR=leaf to stem ratio, CV= coefficient of variation, kk= kindokosha, LSD= Least significance different and ns= non-significant

Performance of Desho grass at Kofele (Hula bara and Gurmichu) study sites

The results of agronomic performance of Desho grass varieties tested for adaptability at Kofele were present in table 3. The agronomic performance of plant height, leaf length and number of tiller per plants were none significance difference ($P>0.05$) among the tested varieties. However, total biomass yield and leaf to stem ratio were significantly difference ($P<0.05$). KK2-DZF # 589 and Areka-DZF # 590 are higher in total biomass yield production than the rest two varieties. They produce 23.02 and 23.71 t/ha, respectively. Areka-DZF # 590 variety is the best in total biomass production at Kofele and shashamanestudy sites, but the lowest in ATARC. The current result is similar with the work of (Bimrewet *al.*, 2018).

Crude protein (CP) contents of the tested varieties were none significance difference ($P>0.05$) at study sites. The mean of the current result (7.5%) was a line with a work of (Bimrew *et al.*, 2018; Birmaduma *et al.*, 2019), which reports 7.3% and 9.5% CP, respectively, but higher than the report of (Ecocrop, 2010). The mean value of the CP content of Desho grass in the current study was higher than the critical value required for normal rumen microbial function (Van Soest, 1994). According to the classification of (Nsahlai *et al.*, 1996), the CP contents feeds, the current result can be classified as medium in CP contents

Table 3. Agronomic performance of Desho grass at Kofele

Variety	Parameters						
	PH(cm)	LL(cm)	TDMY(t/ha)	CP%	NTPP(counts)	LSR	Cover
KK1-DZF # 591	102.33	42.12	18.67 ^b	9.3	99.58	0.87 ^b	89.7
KK2-DZF # 589	101.62	44.40	23.02 ^a	6.8	100.4	1.02 ^a	88.95
Kulumsa-DZF #592	101.67	42.57	19.02 ^b	7.5	112.37	0.94 ^{ab}	89.45
Areka-DZF # 590	101.43	43.33	23.71 ^a	6.5	113.19	0.97 ^{ab}	89.25
mean	101.76	43.10	21.14	7.5	106.38	0.95	89.3
CV (%)	18.41	12.68	10.60	27.5	23.17	9.83	1.4
LSD(0.05)	ns	ns	2.7	ns	ns	0.1	ns

^{a,b} = Means with the same letter in the same row are not significantly different, PH=plant height.

LL=leaf length, TDMY = total dry matter yield, NTPP = Number of tiller per plant LSR=leaf to stem ratio, CV= coefficient of variation kk= kindokosha, LSD= Least significance different and ns= non-significant

Generally, the better yield performances were observed at **Kofele** as compared to the other two sites. The mean average value of the total biomass yield recorded at ATARC, shashamane and **Kofele** were 13.66, 17.40 and 21.14 t/ha, respectively. This indicated the total biomass yield production increased with the increment of altitude. The grass can grow at altitude range of 1500–2800 m above sea level (Leta *et al.*, 2013) and performs best at altitude greater than 1700 m.a.s.l. (Danano 2007). The highest average mean value of plant height is recorded at mid agro ecology (105.92cm) and the lowest is at lowland agro ecology (98.44cm).

Conclusions and Recommendations

The result revealed that the highest total biomass yield was recorded at high land study site and the second was at mid agro ecology. This indicated the grass performance is high at high moisture area. However, the highest crude protein content was recorded at low moisture area (ATARC). Leaf length, number of tiller per plant and leaf to steam ratio were none significant at ATARC site, however, plant height and total dry matter yield were shows significant difference. KK1-DZF # 591 is performed well at this site. Plant height, leaf length per plant and number of tiller per plant were none significance at Shashamane and kofele study sites. But, total biomass yield and leaf to steam ratio were shows significance difference. Therefore all tested varieties under all study sites were well adapted and well performed. Among the tested varieties Areka-DZF # 590 was performed more in total biomass yield production particularly at Shashamane and Kofele study sites, therefore this varieties is recommended for

both study sites. KK1-DZF # 591 was the best at ATARC in total biomass production. Crude protein contents of the tested Desho grass varieties were none significance difference at all study sites. The obtained CP value was sufficient for normal rumen function for livestock production. Further research is needed to exploit its potential by application of different agronomic practices by irrigation especially at low moisture areas.

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On-farm Evaluation of Rhodes Grass (*Chloris gayana*) Cultivars at Babile District of East Hararghe, Ethiopia

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Abstract

Feed shortage is one of major challenges that limit livestock production in Eastern Ethiopia. The study was conducted with objectives to select/recommend adaptable and high biomass yielding Rhodes grass cultivars to the study area and other districts having similar agro-ecologies. Five Rhodes grass cultivars (ILRI-7384, CV-massaba-13329, ILRI-6633, DZ-253 and local check) were laid out in a randomized completely block design (RCBD) with three replications. The result showed that days to emergence, days to 50% flowering, dry biomass yield and seed yield parameters have a significant difference ($P < 0.05$). The longest days to reach 50% flowering was recorded for cultivar DZ-253 was 110.67 days followed by ILRI-6633 (109.83 days) while the shortest days to 50% flowering were obtained from ILRI-7384 (108.83). The results indicated that the maximum dry biomass yield (9.42 t/ha) was recorded by DZ-253 which was followed by local check (8.5 t/ha). Whereas, the minimum aboveground dry biomass yield obtained were (7.53, 7.79 and 7.95 t/ha) were obtained from CV-massaba-13329, ILRI-7384 and ILRI-6633 respectively. The results depicted that higher seed yield (291.3 kg/ha) was attained by DZ-253 while the lowest seed yield was attained by CV-massaba-13329 (252.0kg/ha). Thus it can be concluded that DZ-253 and local check were more desirable and recommended for livestock feed in the study area and areas having similar agro-ecologies in Eastern Ethiopia.

Keywords: *Chloris gayana*, Dry Biomass Yield, Fodder Grass.

1. Introduction

In developing countries livestock sector plays an important role in contributing for food and nutritional security and serves as an important source of livelihood for nearly one billion poor people (Frans and Siboniso, 2010). Keeping livestock is an important risk reduction strategy for vulnerable communities, an important provider of nutrients and traction for growing crops in small holder systems. Livestock products like milk, meat and other products contribute 17 percent to kilocalorie consumption and 33 percent to protein consumption globally (Melkamu, 2014). Cattle largely depends on rangeland grazing or crop residues that are poor in quantity and quality. Seasonal fluctuation in the availability and quality of feed has been serious challenges in livestock production and the feed shortage mostly happens in dry season of the year (Ibrahim and Olaloku, 2000). 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.

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).

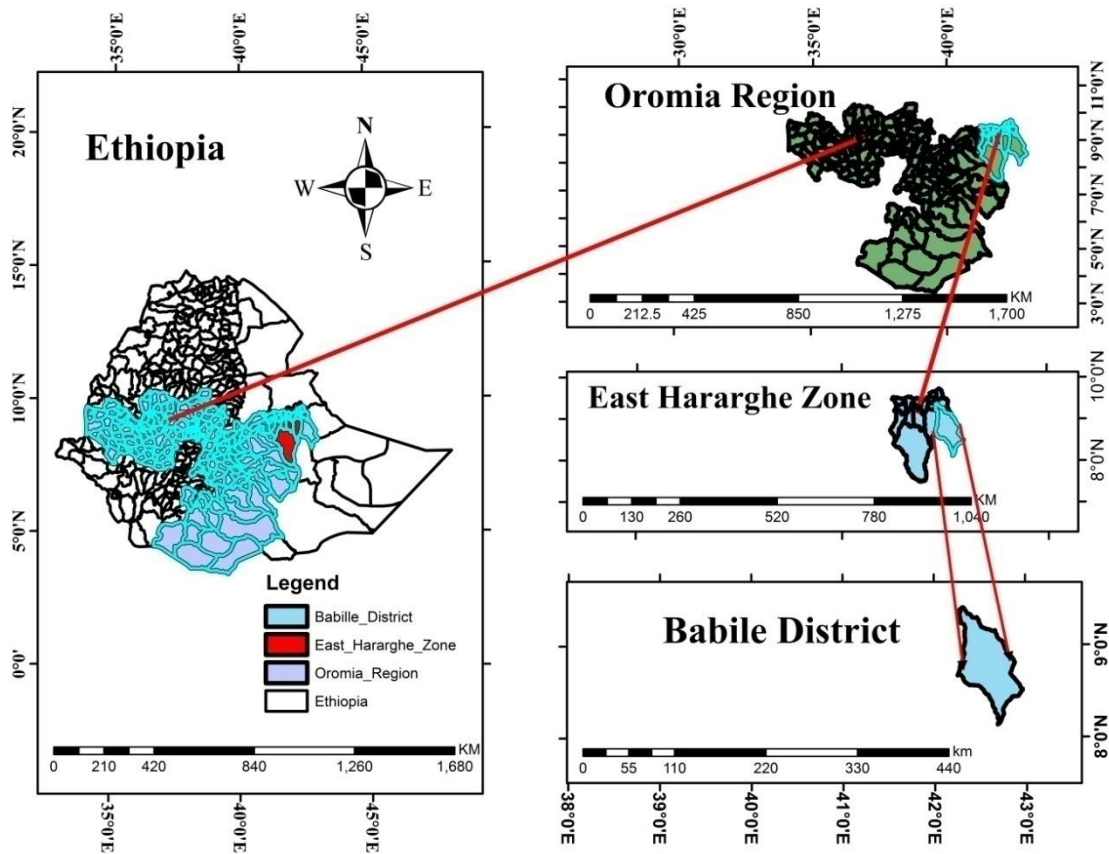
Due to its deep roots, Rhodes grass can withstand long dry periods (over 6 months) and up to 15 days of flooding (Cook *et al.*, 2005; FAO, 2014). Seasonal water logging over 30 cm kills the plant (FAO, 2014). Rhodes grass grows on a wide range of soils from poor sandy soils to heavy clayey alkaline 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; Ecocrop, 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). The feed shortage mostly happens in dry seasons of the year (Ibrahim and Olaloku, 2000). 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

Field experiment was conducted at Bishan Babile and Ifa Kebeles of Babile district of East Hararghe during 2018 and 2019 main cropping season. Babile district is found in the Oromia Regional State of Ethiopia (Figure 1). Babile district is located at 7°00' 90" N Latitude and 43°0' 00" E Longitude (MoA, 2014). Babile district shares boundary with Somali regional state and Fedis district of East Hararghe Zone of Oromia regional State (Figure 1). The total size of Babile district is estimated about 1,325 km² which was divided into 17 major Kebeles and 42 sub-Kebeles. The annual average temperature was 26.5 Celsius with uneven distribution rainfall (MoA, 2014). The people are dominantly agro-pastoral with 70% of the total population living on this mode of life. The remaining population, 15% are agricultural, 10% are pastoral, and 5% petty traders. Maize and sorghum are staple crops, and the livestock production includes cattle, goats, sheep, and camels (MoA, 2014).



Figures 1: Map of the study area

2.2. Experimental Materials

Five Rhodes grass cultivars (ILRI-7384, CV-massaba-13329, ILRI-6633, DZ-253) were collected from ILRI (International Livestock Research Institute)

Bishoftu sub-branch while the local check was the formerly adapted one at Fedis research center.

2.3. Field Preparation and Sowing

The experimental land preparation was done by oxen driven local plough followed by manual ploughing implement (Akafa). The seeds were sown immediately after the beginning of the main rainy season. Sowing was done manually by drilling the seeds in the furrows at a depth of 1-2 centimeters. Then the seed was covered with thin soil by over passing light sticks and fingers over the furrows.

2.4. Treatments and Experimental Design

Two and four farmers were selected from Efa and Bishan Babile Kebeles of Babile district respectively. Five Rhodes grass cultivars (ILRI-7384, CV-massaba-13329, ILRI-6633, DZ-253 and 1 local check) were arranged in a Randomized Complete Block Design (RCBD) with three replications. The gross plot comprised of seven rows of 2 m length ($7 \times 0.25 \text{ m} \times 2 \text{ m} = 7 \text{ m}^2$). A path between plots and blocks were 1 m and 1.5 m respectively. The seed rate was kept at 10-12 kg/haby considering the germination percentage of the experimental seeds.

2.5. Data Collection

Primarily days of emergence, days to 50% flowering, a day to full maturity and plant height was recorded. Height of ten plants from each plot was taken randomly and their mean value was calculated accordingly. 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). Fresh samples of 500gm were collected and submitted for dry matter weight analysis. Finally, the samples were oven dried at 105°C for 24 hours and weighed to determine the dry matter yield of herbage.

2.6. Data analysis

Data on statistical parameters of Rhodes grass samples were subjected to ANOVA based on the model designed for a randomised complete block design (RCBD) according to Gomez and Gomez (1984) and using the computer software package of SAS version 9.1. Mean separations were tested using the least significance difference (LSD) and significant level was considered at ($P < 0.05$).

3. Results and Discussion

3.1. Days to Emergence and Days to 50% Flowering

The over year statistical data analysis result indicates that there was a significant difference ($P < 0.05$) among Rhodes grass cultivars on days to emergence and days to 50% flowering. The mean number of days required for emergence was 10.25, 9.08, 10.33, 10.42 and 8.83 for ILRI-7384, CV-massaba-13329, ILRI-6633 and DZ-253 and local check respectively (Table 1). In this study Rhodes grass cultivars required a range of 8.83 to 10.42 days for first emergence which is almost similar with the study conducted at Deghabour district of Ethiopian Somali region (Mohamed and Gebeyew, 2018) which was recorded 10.66 days. But, other study discovered that Rhodes grass germinates within 7 days after planting (Cook *et al.*, 2005). The alteration on days to emergence might be attributed to soil moisture content, soil fertility and other environmental factors.

The mean numbers of days to 50% flowering were 108.833, 108.92, 109.83, 110.67 and 109.33 for ILRI-7384, CV-massaba-13329, ILRI-6633, DZ-253 and local check respectively. The difference in days to emergence and days to 50% flowering among treatments could be attributed to genetic variation among cultivars and their interaction with the environment.

3.2. Days to Maturity and Plant Height

ANOVA table result for days to full maturity and plant height reveals that there is no significant difference ($P > 0.05$) observed among the mean numbers of days to full maturity for all Rhodes grass cultivars; though, the mean numbers of days to full maturity and plant height were numerically different. The mean numbers of days to full maturity were 154.5, 155.5, 155.3, 151.83 and 151.73 for ILRI-7384, CV-massaba-13329, ILRI-6633, DZ-253 and local check respectively (Table 1). But, numerically, this study revealed that CV-massaba-13329 and ILRI-6633 required more days to full maturity than other cultivars.

The mean plant height values of Rhodes grass cultivars were 145.92, 144.83, 136.08, 147.0 and 145.92 centimeters for ILRI-7384, CV-massaba-13329, ILRI-6633, DZ-253 and local check respectively (Table 1). Numerically, this study revealed that DZ-253 recorded higher plant height values than other cultivars. The average values of Rhodes grass plant height cultivars recorded in this study was almost similar with

the study conducted at Deghabour district of Ethiopian Somali region (Mohamed and Gebeyew, 2018) which was 139.10cm. But, a 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. On the other hand FAO (2009) discovered that Rhodes grass grows up to the height of 90 cm.

3.3. Dry Matter Yield

The aboveground dry matter biomass yield data analysis indicates that, there was a significant difference ($P < 0.05$) among Rhodes grasscultivars. The highest mean aboveground dry matter biomass yield (9.42 t/ha) was recorded from DZ-253 which was followed by local check (8.5 t/ha) (Table 1).Growth performance of Rhodes grass varies with type of cultivar, age of plant and other environmental factors (FAO, 2009). Rhodes grass productivity generally ranges from 7 to 25 tons of DM/ha per year, depending on variety, soil fertility, environmental conditions and cutting frequency (Cook *et al.*, 2005; CASCAPE, 2015; HARC, 2004). The average aboveground dry matter biomass yield values of Rhodes grass cultivars recorded in this study were almost similar with other studies conducted by different authors. 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). Additionally, a study conducted at Adami Tulu Agricultural Research Center (ATARC) and NegeleArsi Farmers Training Center (FTC) also indicated that the average herbage dry biomass yield of Rhodes grass varies from 7.8 to 9.16 tons DM/ha per year without manure application (Tesfaye *et al.*, 2020) which is almost similar with the results of this study.



Figures 2: Showed the performance of Rhodes grass cultivar.

3.4. Seed Yield

Analysis of variance showed that seed yield was significantly ($P < 0.05$) influenced by Rhodes grass cultivars. The highest Rhodes grass seed yield was obtained from DZ-253 (291.3 kg/ha) and local check (280.6 kg/ha) respectively (Table 1). Low seed yield was recorded as compared to a study conducted by other scholars lower than other (Tesfaye *et al.*, 2020; Dawit *et al.*, 2020) at Adami Tulu and Arsi Negele were 370.55 and 313.88 Kg/ha respectively. This variation in seed yield productivity might be due to 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: The over year and over location yield and yield component mean values of Rhodes grass cultivars at Babile district.

Treatments	DE	DF50%F	DFM	PH (cm)	DBMY (t/ha)	Seed yield (Kg/ha)
ILRI-7384	10.25 ^a	108.833 ^b	154.5	145.92	7.79 ^b	265.3 ^c
CV-massaba-13329	9.08 ^b	108.92 ^b	155.5	144.83	7.53 ^b	252.0 ^d
ILRI-6633	10.33 ^a	109.83 ^{ab}	155.33	136.08	7.95 ^b	275.7 ^{bc}
DZ -253	10.42 ^a	110.67 ^a	151.83	147	9.42 ^a	291.3 ^a
Local Check	8.83 ^b	109.33 ^{ab}	151.73	145.92	8.50 ^{ab}	280.6 ^{ab}
Mean	9.78	109.4	154.8	144.13	8.45	273
CV	2.46	0.97	1.5	5.02	12.03	2.08
LSD (0.05)	0.194	0.714	NS	NS	0.415	4.63

^{a,b,c} = Mean within a column with different superscripts differ significantly ($P < 0.05$), DE= days to emergence, DF%F = days to 50% flowering, DFM = days to full maturity, PH = plant height in centimeters, DMY = Dry Matter Yield, t/ha = Ton per Hectare, kg/ha = Kilogram per Hectare, CV = Coefficient of Variation, LSD = Least Significance Difference, NS= Not Significant.

4. Conclusion and Recommendation

The feed shortage mostly happens in dry seasons of the year in Eastern Ethiopia. To overcome the shortage of animal feed in Eastern Ethiopia the study was conducted to select/recommend adaptable and high biomass yielding Rhodes grass cultivars for the study area and other areas having similar agro-ecologies. The results of this study revealed that the highest herbage dry matter yields were recorded from DZ-253 and local check cultivars. From this study, it can be concluded that DZ-253 and local check are recommended for use in Babile district and other districts having similar agro-ecologies in Eastern Ethiopia. However, further study will be needed how to increase the productivity of Rhodes grass using different forage production strategies.

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Adaptation trial of Cassava (*Manihote sculenta* Crantz) in Semi-arid Environments of Borana zone

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Abstract

The study was conducted in Yabello Pastoral and Dryland Agriculture Research Center at the beginning of the long rainfall season, with the objective of evaluating adaptation performances and yield of below and aboveground biomass of cassava varieties. 25-30 cm cuttings of four varieties of cassava plant (Kello, Qulle, Hawassa-4, and Chichu) were collected mainly from Oromia and SNNP regions based on their potential. These 25-30 cm cuttings were planted with 1m×1m intra- and inter-row spacing in Randomized Complete Block Design (RCBD) with four rows; five plants per row and twenty plants per plot were planted. The agronomic parameters were analyzed using SAS software and the mean differences were compared using LSD at $p \leq 0.05$. Among all varieties, Chichu, Kello and Hawassa-4 scored larger leaves dry matter yield of 0.039, 0.037, and 0.032 tons/ha, respectively. In stem dry matter yield, Chichu (0.25 t/ha) had the largest score, followed by Qulle (0.15 t/ha) and Hawassa-4 (0.14 t/ha), respectively while Kello had least score (0.073 t/ha). Chichu, Hawassa-4 and Kello varieties scored larger tuber dry matter yields of 0.323, 0.287, and 0.264 tons/ha, respectively. Generally, in both above and below ground dry matter yield, Chichu, Hawassa-4 and Qulle had had larger values than Kello and well performed in all agronomic parameters in the semi-arid of Borana rangeland and hence we recommend them in semi-arid environments of Borana zone for both food and feed production purpose.

Key: = Biomass yield, Cassava varieties, Without rainfall, Agronomic parameters

1. Introduction

Cassava (*Manihote sculenta* Crantz) is one of the staple food crops for millions of people who live in the world (Tewodros, 2012). It is a tropical root crop and has been grown exclusively for tuber production, mainly for human consumption (Ruiz et al 1981). Its leaves also contain a high crude protein (CP) content of 25.5-29.8% of dry matter (DM). Furthermore, it is particularly important for food security since many tropical areas often experience unfavorable environmental conditions (Nweke and Enete, 1999). It is originated in tropical Latin America (Tewodros, 2012) and was first introduced to Ethiopia by the British (Tewodros et al., 2012). After the introduction, the crop has been found to have an excellent adaptation and growth performance in different agro-ecologies with productivity variation (Amsalu, 2003) which is by far greater than the global average tuber yield of 46t/ha (Edosa, 1996). Its use as a potential food crop in Ethiopia has increased during and after the 1984 famine (Onwueme and Charles, 1994; Amsalu, 2003). Cassava is one of the most drought-tolerant crops and can be successfully grown on marginal soils, giving

reasonable yields where many other crops cannot do well (Edwards, 1991). Cassava is propagated exclusively from cuttings.

Cassava is also used as livestock feed, and regularly fed to sheep and goats on small-scale subsistence farms in Africa (Wanapat and Petlum, 2001). Availability of animal feed is one of the greatest constraints to the expansion of the livestock industry in developing countries. This plant is well known for its adaptability to poor soil conditions, drought resistance and pest tolerance. It is a vitally important feed resource, which is abundantly available in the tropical countries. Cassava can be cultivated for root production, human food and energy source for animals. Recently, managing cassava for foliage production has been found to have more potential as it is a high rumen by-pass protein source for ruminants and thus can improve production and reduce feed costs. Wanapat et al (1997) firstly reported the potential of cassava foliage made into hay (cassava hay), which combined leaves, stems and petiole, as a feed for ruminants. Alternatively, by harvesting the whole upper green part from cassava foliage at the early growth stage (3-4 months) and thereafter every 2-3 months for subsequent harvests have been demonstrated and established for making hay by Wanapat et al (1997). There is a great deal of current interest in supplementing the feeding of animals with cassava in Africa. Traditionally, cassava roots are processed by various methods into numerous products and utilized in various ways according to local customs and preferences. However, there is limited information on cassava in terms of quality and feeding value for livestock species in the Borana zone though cassava offers tremendous potentials as a cheap source of energy feed for animals, provided it is well balanced with other nutrients. Therefore, the objectives of this study was to evaluate the adaptation potential and yield of cassava varieties and to explore the potential use of cassava as an important livestock feed resource in semi-arid environments of Borana zone.

2. Materials and Methods

2.1. Description of the study area

The study was conducted in the Yabello district of the Borana zone. Yabello town, the capital of the Borana zone, is located 565km south of Addis Ababa. The Borana rangelands are found in the south most of Ethiopia. They are located at 4-6°N and 36-42°E sloping gently from 1600m.a.s.l in the North East to about 1000m.a.s.l in the extreme South that borders Northern Kenya and about 1780m.a.s.l in the central vicinity. The Borana rangelands are occupied almost entirely by pastoral populations, resource use is largely communal, though with crop cultivation and private enclosures that appear to be increasing in recent decades. Rainfall is bimodal with the long rains accounting for 60% of the total rainfall occurring between March and May and the short rains comprising 27% of the total rainfall occurring between September and November. Spatial and temporal variability in both the quantity and distribution of rainfall renders the area Semi-arid, with an average annual rainfall varying from 353 to about 900mm per annum (McCarthy, Kamara and Kirk, 2002). A cool dry season occurs from June to August, while a warm dry season occurs from December to February. The average temperatures vary from an annual minimum of 13.1°C to an annual maximum of 25.2°C.

2.2. Site selection and land preparation

The specific site was selected with the participation of local elders, experts from each district's PDOs (Pastoral development offices) and community leader. The site was fenced with local materials firmly to

exclude livestock interference. Further, a guard was assigned for the protection of the study site. Before the collection of the plant, the site and seed-beds were prepared. Unwanted plant materials were removed from the study site and some soil amendments were conducted accordingly.

2.3. Experimental materials and design

The cuttings of 25-30cm of the four cassava varieties (Kello, Qulle, Hawassa-4, and Chichu) were collected from Oromia and SNNP regions based on their potential. The experiment was laid in RCBD (randomized complete block design) with four replications in 2m spacing between blocks and followed all agronomic practices as needed during the growing period. Plantation was done in rows per plot at a spacing of 1m between plants and 1m between rows.

2.4. Planting and data collection

Planting of all plots was done in the main rainy season (Mid-March) in a furrowed seed beds. Weeding was performed when the cassava plants are 20 to 25 cm tall, that is, four weeks after planting. The second time weeding was done one or two months after the first weeding. All the data were collected 18 months after planting as had been suggested by (Tewodros and Biruk, 2012). Accordingly, data on plant height (cm), number of branch/plant, average canopy diameter/plant (m), average stem girth (cm), average number of roots/plant, average length of roots/plant (cm), average diameter of roots/plant (cm), root fresh weight (kg/plot), above-ground biomass (kg/plant) and root dry weight (kg/plot) were recorded. Three plants (large, medium and small) cassava were taken purposively from the plot and were floured to get the dry matter yield of the product. After measuring the yield, the amounts were converted into tons per hectare.

2.5. Data analysis

The data obtained were analyzed using One way Analysis of Variance (ANOVA) in a Completely Randomized Block Design. Analysis of variance for all parameters was performed using the general linear model's procedure of SAS (2002). Differences between means were compared using the least significance difference (LSD) at a 0.05 level of significance.

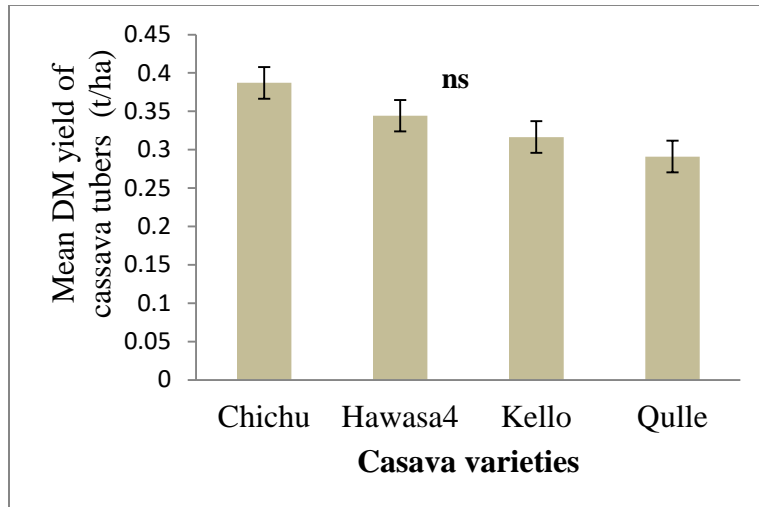
$$Y_{ij} = \mu + \beta_i + \gamma_j + e_{ij}$$

When: Y_{ij} = observations, μ = Overall mean, β_i = Block effects, γ_j = variety effects, and e_{ij} = Experimental error effect

3. Results and Discussion

3.1. Tuber Dry Matter Yields of Cassava varieties

All cassava varieties were not significantly different ($P > 0.05$) in their tuber dry matter (Fig.1). Among the Cassava varieties, Qulle scored the least tuber dry matter (DM) yield (0.243 tons/ha) while Chichu, Hawassa-4 and Kello scored larger tuber dry matter yield of 0.323, 0.287, and 0.264 tons/ha, respectively. The mean DM yield of cassava varieties were different from each other because the sizes/length of tubers were different between cassava varieties; for instance, Chichu variety had larger size of tubers than other cassava varieties.

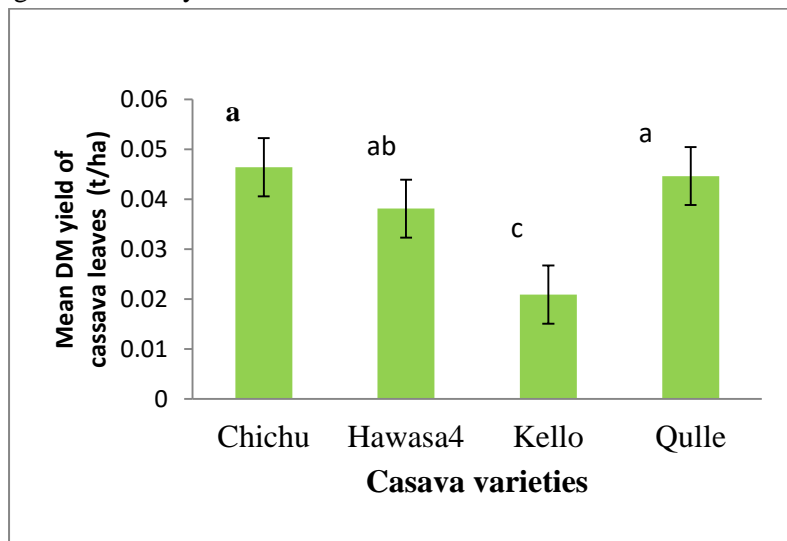


¹ns = non-significant difference at $P > 0.05$

Figure 1: Mean of DM yield of cassava tubers in tons per hectare (t/ha)

3.2. Leaves Dry Matter Yields

Kello variety was significantly different ($P < 0.05$) in leaves dry matter from Chichu, Hawassa-4 and Kello varieties while there was no significant difference ($P > 0.05$) in leaves dry matter among Chichu, Hawassa-4 and Kello cassava varieties (Fig.2). Among the Cassava varieties, Kello scored the least leaves dry matter yield (0.017 tons/ha) while Chichu, Kello and Hawassa-4 scored larger leaves dry matter yield of 0.039, 0.037, and 0.032 tons/ha, respectively. The mean values of leaves DM yield among cassava varieties were different from each other because the branch numbers of cassava varieties were different between varieties; e.g. Kello variety had lower numbers of branches than other cassava varieties.

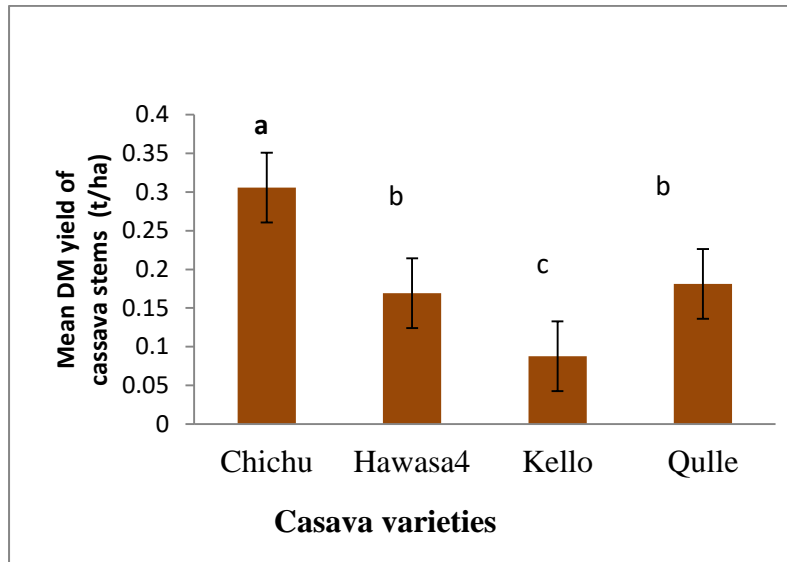


¹ Bars of with different letters are significant different at $P < 0.05$

Figure 2: Mean leaves DM yield in t/ha

3.3. Stem Dry Matter Yields of cassava varieties

Among cassava varieties, there were significant differences ($P < 0.05$) in the dry matter of stems. Chichu and Kello varieties were significantly different ($P < 0.05$) in stems dry matter yield from other varieties while there was no significant difference ($P > 0.05$) in leaves dry matter between Hawassa-4 and Qulle varieties (Fig.3). Among the Cassava varieties, Chichu scored the largest stem dry matter yield (0.25 tons/ha) followed by Qulle and Hawassa-4 with 0.15 and 0.14 t/ha, respectively, while Kello was least in stem dry matter yield, 0.073 tons/ha. These mean values of stem DM yield of cassava varieties were different values due to difference in the circumferences, height and branch numbers of stems. For instance, of all cassava varieties, Chichu had higher stem circumferences and branch numbers than other cassava varieties.

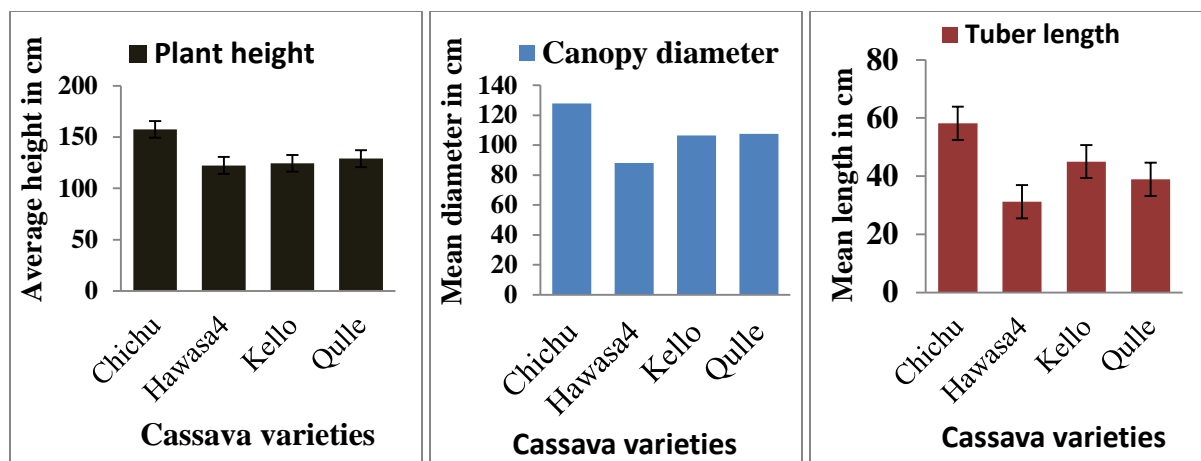


¹ Bar of different letters are significantly different at $P < 0.05$

Figure 3:- Mean stems DM yield in t/ha

3.4. Plant height, tuber length, and Canopy diameter

Plant height of cassava varieties ranged from 157-122 cm. Chichu scored the largest plant height of 58.2 cm, followed by Kello, 45 cm; but Hawassa-4 had the least plant height, 31.2 cm. Their tuber length ranged from 58.2-31.2 cm. Chichu scored the largest tuber length of 58.2 cm, followed by Kello (45 cm); but Hawassa-4 had the least tuber length, 31.2 cm. The canopy diameter of cassava varieties was ranged from 128-88 cm. Among the Cassava varieties, Chichu scored the largest canopy diameter of 127.83 cm, followed by Qulle and Kello with 107.42 and 106.42 cm, respectively, while Hawassa-4 had the least in canopy diameter, 88.13 cm. Generally, Chichu showed the highest value in canopy diameter, tuber length, plant height than Qulle and Kello though Hawassa-4 was the lowest.



¹ Bar with different letters are significantly different at $P < 0.05$

Figure 4: Mean value of Canopy diameter, Plant height, and tuber length of cassava varieties

3.5. Plant Leafiness

Leafiness is one criterion for a plant to be eligible for forage because the leaf part of forage species determines the yield of forage biomass. Based on this fact leafiness was inculcated in the data to be collected and out of four cassava varieties 80%, 10%, 10% and 0% were excellent, good, fair and poor, respectively (Fig. 5).

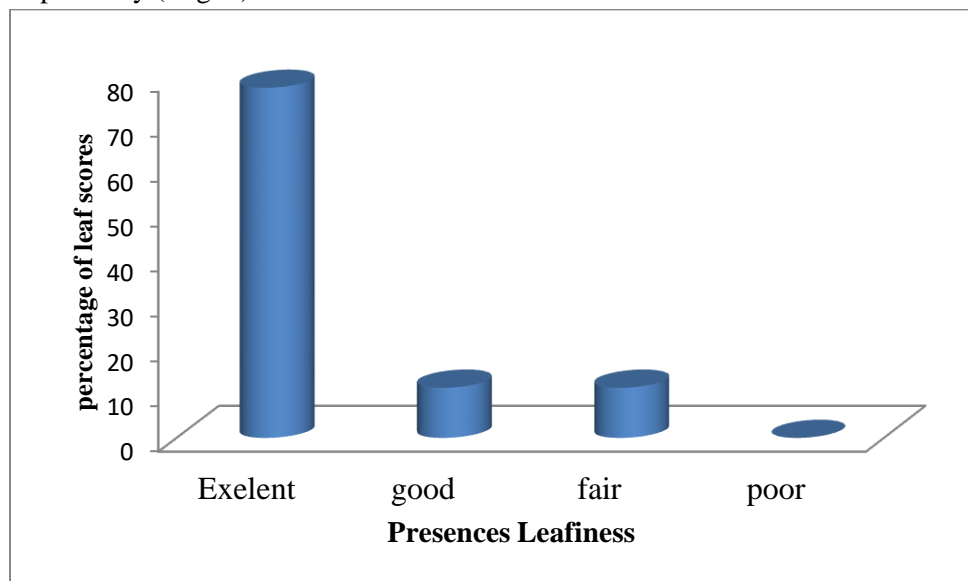


Figure 5: percentage of cassava varieties leafiness scores

4.6. Plant vigorousness

In forage production, the persistence of vigorousness is another criterion that would be considered as an indicator of good forage species because it indicates that the strength of the plant to tolerate and persist in the given environmental calamities. Taking this into consideration, from the cassava species trial 88.89%, 11.11%, 0% and 0% were excellent, good, fair and poor, respectively (see Fig.6). All dual purpose

cassava species had an excellent vigorous trait that could be taken as the potential for advanced screening in any semi-arid of Borana zone with only rainfall availed in the areas.

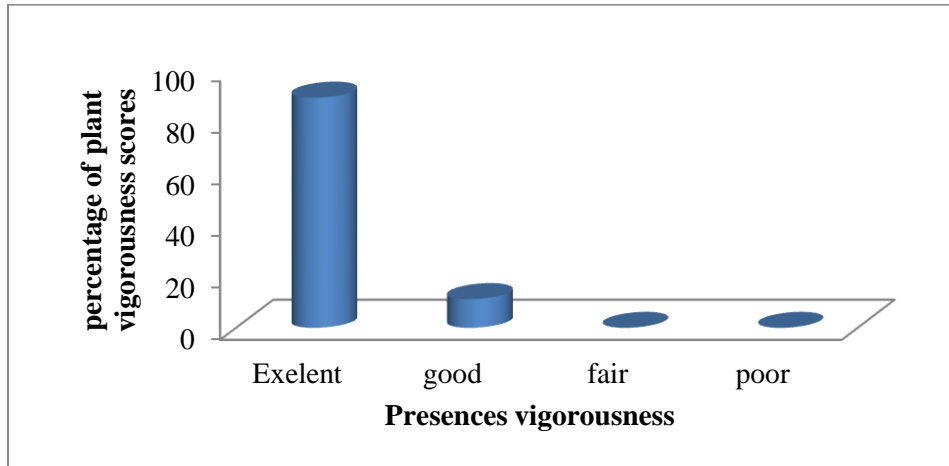


Figure 6:- percentage of cassava varieties vigorousness scores

4.7. Resistance to pest and diseases

The data showed that 66.67 %, 22.22 %, 11.11 %, 0% and 0% of the cassava varieities had no infestation, very low infestation, low degree of infestation, moderate infestation, high degree of infestation andvery high degree of infestation. During data collection, some worms were observed attacking leaves of cassava but not with much damage to the leaf part of cassava varieties.All cassava varietiesdidnotlose their greenness and dried due to attack by diseases or warms.

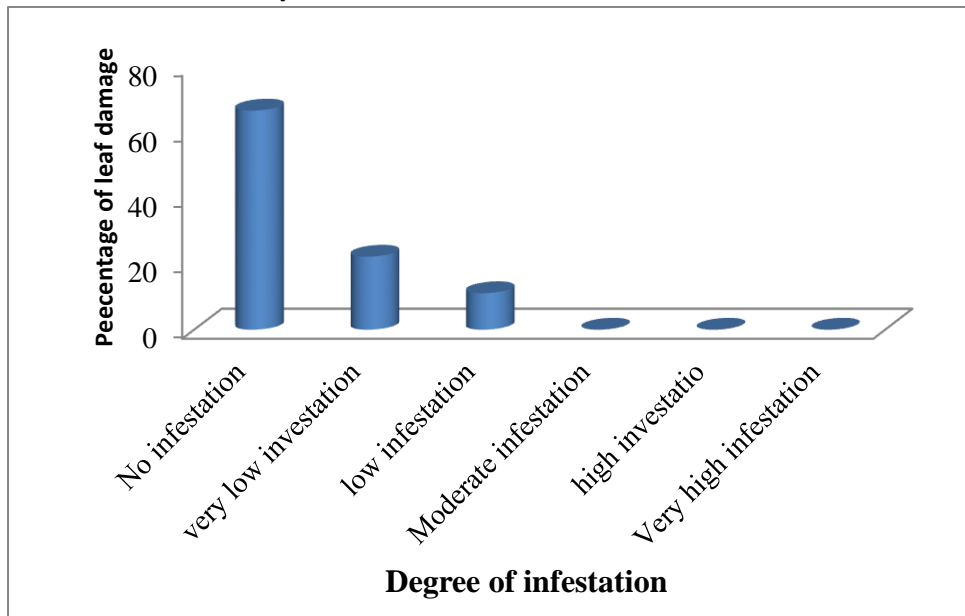


Figure 7:- percentage of cassava varieties leaf damage scores

4. Conclusions and recommendation

The four varieties of the cassava plants (Kello, Qulle, Hawassa-4, and Chichu) were adapted in semi-arid environments of the Borana zone without irrigation as water supplement. All the dual purpose of cassava varieties, Chichu, Hawassa, Qulle and Kello, had performed excellent in vigorosity but they had different in their dry matter yield. Therefore, Chichu varieties could be taken as potential materials for advance screening in any semi-arid areas in Borana zone where rainfall is the only source of water for plant growth. Because among these varieties, Chichu has the highest in total (above and belowground) dry matter yield, followed by Hawassa-4 and Kullo varieties. This cassava varieties that have good forage indicators should be scaled-up and developed in pastoralist and agro-pastoralist areas because its multipurpose use and high preference after use by livestock and human consumption under arid and semi-arid conditions in the study area especially during the dry season has significant contribution to food security. Therefore, the study varieties were good performance when rainfall is good especially variety Chichi showed that better performance in above and below ground biomass yield per hectare.

5. Acknowledgments

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Evaluating Rehabilitation of Degraded Rangeland through Cattle Night Penning in Borana Zone, Southern Ethiopia

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Abstract

The study was conducted in Yabello Pastoral and Dryland Agriculture Research Center ranch towards the end of the dry season, with the objective of determining the effectiveness of rehabilitating the moderately degraded rangeland by cattle night penning in Borana rangelands. A 10m x 5m area was fenced off with local materials for a number of livestock per plot that was calculated based on 1.6 m² per unit of cattle. The experimental design was RCBD in three replications with five treatments, namely, T₁ = moderately degraded rangeland + night penning for 4 days, T₂ = moderately degraded rangeland + night penning for 6 days, T₃ = moderately degraded rangeland + night penning for 8 days, T₄ = moderately degraded rangeland + night penning for 10 days and T₅ = control (exclosure). Then five sub-quadrants with 0.5 m x 0.5 m size were placed in four corners and a center of 10 m x 5 m plots to determine the herbaceous composition and soil properties. The collected data were analyzed using one-way ANOVA for significance test at an alpha level of 0.05.

As the result indicated cattle night pen is an effective mechanism for improving herbaceous botanical composition of desirable perennial and annual species than control plots (exclosure). In this study, the proportion of basal and soil erosion and compaction were significantly different ($P < 0.05$) between cattle night pen treatments and the control. Also, grass and forbs dry matter yields were significantly different ($P < 0.05$) among cattle night pen treatments and the control (exclosure).

Key: cattle night pen, grass, non-grass, dry matter yield, soil properties

1. Introduction

Degradation of pastoral rangelands has been associated with restricted livestock mobility, poor grazing management practices resulting in overgrazing (Li et al., 2011). This has serious negative consequences for pastoral livestock production, wildlife conservation and pastoral livelihoods. African pastoral rangelands are some of the most degraded rangelands in the world (Ritchie et al., 2012). The ongoing degradation of African pastoral lands has been largely associated with inappropriate grazing practices (Maraseni et al., 2008). Consequently, restoration and improvement of the productivity of these rangelands require innovative approaches such as planned grazing, rest rotation and bunched herding, which has been suggested as a tool for restoring and enhancing rangeland health (Ritchie et al., 2012). The concentrated cattle trampling, dunging and urination have maintained the grasslands for millions of years by breaking soil crusts, planting seeds, mulching the soil surface with trampled vegetation and

fertilizing the landscape. Findings of Brunbjerg et al., (2015) and Boakye et al., (2013) attested that the effects of cattle trampling induced an increment in plant species richness and number of annual plants species and interaction with fire is often creating a mosaic of succession communities and increases spatial heterogeneity in the rangeland due to cattle manure make the rangeland more productive than would be the case in their absence through enhancing soil fertility. The proponents of holistic cattle trampling management argue that when animals are concentrated in small areas for short periods, they break the ground, allowing water and nutrient flow, whilst sowing seeds and adding fertilizer through dung and urine (Strauch, 2009). In the management of rangeland, inorganic fertilization is used as one option. In organic fertilization in a large area or extensive farming requires a high cost to the organic fertilizer. So, where ample manure is available, organic fertilization is effective for range rehabilitation. This manure application may be practiced by over-sowing or reseeding activities, in which it requires high labor and cost for extensive farming.

In the study area, the rangeland is characterized by low productivity, soil erosion, and low fertility, reduced soil water availability for plant growth (Adumasi et al., 2010). Conversely, there is no research that has been conducted to evaluate the effect of holistic rangeland management as rangeland productivity improvement interventions. Therefore, it is imperative to evaluate the effect of holistic rangeland management for restoring degraded rangelands.

Objective

- To determine the effectiveness of rehabilitating the moderately degraded rangeland by cattle night penning

2. Material and methods

2.1. Selection of Experimental Site and Design

The study was conducted at the Yabello district of Borana zone. The experiment was conducted in YPDAR Constation rangeland experimental site. A three 750 m² moderately degraded rangeland area was selected from the YPDARC. The uniformity and condition of the rangeland to be selected were considered. The sites were fenced by participating pastoral communities through active participation of district administrative, development actors and researchers. The selected rangeland was divided into plots of 10m x 5m area and fenced off with local materials. Each plot of degraded rangeland received treatments randomly and the control plot of degraded rangeland was exclosure. The experimental design was RCBD with five treatments and three replications. All treatments were conducted during mid-long rainy season. After the application of treatments, the experimental sites were protected until data collection. The number of livestock per plot was calculated based on 1.6 m² per unit of cattle.

Experimental Treatments

Treatment (T₁) = moderately degraded rangeland + night penning for 4 days

Treatment (T₂) = moderately degraded rangeland + night penning for 6 days

Treatment (T₃) = moderately degraded rangeland + night penning for 8 days

Treatment (T₄) = moderately degraded rangeland + night penning for 10 days

Treatment (T₅) = control (Exclosure)

2.2. Data Collection

Species composition: At each of the experimental site, the herbaceous species composition was assessed by harvesting three to five quadrates of 1 m x 1 m (depending on the homogeneity and heterogeneity of the range sites) randomly by throwing the quadrant each time towards the back and the herbaceous vegetation within the quadrant was cut at ground level using hand shears. The identification of the species composition was undertaken at two levels. Plant species were identified very easily in the field by using guidebooks (Reinhard and Admasu, 1994). But those plants which could not be identified in the field, a herbarium sample of each species were pressed using plant presser, labeled, and then sent to the National herbarium at Addis Ababa University for identification.

Herbaceous basal cover: For the herbaceous basal cover, 0 to 10 points were considered. A representative sample area of 1m x 1m was selected for detailed assessment and three to five measurements were taken at each sample site. The square meter was divided into eight equal parts and all basal covers of plant species in the quadrant were drawn to one of the eighths to facilitate visual estimations. Only basal covers of live plant species were considered. The rating for basal cover within the same square meter was given the maximum score (10 points) and the minimum score when the basal cover was less than 3%.

Herbaceous biomass production: The herbaceous biomass yield data such as fresh biomass yield (FBY) and dry matter yield (DMY) were collected from each experimental plot at age of 50% heading. Three samples were randomly taken per each plot from quadrat plot of 0.50m x 0.50 m area using a sickle and transported to Yabello Pastoral and Dryland Agriculture Research Center. The representative samples were oven-dried at 105°C for 24hrs at Yabello Pastoral and Dryland Agriculture Research Center's laboratory. Then the dry matter yield per each experimental treatment was calculated by the final weight collected from oven-dried and was divided by initial weight before subjecting to the oven-dried.

Soil composition: Soil samples were collected from three quadrants along the transects at 0-15 cm soil layers, which were air-dried and sieved through 2 mm and 210 µm meshes and then the composite samples were made. Soil organic matter was determined through wet oxidation (Nelson and Sommers, 1982). Total N was determined by the Kjeldahl method (Steven-son, 1982). Available P was extracted with 0.5N NaHCO₃ at pH 8.5 (Alifragis, 2010). Soil pH was determined by using a glass electrode. Soil bulk density and soil water content were also determined following standard procedures.

2.3. Statistical Analysis

Effects of cattle trampling on herbaceous biomass, grass basal cover, herbaceous species and soil composition were analyzed by one-way ANOVA; significant differences for all statistical tests were evaluated at the level of $P \leq 0.05$. Means were separated using least significant difference (LSD) test with the following model

$$Y_{ij} = A + \beta_i + t_j + e_{ij}$$

Where: Y_{ij} = dry matter yield, A = General mean of the treatments, β_i = block effects (location),
 t_j = treatment effects and e_{ij} = Random error

3. Result and Discussion

3.1. Effect of cattle night penning on the herbaceous botanical composition of degraded rangeland

Rangeland is composed of grasses, legumes, sedges, and other heterogeneous plants in various families, which could be herbaceous or woody forms (McIllroy 1972). Herbaceous species of native to rangeland that have been identified at the experimental site are presented in Table 1. Nine types of grass and thirteen non-grass herbaceous species belonging to different families were identified in all the plots. The productive perennial grass species identified were *Panicum maximum*, *Cynodon dactylon*, *Cenchrus ciliaris*, *Eleusine intermedia* and *Panicum maximum* while the palatable forb species that were identified include *Commelina africana*, *Pollichia mpestris*, and *Barleria spinisepala*.

Most herbaceous species recovery identified in this study had similarities with previous reports on rangeland species composition in the semi-arid of Borana rangeland, indicating that the cattle night pen affects recovery of species composition of degraded rangeland. The different species composition of rangeland is classified based on their distribution as decreaser and increaser attributes in terms of pasture quality, quantity and persistence. Hence, the presence of more desirable perennial and annual herbaceous species in the treatment of night pen plots than control plots would indicate the cattle night penning has a higher degree of recovering degraded rangeland than usual traditional enclosure rangeland management.

Table 1. Effect of cattle night penning on the herbaceous botanical composition of degraded rangeland

Scientific name	Vernacular name	Growth form	Life form	Treatments				
				Four day-night pen	Six day-night pen	eight day-night pen	Ten day-night pen	Control
<i>Cenchrus ciliaris</i>	Mata guddeessa	G	P	2	5	0	0	0
<i>Oxygonum sinuatum</i>	Mogooree	F	A	0	2	2	0	0
<i>Abutilon hirtum</i>	Gurbiidaalati	F	P	0	0	0	5	0
<i>Aristida kenyensis</i>	Biilaa	G	A	0	40	11	0	0
<i>Barleria spinisepala</i>	Qilxiphee	F	P	0	7	1	2	2
<i>Bercharia species</i>		G	A	2	0	6	1	0
<i>Chenopodium opulifolium</i>	Ononuu	F	A	0	0	0	8	4
<i>Chlorophytum gallabatense</i>	Miirtuu	F	A	0	0	3	0	0
<i>Commelina africana</i>	Qaayyoo	F	A	0	1	4	1	5
<i>Cynodon dactylon</i>	Sardoo	G	P	21	11	15	5	0
<i>Cyperus bulbosus</i>	Saattuarbaa	G	A	8	17	0	0	0
<i>Cyperus species</i>	Saattuu	F	A	0	0	1	37	0
<i>Eleusine intermedia</i>	Coqorsa	G	P	4	1	1	0	0
<i>Endostemon kelleri</i>	Urgoo	F	P	6	0	2	7	25
<i>Eragrostis cilianensis</i>	Ardaa	G	P	31	0	0	0	0
<i>Justicia odora</i>	Agaggaroo harree	F	P	0	0	1	0	1
<i>Panicum maximum</i>	Loloqaa	G	P	23	5	0	0	0
<i>Plectranthus caninus</i>	Harc'a'a	F	A	0	0	6	0	0
<i>Pollichia campestris</i>	Guungumakorbe essaa	F	P	0	1	1	0	1
<i>Sporobolus pellucidus</i>	Salaqoo	G	A	2	2	1	0	0
<i>Tagetes minuta L.</i>	Suunkii	F	A	0	7	41	32	62
<i>Volkensinia prostrate</i>	Gurbii	F	P	0	1	6	1	0
				100	100	100	100	100

¹G= Grass, F= Forbs, P= Perennial, A= Annual, Control or enclosure

3.2: Effect of cattle night penning on herbaceous species biomass yield of degraded rangeland

Total dry matter yields of herbaceous species were significantly different among the treatments and higher results were obtained from treatment with eight-day cattle night penning, followed by ten-day cattle night penning and least for the control (exclosure). However, a non-significant result was obtained for the composition of grasses between all treatments and the control. Unlike the grass species composition, the species composition for non-grass species was found to be significantly different among all the treatments, with higher for 10 day cattle night penning treatment because it obtains high amount of cattle dung and urine due to relatively elongated night penning days. All the treatments highly generated the growth of non-grasses than grass species because forbs/non-grass herbaceous species respond less to N than grasses as grass dominated pastures give greater responses to N (Steele 2008). The percentage increase in the proportion of non-grass reflects the role of cattle dung and urine influencing the first rehabilitation of degraded rangelands of herbaceous botanical composition in succession progress.

Generally, there was higher herbaceous botanical composition across all the treatment plots than the control or exclosure rangeland management system. This may indicate that limited cattle dung and urine that serves as sources of organic matter and total nitrogen had a direct suppressing effect on the growth of herbaceous botanical composition of degraded rangeland. Night pen treatment areas also easily recovered through initiating germination of soil seed bank and above seed of herbaceous species because degraded rangeland was rested due to livestock has not interested to allow for grazing.

The highest result of herbaceous composition recovery was observed in the cattle night penning plots. This is justified in Whiteman (1980) as the application of nitrogen fertilizers to grass-legume pastures has dramatic effects on the legume component by altering the botanical composition. The presence of high levels of nitrate or ammonium will inhibit nodulation and reduces the rate of nitrogen fixation that leads to a reduction in legume content. Similarly, Miles and Manson (2000) explained that when legumes are growing with grasses, the grasses are strong competitors for available nitrogen, and take up most of that applied.

Table 2. Effect of cattle night penning on herbaceous species biomass yield of degraded rangeland

Treatments	DMY (ton/ha)	Grasses	Non-grass
Four day cattle night penning	0.56 ^{cd}	0.19 ^a	0.36 ^c
Six day cattle night penning	0.70 ^{bc}	0.20 ^a	0.50 ^{bc}
Eight day cattle night penning	1.09 ^a	0.23 ^a	0.86 ^{ab}
Ten day cattle night penning	1.0 ^{2ab}	0.09 ^a	0.92 ^a
Control (Exclosure)	0.32 ^d	0.10 ^a	0.22 ^c
P-value	0.0002	0.45	0.0.001

¹abc Means in the same column without a common letter are different; DMY- Dry Matter Yield

3.3: Effect of cattle night penning on the basal cover, soil erosion and compaction of degraded rangeland

The result revealed that all treatments of cattle night penning plots were significantly different ($P < 0.05$) from control plots in the ground cover of herbaceous species but not between the cattle night penning treatment plots (Table 2). Percentage ground covers of herbaceous plant species of cattle night penning plots were higher than control (exclosure).

Table 3 showed that the effect of cattle night penning is an important factor that had an impact on herbaceous species recovery of above-ground biomass of degraded rangeland. The result revealed that all treatment of cattle night penning plots were significantly different ($P < 0.05$) within control plots in soil erosion but not between the cattle night penning treatment plots (Table 3). Percentage soil erosion of cattle night penning plots was lower than enclosure because of higher recovery of basal cover in cattle night pen plots than the enclosure.

The result revealed that all treatments of cattle night penning plots were significantly different ($P < 0.05$) within control plots in soil compaction and also between the cattle night penning treatment plots (Table 3). Percentage soil compaction of cattle night penning plots was lower than usual rangeland management system of the traditional enclosure but compaction increased as increased cattle night penning days. Therefore, eight-day cattle night penning was better to avoid soil compaction of degraded rangeland.

Table3:- Effect of cattle night penning on the basal cover, soil erosion and compaction of degraded rangeland

Treatments	Basal cover (%)	Soil erosion (%)	Soil compaction (%)
Four day cattle night penning	52.22 ^a	1.11 ^b	15.56 ^b
Six day cattle night penning	51.33 ^a	0.00 ^b	3.67 ^c
Eight day cattle night penning	60.56 ^a	2.56 ^b	4.11 ^c
Ten day cattle night penning	54.44 ^{2a}	0.00 ^b	7.78 ^{bc}
Control (Exclosure)	21.22 ^b	15.56 ^a	39.22 ^a
P-value	0.0005	0.002	0.0001

¹*a,b,c Means in the same column without a common letter are different; basal cover, soil erosion and compaction*

3.4: Effect of cattle night penning on soil organic matter, organic carbon and total nitrogen of degraded rangeland

The result of cattle night penning on organic matter, organic carbon and total nitrogen of degraded rangeland rehabilitation of the study sites are indicated in table 4. Rehabilitation of degraded rangeland through cattle night pen treatment has a great impact on soil organic matter. Soil organic matter and carbon were significantly different ($P > 0.05$) between treatments and the control. Ten and eight-day cattle night pen treatments were higher in soil organic matter and carbon than the six and four day cattle night penning and the control plots as cattle night penning increase soil organic matter in the degraded areas due to the increased level of dung. The Statistical analysis indicated that the soil total nitrogen was significantly different ($P > 0.05$) between treatment and the control because as cattle night penning increase the cattle urine frequency increased which can increase soil nitrogen of degraded areas. Ten-day cattle night penning has high soil total nitrogen than other treatment and followed by eight-day cattle night penning treatment while least for only enclosure plot. Whereas, the percentage of soil organic matter and carbon and total nitrogen of large value of cattle night pen was lower than the standard requirement of forage production i.e. medium average content of soil in their soil organic matter and carbon and total nitrogen respectively (Frank, 1990). This is due to effect of overgrazing and climatic factors (drought), the plant material that services for increasing soil decomposition was lower on degraded areas of rangeland types. The result agreed with Dormaar et al. (2009), Kumasi et al. (2010) and Bagheri et al. (2009) that concluded the removal of vegetation by herbivores reduces ground covers and

soil organic matters and nitrogen. Tessema et al. (2011) also reported that the concentration of total nitrogen was higher in light grazing as compared with sites exposed to heavy grazing. The interaction effect between districts and rangeland types was significantly different ($P < 0.05$); in soil organic carbon (OC %) due to soil type different between districts but not significantly different ($p > 0.05$) between district and depth, rangeland types and depth, and district, rangeland types and depth in soil percentage of OC, OM and TN.

Table 4: Effect of cattle night penning on soil organic matter, organic carbon and total nitrogen of degraded rangeland

Treatments	%OC	% OM	% TN	A [^] P
Four day cattle night penning	3.4 ^{bc}	5.8 ^{bc}	0.14 ^{cd}	31.7 ^{ab}
Six day cattle night penning	3.9 ^{ab}	0.7 ^{ab}	0.19 ^{bc}	49.3 ^{ab}
Eight day cattle night penning	4.1 ^a	7.0 ^a	0.21 ^{ab}	44.9 ^{ab}
Ten day cattle night penning	4.2 ^a	7.2 ^a	0.24 ^a	54.3 ^a
Control (exclosure)	3.0 ^c	5.2 ^c	0.12 ^d	13.9 ^b
P-value	0.004	0.004	0.002	0.15

¹*a,b,c Means in the same column without a common letter are different; %OC = percentage of organic carbon, %OM = percentage of organic matter, %TN = percentage of total nitrogen and AP = available phosphorous*

4. Conclusion and recommendation

Generally, there was higher herbaceous botanical composition and dry matter yield across all the treatment plots than the usual exclosure rangeland management system. Also the proportion of soil erosion and compaction of cattle night penning treatment plots was lower than control/exclosure because of higher recovery of basal cover in cattle night pen plots than the exclosure. The percentage of soil carbon, organic matter and total nitrogen were higher in the treatment plots than the control plot. Confining animals for a short period in the night pen might disturb soil compaction by their hoof and provide spaces for water and nutrient flow. This may indicate that cattle dung and urine had a direct suppressing effect on the growth of herbaceous botanical composition and soil properties of degraded rangeland. Therefore, moderate cattle night penning i.e. eight-day cattle night pen is better for rapidly recovering degraded rangeland with reducing soil compaction effect.

5. Acknowledgments

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Effect of seeding ratio and time of planting of cowpea (*Vigna unguiculata*) intercropping with maize (*Zea mays*) on agronomic parameters, forage biomass and grain yield of maize

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Abstract

The study was conducted at Adami Tulu and Dugda districts of Oromia regional state, Ethiopia to determine the optimum level of seeding ratio and planting time of cowpea under maize for optimum forage biomass production and maize grain yield. Combinations of four levels of cowpea seeding ratios and four different cowpea planting dates were laid out in a randomized complete block design in factorial arrangement with three replications. The levels of seeding ratios were 100%, 75%, 50%, 25%, 0% (sole maize) for the two districts. The four planting dates for cowpea were simultaneously planting with maize, 10 days after maize planting (DAMP), 20 DAMP and 30 DAMP. The results indicated that increasing seeding ratio of cowpea from 25% to the highest level (100%) resulted in significantly increased cowpea forage biomass yield. Time of cowpea planting in maize also influenced the plant height and biomass yield of cowpea. The highest forage biomass yield was recorded from simultaneously planting of the two crops. On the other hand, seeding ratio of cowpea has significantly influenced the grain yield of maize. It was also indicated that the time of cowpea planting in maize have significantly affected the grain yield of maize with simultaneously planting resulting in the lowest grain yield. The total LER in most of the intercropping system was more than one showing that intercropping of forage legumes with maize is more advantageous than sole cropping of maize. The optimum forage legume biomass yield (1.78 t/ha) was obtained from the combination of seeding ratio of 75% with 10 DAMP without significantly ($p > 0.05$) reducing the grain yield of maize. Hence this combination was recommended for production of cowpea forage and maize grain from intercropping of the two crops in the study areas. From these results, it can be concluded that additional forage can be produced by intercropping cowpea with maize at their appropriate seeding ratio and planting time with a little or no sacrifice in maize grain yield. Moreover, it is important to further demonstrate and promote the recommended maize-cowpea intercropping practices for the end users of the study areas and similar agro-ecologies.

Keywords: Biomass yield, cowpea, intercropping, planting date, seeding ratio

Introduction

Livestock production system is one of the main agricultural activities in the mid rift valley of Oromia regional state, Ethiopia. Even though the rift valley has a great livestock potential, the production and productivity of the livestock is very low mainly due to the shortage of feed resources. Feed shortage in terms of both quantity and quality is the major constraint to livestock production and productivity especially during the dry season (Ahmed *et al.*, 2010, Ulfina *et al.* 2013, Gelayenew *et al.* 2016). Feed

supply from natural pasture fluctuates following seasonal dynamics of rainfall (Solomon *et al.*, 2008). Grazing as a source of livestock feed has begun to decline in recent years, as a result of increased areas of cultivation, and changing patterns of land use. Despite, these problems, ruminants continue to depend primarily on forages from natural pastures and crop residues. So, an adequate supply of livestock feed both in quantity and quality is crucial to the livelihoods of millions of people across the developing world, and not just for smallholders, but also for pastoralists and the large number of landless who depend mainly on common land for grazing (Sanford and Ashly, 2008).

Maize (*Zea mays*), is one of the important dual purpose crop used in human diet and animal feed. It has the potential to supply large amounts of energy-rich forage for animal diets, and its fodder can safely be fed at all stages of growth without any danger (Dahmardeh *et al.*, 2010). Cowpea is also among the legumes adapted in the mid and lowland agro ecologies (Ayana *et al.*, 2013). It is considered as relatively tolerant to drought because of its tendency to form a deep taproot (Gomez, 2004). Cowpea is mainly used for grain and animal feed because of their high feed potential for dry matter and quality (Bilatu *et al.*, 2012, Alemu *et al.* 2016). Cowpea provides nutritious grain and an inexpensive source of protein for both rural poor and urban consumers. Its grain contains about 25% protein and 64% carbohydrate (Quin, 1997) and therefore has a tremendous potential to contribute to the alleviation of malnutrition among resource-poor farmers. It also provides high quality legume hay for livestock. In addition, cowpea contributes to the sustainability of cropping systems and soil fertility improvements in marginal lands by providing ground cover and plant residues, fixing nitrogen, and suppressing weeds. Moreover, the acceptance of such a dual purpose crop by small holder farmers is very high.

Intercropping is a means of reducing household risks during poor growing seasons and producing modest surpluses during favorable seasons (Woomer *et al.*, 1997). The yields of intercropping are often higher than in sole cropping systems (Lithourgidis *et al.*, 2006). The reasons are mainly that resources such as water, light and nutrients can be utilized more effectively than in the respective sole cropping systems (Liu *et al.*, 2006). Cereal-legume intercropping is important in subsistence farming communities as a means of improving soil fertility and increasing land use intensity in situations of limited land availability (Saidi *et al.*, 2010). Different studies indicated that forage legumes integration through intercropping did not have a significant effect on maize grain and biomass yield (Mergia Abera, 2014). The shade tolerance characteristic of cowpea makes it compatible as an intercrop with maize, millet and sorghum (Singh and Emechebe 1998).

Studies conducted at Adami Tulu Agricultural Research Center indicated that, for areas where the rainfall situation is erratic and irregular, the dual purpose cowpea is an appropriate crop especially if it is cultivated as a mixture with cereals (Ayana *et al.*, 2013). In addition to sole cropping of cowpea, farmers with limited crop land prefer the intercropping systems (Singh *et al.*, 2003). Most studies indicated that forage legumes did not appear to reduce cereal yield when intercropped (Dahmardeh *et al.*, 2010, Sarkodie-Addo and Abdul-Rahaman, 2012, Hamid *et al.*, 2014). However, due to high competition and shading effect, intercropping may result in decrease in yield of one or both of the individual crops in the mixture unless appropriate seed ratios and planting time is followed.

Therefore, to get the optimum benefit from intercropping; seed rate, planting time and other agronomic practices need to be adjusted depending upon the purpose and growing conditions. Regardless of the importance of dual purpose cowpea as food-feed value, there is lack of adequate information with regard to optimum seeding ratio and appropriate planting time to produce reasonable grain and forage yields from the crops in mixture. Hence, the objective of this study was to identify the optimum level of seeding ratio and appropriate time of cowpea planting to produce a reasonable amount of maize grain and forage biomass yields from the mixture.

Materials and Methods

Study site

The study was conducted for two consecutive years (2018/19-2019/20) at Adami Tulu Agricultural Research Center and Farmers Training Center (FTC) of Dugda district. These sites represent the lowland agroecology. Adami Tulu and Dugda districts are located in the mid rift valley of Oromia, south of Addis Ababa on Addis Ababa-Hawasa road. They are found at an altitude of 1650 meters above sea level (m.a.s.l). The average annual rainfall of the areas is about 727.1 mm, whereas their average annual minimum and maximum temperatures are 11.8°C and 28.3°C, respectively (ATARC metrology data, 2015-2017).

Crop management

Maize crop varieties of BH 540 which is inappropriate for lowland altitude and is currently under production in the study areas was used for this study. With regard to cow pea, an adapted dual purpose variety known as Black eye bean was used for the intercropping. Maize was planted in rows at distance of 75cm and 25cm between rows and between plants, respectively with a seed rate of 25 kg/ha. Initially 2-3 seeds of maize were planted per hole. Twenty five days after sowing, seedlings of maize were thinned to retain one healthy seedling per hole. Germination test was done for cowpea before sowing. Cowpea was intercropped with maize at different time of planting and seed rates. DAP fertilizer at a rate of 100kg/ha was applied to all plots before sowing while 100kg/ha of UREA was applied to all plots except the sole cowpea treatment.

Experimental design

Combinations of four levels of cowpea seeding ratio and four different times of cowpea planting dates arranged factorially with three replications were laid out in a Randomized Complete Block Design. The levels of seeding ratios were 100%, 75%, 50%, 25% and 0% (sole maize) while the planting dates of cowpea include simultaneously planting with maize, 10 DAMP, 20 DAMP and 30 DAMP. The seeding ratio of cowpea was calculated on the basis of the recommended sole seeding rates (40kg/ha) for sole cowpea production. The plot size of 3.75m x 3.5m and spacing between the plots and replications of 0.5 and 1m respectively were used. All other cultural practices including thinning and weeding were kept at normal and uniform for all the treatments.

The intercropping advantage was assessed by calculating the land equivalent ratio (LER), an index of intercropping advantage, and a reflection of the degree of inter-specific competition or facilitation in an

intercropping system. Partial LER was used to compare in between individual LERs (LER_f and LER_m), which indicated competitive effects as proposed by Mead and Willey (1980).

$$\text{LER} = (\text{LER maize} + \text{LER cowpea}),$$
$$\text{LER maize} = (\text{YMM} / \text{YM}) \text{ and } \text{LER cowpea} = \text{YCM} / \text{YC},$$

Where; YMM= Yield per unit area of maize in mixture, YCM = Yield per unit area of cowpea in mixture
YM and YC are the yields of maize and cowpea as sole crops, respectively,

Data collection and analysis

All relevant agronomic and yield data including plant height of cow pea and maize, stover (stalks) biomass yield, biomass yield of cowpea, grain yield of maize were collected. Three plants for each crops were randomly selected from each plot and plant height was measured from the base of the plant to the flag leaf. The mean plant height was then calculated. The cowpea biomass yield was estimated by harvesting the plant at 50% flowering stage. Plants in the middle row of the plots were harvested and weighed immediately to obtain fresh yield. The fresh sample was taken and dried to constant weight using forced air-drying oven to determine the dry matter yield using the methods described by AOAC (2000). Maize grain yield was determined by taking a sample from one row of each plot when the plants were fully matured and dried. After the maize is harvested, sample of stover was taken from one row and oven dried to a constant weight and dry biomass yield (t/ha) was recorded. The collected data were organized and subjected to analysis of variance using the SAS statistical procedures. Means were separated using least significant difference (LSD) at 5% significant level.

Results and Discussions

Analysis of variance for the effects of seeding ratio and time of cowpea planting under maize crop are presented in table 1. Cowpea plant height was significantly affected ($p < 0.01$) by time of planting and the interaction of seeding ratio and time of planting ($p < 0.05$). However, the plant height was not influenced ($p > 0.05$) by seeding ratio. The highest plant height of cowpea (94.01 cm) was recorded from simultaneously planting of maize and cowpea while the shortest plant height (53.2 cm) was recorded from cowpea sown at 30 DAMP (Table 2). A decreasing trend of plant height was observed as time of cowpea planting under maize elapsed. Considering the combination of both factors, it was observed that simultaneously planting of cowpea and maize for all cowpea seeding ratios have produced significantly the longest cowpea plants followed by the 10 DAMP. The shortest plant was observed from seeding ratio of 100% with 30 DAMP (Table 3). These findings suggest that plant heights of cowpea are depressed by the higher shade effect experienced when established at 30 DAMP.

Cowpea forage biomass yield was significantly affected by application of different levels of seeding ratio ($p < 0.001$) and time of cowpea planting ($p < 0.001$) and their interaction ($p < 0.05$). The maximum biomass yield (1.48 t/ha) of cowpea was recorded from the highest seeding ratio (40 kg/ha) while the least biomass yield (0.74 t/ha) was obtained from the lowest seeding ratio of cowpea. This may be because, the higher plant populations achieved earlier canopy closure and intercepted more light than the lower plant populations resulting in the lower dry matter accumulation and hence the least biomass yield of

cowpea. The effect of time of cowpea planting indicated that the significantly highest biomass yield (2.64t/ha) was obtained from simultaneously planting of cowpea and maize while the lowest biomass yield (0.12t/ha) was recorded from 30 DAMP. The biomass yield showed a declining trend as the time of planting increased from simultaneous planting to 30 DAMP. This could be due to the higher maize canopy at 30 DAMP which diminished the amount of light penetration. Interaction of the two factors, seeding ratio and time of under sowing of cowpea in maize, resulted in maximum biomass yield of 3.18t/ha from the combination of seed ratio of 100% and simultaneous planting of the two crops. It was followed by 2.83t/ha and 2.71 t/ha for 75% seed ratio and simultaneously planting and 50% seed ratio and simultaneously planting, respectively. The least biomass value (0.029t/ha) was recorded from the combination of seed ratio of 25% and 30 DAMP.

The lowest agronomic and forage biomass performances of cowpea planted at 30 DAMP could be mainly because of the shading effect of maize canopy that blocked the transmission of light to the undersown cowpea. The intercropping would be more productive if the effect of maize shade could be reduced. The manipulation of time of sowing cowpea under maize crop is one of the potential ways of reducing the negative effect of the shade of maize on cowpea. Studies also confirmed that shading effect is one of the reason for the low productivity of cowpea in maize-cowpea intercropping systems (Ewansiha *et al.*, 2015). It was also reported that higher shade affected the growth of cowpea during the critical stages of growth leading to smaller plants with fewer branches which contributed to the low biomass yield (Terao *et al.*, 1997).

Both seeding ratio and time of cowpea planting and the interaction of seeding ratio and time of planting didn't have a significant effect ($P>0.05$) on maize plant height and maize stover yield. Maize grain yield was significantly affected by seeding ratio ($p<0.05$), time of cowpea planting ($P<0.01$) and the interaction effect of seeding ratio and time of cowpea planting ($P<0.05$). The maximum grain yield (60.07t/ha) was recorded from sole maize planting while the lowest yield (50.14t/ha) was obtained from 100% seed ratio of cowpea. As compared to sole maize planting, the mean grain yields of 10.8, 9.1, 11.9 and 15.69 %, respectively were decreased due to 25, 50, 75 and 100% of cowpea seed ratio planted under maize. Generally, the results showed that high seeding ratio of cowpea in maize crop resulted in maize grain yield reduction. This could be due to the densities of cowpea at higher seeding ratio which reduces the sunlight and competes for nutrient that can be utilized by maize crop. Maize grain yield increased with increase in time of cowpea undersowing. Planting cowpea at 30 DAMP produced the highest (57.72qu/ha) maize grain yield while simultaneously planting of maize and cowpea resulted in the lowest (49.02qu/ha) maize grain yield. The reduced grain yield from simultaneously planting of cowpea with maize could probably due to the fact that the competition effect of cowpea for soil nutrient and sun light. The interaction effect of seed ratio (50%) and time of cowpea planting (30 DAMP) resulted in the highest value (60.21qu/ha) of maize grain yield. The lowest value of maize grain yield (43.1qu/ha) was obtained from the combination of cowpea seeding ratio of 100% and simultaneously planting of maize and cowpea. It was also showed that simultaneously planting of cowpea in maize crop had a negative effect on maize grain yield as compared to the delay planting. The result indicated that the optimum forage legume biomass yield (1.55 t/ha) and (1.78) were obtained from the combination of seeding ratio of 50% with 10 DAMP and 75% with 10 DAMP, respectively without significantly ($p>0.05$) reducing the grain yield of maize. Generally, the result showed that planting cowpea under maize crop with seeding ratios of

50% and 75% at 10 DAMP resulted, respectively in forage biomass yield of 1.55t/ha and 1.78t/ha and maize grain yield of 54.5ku/ha and 53.68qu/ha without significantly reducing the grain yield of maize.

On the other hands, the result indicated that significantly higher partial LER of cowpea (0.69) was obtained from 100% seeding ratio with simultaneously planting while the maximum total LER (2.11) was obtained from 100% seeding ratio with simultaneously planting (Table 4). However, the total LER of most of the treatment combinations did not differ significantly except for treatments with seeding ratio of 25, 75 and 100% planted 10 DAMP combinations. The variations observed in LER could be ascribed to the difference in the amount of dry biomass produced from cowpea and maize stover. The total LER in all cases was more than one, showing that intercropping of the forage legumes with maize is more advantageous than sole cropping of maize. Therefore, the total LER indicated that intercropping of maize and cowpea was productive and had a yield advantage over the sole maize cropping. According to Workayehu (2014), when $LER < 1$ there is obvious disadvantage of the intercropping and the available resources are used more efficiently by the sole cropping than the intercropping. Similar results were reported for different proportion of plant mixtures (Mucheru-Muna *et al* 2010, Lithourgidis *et al* 2006). Land equivalent ratio greater than unity has also reported in sorghum/lablab intercropping (Ibrahim *et al.*, 1993). From these results, it can be concluded that additional forage can be produced by intercropping cowpea legume at their appropriate seeding ratio and planting time with a little or no sacrifice in maize grain yield.

Table 1. Mean square values of agronomic and yield components of cowpea and maize in response to different level of seeding rates and time of cowpea planting under maize crop at lowland sites

Source of variation	Df	Cowpea plant height (cm)	Cowpea biomass yield (t/ha)	Maize plant height (cm)	Maize grain yield (qu/ha)	Maize stover yield (t/ha)
Seeding rate	3	91.74ns	2.29***	90.04ns	67.29ns	6.48ns
Time of cowpea planting	3	8079.82**	29.07***	26.68ns	554.83**	5.39ns
S*T	9	34.99*	0.29*	135.56ns	8.27*	2.22ns
Error	80	68.27	0.36	251.6	113.1	11.26

Table 2. Effect of seeding ratio and time of planting on agronomic and yield performance of cowpea and maize at lowland sites

Treatments	Cowpea plant height (cm)	Cowpea biomass yield (t/ha)	Maize plant height (cm)	Maize grain Yield (qu/ha)	Maize Stover yield (t/ha)
25	77.87	0.74 ^b	185.35	53.55 ^{ab}	7.29
50	79.58	1.22 ^a	182.92	54.59 ^{ab}	7.17
75	78.67	1.22 ^a	183.33	52.87 ^{ab}	7.23
100	79.13	1.48 ^a	180.63	50.14 ^b	6.34
Sole maize(0)	-	-	184.88	60.07 ^a	7.08
LSD (0.05%)	NS	0.34	Ns	8.34	Ns

Time of cowpea planting					
Simultaneously	94.01 ^a	2.64 ^a	182.31	49.02 ^b	6.57
10 DAMP	86.10 ^b	1.31 ^b	183.51	53.00 ^{ab}	6.91
20 DAMP	81.87 ^b	0.58 ^c	182.90	54.18 ^{ab}	7.48
30 DAMP	53.20 ^c	0.12 ^d	184.14	57.72 ^a	7.19
CV	10.3	31.4	8.71	20.69	31.3
LSD (0.05%)	4.76	0.34	Ns	6.8	Ns

¹DAMP=Days After Maize Planted, CV=Coefficient of variation,LSD=Least significant difference, NS= Non significant,

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

Table 3. Interaction effect of seeding ratio and time of planting on agronomic and yield performance of cowpea and maize at lowland sites

Treatment combination		Cowpea plant height (cm)	Cowpea biomass yield (t/ha)	Maize plant height (cm)	Maize grain Yield (qu/ha)	Maize Stover yield (t/ha)
Seeding ratio (%)	Time of cowpea planting					
25	Simultaneously	93.77 ^a	1.86 ^b	181.0	46.3 ^{cd}	5.78
	10 DAMP	86.77 ^{ab}	0.75 ^{cd}	190.6	54.8 ^{abcd}	7.76
	20 DAMP	78.5 ^b	0.32 ^{cde}	179.9	53.6 ^{abcd}	7.50
	30 DAMP	52.44 ^c	0.029 ^e	189.8	59.51 ^{ab}	8.12
50	Simultaneously	94.16 ^a	2.71 ^a	183.5	48.0 ^{cd}	6.60
	10 DAMP	86.16 ^{ab}	1.55 ^b	177.4	57.50 ^{abc}	6.56
	20 DAMP	83.66 ^b	0.56 ^{cde}	184.1	55.63 ^{abcd}	7.57
	30 DAMP	54.33 ^c	0.06 ^e	186.7	60.21 ^a	7.97
75	Simultaneously	93.78 ^a	2.83 ^a	180.5	48.2 ^{bcd}	7.61
	10 DAMP	86.11 ^{ab}	1.78 ^b	184.0	58.8 ^{ab}	6.69
	20 DAMP	80.5 ^b	0.59 ^{cde}	189.6	53.98 ^{abcd}	7.93
	30 DAMP	54.3 ^c	0.13 ^{de}	179.2	56.24 ^{abcd}	6.71
100	Simultaneously	94.3 ^a	3.18 ^a	181.6	44.4 ^d	5.81
	10 DAMP	85.5 ^{ab}	1.61 ^b	182.0	48.02 ^{bcd}	6.64
	20 DAMP	84.8 ^{ab}	0.87 ^c	177.9	53.51 ^{abcd}	6.95
	30 DAMP	51.9 ^c	0.26 ^{cde}	180.9	54.92 ^{abcd}	5.97
Sole maize		-	-	184.9	60.07 ^a	7.08
Mean		78.8	1.16	183.2	53.83	7.01
LSD (0.05%)		9.52	0.68	Ns	11.66	NS
CV		10.52	27.1	8.71	20.74	32.7

¹DAMP=Days After Maize Planted, CV=Coefficient of variation,LSD=Least significant difference, NS= Non significant,

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

Table.4 Partial and total land Equivalent Ratio (LER) of biomass of cowpea and maize at lowland sites.

Treatment combination		Partial Land Equivalent ratio		Total LER
Seeding ratio (%)	Time of cowpea planting	Cowpea	Maize	
25	Simultaneously	0.34 ^{cd}	1.03	1.36 ^{bc}
	10 DAMP	0.23 ^{de}	1.38	1.62 ^{abc}
	20 DAMP	0.08 ^{efgh}	1.37	1.47 ^{abc}
	30 DAMP	0.01 ^h	1.38	1.39 ^{bc}
50	Simultaneously	0.46 ^{bc}	1.31	1.77 ^{abc}
	10 DAMP	0.45 ^{bc}	1.28	1.74 ^{abc}
	20 DAMP	0.16 ^{efg}	1.54	1.71 ^{abc}
	30 DAMP	0.008 ^h	1.58	1.59 ^{abc}
75	Simultaneously	0.50 ^b	1.50	1.99 ^{ab}
	10 DAMP	0.49 ^b	1.62	2.11 ^a
	20 DAMP	0.20 ^{def}	1.46	1.66 ^{abc}
	30 DAMP	0.03 ^{gh}	1.21	1.23 ^c
100	Simultaneously	0.69 ^a	0.99	1.69 ^{abc}
	10 DAMP	0.52 ^b	1.29	1.81 ^{abc}
	20 DAMP	0.32 ^{cd}	1.30	1.62 ^{abc}
	30 DAMP	0.08 ^{fgh}	1.07	1.15 ^c
CV (%)		29.6	28.4	23.9
LSD (0.05)		0.15	Ns	0.67

¹DAMP=Days After Maize Planted, CV=Coefficient of variation,LSD=Least significant difference, NS=Non significant,

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

Conclusions

The result of maize-cowpea intercropping experiment indicated that increasing seeding ratio of cowpea from 25% to the highest level (100%) resulted in significantly increased cowpea forage biomass yield. Time of cowpea undersowing in maize crop also influences the plant height and biomass yield of cowpea with the higher forage biomass yield recorded from simultaneously planting of cowpea and maize crop. Interaction effect of seeding ratio and time of cowpea planting also indicated that the highest seeding ratio of cowpea with simultaneously planting resulted in higher cowpea biomass yield. On the other hands, seeding ratio of cowpea significantly influenced the grain yield of maize with the lowest yield obtained from 100% of cowpea as compared to the sole maize planting. It was also indicated that the time of cowpea undersowing in maize have significantly affected the grain yield of maize with simultaneously planting resulted in the lowest grain yield. Maize grain yield was also influenced by the interaction effect of seeding ratio and time of planting of cowpea. Using the highest seeding ratio of cowpea with simultaneously planting resulted in higher cowpea biomass yield and a reduction in maize grain yield. The biomass yield of cowpea decreased as cowpea seeding ratio declined and the time of cowpea planting

under maize advanced. The time of cowpea planting under maize also influenced the maize grain yield. On the other hand, the total LER in most of the intercropping system was more than one showing that intercropping of the forage legume with maize is more advantageous than sole cropping of maize. From these results, it can be concluded that additional forage can be produced by intercropping cowpea legume at their appropriate seeding ratio and planting time with a little or no sacrifice in maize grain yield. Generally, the optimum forage legume biomass yield (1.78 t/ha) was obtained from the combination of seeding ratio of 75% with 10 DAMP without significantly ($p > 0.05$) reducing the grain yield of maize. Hence these cowpea seeding ratio and time of planting were found as the best combinations for production of cowpea forage biomass and maize grain from the intercropping of the two crops for the study area. Moreover, it is important to further demonstrate and promote the recommended maize-cowpea intercropping practices for the end users of the study areas and similar agro-ecologies.

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Evaluation of Pigeon pea (*Cajanus cajan*) for Agronomic, Performance, Yield and Nutritional Composition at Adola District, East Guji Zone of Oromia.

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Abstract

The study was conducted with the objective to identify adaptable, high biomass, good quality and seed yield of Pigeon pea varieties Adola sub-site of Bore Agricultural research center. Four pigeon pea varieties Tsigab, Degagsa-(11575), Belabas-(16527) and 16555 were tested in randomized complete block design (RCBD) with three replications. Analysis of variance showed significant differences ($P < 0.01$) were observed among the tested treatments in days to 50% emergency, days to 50% flowering, number of branches per plant, pod length, plant height, leaf to stem ratio and biomass yield. . The highest branch number was recorded from Tsigab variety (21.6), whereas the lowest branch number was obtained from Degagsa-(11575) genotype (9.75). The longest pod was recorded from Degagsa-(11575) (5.16 cm) while the shortest pod length was recorded from Belabas-(16527) variety (3.14 cm). The longest plant height was measured from Degagsa-(11575) variety (159.75 cm) followed by 16555 genotype (113 cm) while the shortest plant height was obtained from Belabas-(16527) variety (78.63 cm). The maximum biomass yield was produced from Tsigab variety (2.17 ton/ha) followed by 16555 genotype (1.27 ton/ha) while the lowest biomass yield was obtained from Degagsa-(11575) variety (1 ton/ha). With regards to crude protein (CP) content, Tsigab variety had showed the maximum (30.4%) value while the minimum (23.1%) was observed from genotype 16555. Hence, based biomass yield, seed yield and CP content Tsigas variety was recommended for further promotion in the midland of Guji zone and similar agro- ecologies.

Keywords: *Cajanus cajan*, Biomass yield, Nutritive value, pigeon pea.

1. Introduction

Animal feed is the most important input in livestock production and it is an essential prerequisite for any substantial and sustained expansion in livestock production (Samuel *et al.*, 2008; Lagasse *et al.*, 2010). According to Sefa (2017), animal feeds including; natural pasture, fodder crops, fodder trees, crop residues and non-conventional feeds are used in different parts of Ethiopia. Green fodder (grazing) is the major type of feed (54.59%) followed by crop residues (31.60%), hay (6.81%) and industrial byproducts (1.53%), (CSA, 2017). Alemayhuet *al.*, (2017) reported that feed in terms of both quantity and quality is bottleneck to livestock production in Ethiopia. This problem of feed shortage is more aggravated during the dry season (Zewdie, 2010). Even during years of good rainy season, forage is not sufficient to feed livestock in the highlands (Melese *et al.*, 2014). Data from different parts of the country also indicated that available feed satisfy only about 78.2% of demand (Sefa, 2017)

Pigeon pea (*Cajanus cajan*(L.) is one of the leguminous crops that have been cultivated for human and livestock consumption in many parts of the world. It is a legume plant belonging to the family of “Fabaceae” or “Leguminosae” and widely used as fodder and feed for livestock (Rao *et al.*, 2002). Pigeon pea is one of the most common tropical and subtropical legumes cultivated for its edible seeds. Pigeon pea is fast growing, hardy, widely adaptable, and drought tolerant (Bekele-Tessema, 2007). It can be considered of utmost importance for food security regions places where rain failures are prone to occur (Crop Trust, 2014). At the end of the dry season, pigeon pea provides green forage of outstanding value when other forages have disappeared (Sloan *et al.*, 2009).

Pigeon pea has numerous uses in animal feeding. The leaves and pods are valuable and palatable protein-rich fodder. Leaves are sometimes used to replace alfalfa in ruminant’s diets where alfalfa cannot be grown. Seed processing by-products and sometimes the seeds themselves are used as livestock feed (Pathak *et al.*, 1993). The seeds can be fed to poultry, and mixtures of pigeon pea with maize grain were successful in Hawaii. Bees actively feed on pigeon pea and produce a honey with a distinctive color (greenish) in the comb (Orwaet *et al.*, 2009). Pigeon pea is also a good host for lac insect and silkworms (Cook *et al.*, 2005).

Pigeon pea is a tropical grain legume and is among important pulses grown for food, feed and soil fertility improvement. It is mainly grown in India and in tropical and sub-tropical regions of Africa, Asia and America. It is a cheap source of protein (20%), other soluble vitamins and essential amino acids (Singhet *al.*1990). In Southern and Eastern Africa, pigeon pea has been neglected and very little attention has been put in its research (Damaris, 2007). Farmers in the region still use unimproved late maturing cultivars due to poor access to improved seed (Franklin Simtowe *et al.*, 2011, ICRISAT, 2009). However, there is limited information with respect to pigeon pea adaptation, biomass, seed yield and its nutritional quality in Guji Zone. Therefore, the study was undertaken with the objective to identify and evaluate better adaptable, biomass yield, seed yield and quality of Pigeon pea variety/genotype for the 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 at a distance of 470 km from Addis Ababa and 120 Km from the zonal capital city, NegeleBorena. 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 the district is 1254.56km². 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%). The major soil type of the district is nitosols (red basaltic soils) and orthic Acrosols(Yazachew and Kasahun, 2011).

2.2. Experimental treatments and design

The study was conducted using four varieties/ genotypes including; Tsigab, Degagsa (11575), Belabas- (16527) and 16555. The experiment was conducted in randomized complete block design (RCBD) with three replications. Seeds were sown in rows spaced 0.5 m on a plot size of 4 m x 3 m =12 m². Spacing of 1m between plots and 1m between blocks were used. Seeding rates 30 kg ha⁻¹ was used for all types of

pigeon pea varieties and genotypes. For all treatments, 100 kg ha⁻¹NPS fertilizer rate was uniformly applied at sowing time.

2.3. Data collection

Data on days to 50% flowering, days to seed maturity, plant height, number of branches, pod per plants, pod length, seed per pods, leaf to stem ratio, biomass yield, seed yield and nutritional composition were recorded. Three plants for each crops were randomly selected from each plot and plant height was measured from the base of the plant to the flag leaf and mean plant height was calculated. Forage legume biomass yield was estimated by harvesting forage at 50% flowering stage. Plant in middle row of the plots were harvested and weighed immediately to obtain fresh yield. To determine grain yield, the pods were harvested from the rest rows at optimum physiological maturity by hand picking.

2.4. Chemical analysis

For forage quality analysis, chopped herbage of the three replications were pooled into one and properly homogenized and one representative subsample was taken for each variety/genotypes. The DM and ash percentage were determined by oven drying at 105°C overnight and by igniting in a muffle furnace at 500°C for 6 hours, respectively. Nitrogen (N) content was determined by Kjeldahl method and CP was calculated as N x 6.25 (AOAC, 1995). The neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) fractions were analyzed according to Van Soest and Robertson (1985).

2.5. Statistical analysis

All collected data were analyzed using general linear model procedure SAS (SAS, 2002) version 9.1. Means were separated with 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, μ = overall, mean, A_j = the j th factor effect of treatment, B_i = the i th factor effect of block/ replication, e_{ijk} = the random error.

3. Results and Discussions

Days to Emergence

The analyzed result show that days to 50% emergence was significantly ($P < 0.05$) different between the tested treatments (table 1). The shortest days (9 days) to 50% emergence was recorded from *Tsigab* variety and 16555 genotype. While it takes longer days (9.67 days) for Degagsa-(11575) variety to attain 50% emergence.

Days to Flowering

Significant variations ($P < 0.05$) in days to flowering between the treatments were observed (Table 1). The shortest days to 50% flowering was obtained from *Tsigab* (92.6 days) variety followed by 16555 genotype (109 days), while it takes longer days (124 days) for Degagsa-(11575) variety as compared to others. This might be due to genetic differences of the varieties/genotypes. This result is in line with the previous reports (James AD. 1983; Varshney RK *et al.*, 2009; Wondewosenet *et al.*, 2014).

Days to Maturity

Days to maturity was not significantly ($P > 0.05$) different between treatments. Numerically, the shortest days to maturity was obtained from Belabas-(16527) (162 days) variety followed by 16555 genotype (186 days), while it takes longer days (203 days) to mature for Degas a-(11575) variety as compared to the other treatments'.

Number of branches per plant

Analysis of result showed that number of branch per plant was highly significant differences ($P < 0.01$) among the varieties/ genotypes (Table 1). The highest number of branch was recorded from Tsigab variety (21.6) whereas the lowest branch number was obtained from Degagsa-(11575) (9.75). This result is in conformity with the result of (Ezeaku *et al.*, 2008).

Pods per plant and seeds per pod

The result showed that there was no significant ($p > 0.05$) differences among the treatments for pods per plant and seeds per pod (Table 1). This result was disagreed with the report of (Chattopadhyay and Dhiman, 2006; Dahat *et al.*, 2006).

Pod length

The tested varieties/genotypes were significantly ($P < 0.05$) differ for pod length. The longest pod was recorded from Degagsa-(11575) (5.16 cm) while the shortest pod length was recorded from Belabas-(16527) variety (3.14 cm). This result is in conformity with (Ezeaku *et al.*, 2008).

Plant Height

The plant height was significantly ($P < 0.05$) varied among the treatments. The longest plant height was measured from Dagagsa-(11575) variety (159.75 cm) followed by 16555 genotype (113 cm) whereas the shortest plant height was obtained from Belabas-(16527) (78.63 cm). This result is disagreed with the report of (Denbela *et al.*, 2020) (217.6).

Leaf to Steam ratio

There were significant ($P < 0.05$) different in leaf to steam ratio of the tested treatments. The highest leaf to steam ratio was obtained from Tsigab variety (1.07) followed by Belabas-(16527) (0.71) whereas the lowest leaf to steam ratio was obtained from 16555 genotype (0.67).

Biomass Yield

Significant variations ($P < 0.05$) in biomass yield was observed among the treatments (were observed Table 1). The highest biomass yield was produced from Tsigab variety (2.17 ton/ha) followed by 16555 genotype (1.27 ton/ha) while the lowest biomass yield was obtained from Belabas-(16575) (1 ton/ha).

Seed Yield

Genotypes were showed no significant different ($P > 0.05$) in seed yield performance, but numerically has different values (Table 1). The highest seed yield was recorded from Tsigab variety (29 qt/ha) followed by Degagsa (11575) (23.1 qt/ha), whereas the lowest seed yield was obtained from 16555 genotype (13.2 qt/ha). This result is similar to the report of (Sharma *et al.*, 1981) with the mean yield of 1.37 t/ha.

3.2. Chemical Composition

The chemical composition of the tested pigeon pea varieties/genotypes were indicated in table 2. According to the laboratory result, Belabas-(16527) variety has the highest dry matter (91.7%) while 16555 genotype had the lowest (87.95%) dry matter value. This result is lower than the value (90%) reported by Daniela (*et al*, 2020).

The highest (30.4%) CP was obtained from Tsigab variety while 16555 genotype had the lowest crude protein of (23.1%). On the other hands, genotype 16555 had recorded the highest (68.6%) NDF while Tsigab variety had the lowest (47.3%) NDF value. Among the tested treatments, Tsigab variety had the highest ADF (45.9%) while Degagsa-(11575) had the lowest (37.5%) ADF value. Tsigab also recorded the highest (17.4%) value of ADL while 16555 has the lowest (5.2%) of ADL. The data also showed that Belabas-(16527) variety had the highest (85.1%) OM while 16555 genotype had the lowest (79.2%) OM as compared to other varieties.

Table 1. Combined mean values of different agronomic traits of four pigeon pea cultivars.

Cultivars	DE	DF	DM	Nbpp	Ppp	Pl (cm)	Spp	Ph(cm)	LSR	Bmy (t/ha)	SyY(qt/ha)
<i>Tsigab</i>	9c	92.6c	193	21.6a	55	3.9b	3.5	104.9ab	1.07a	2.17a	29
<i>Dagagsa-11575</i>	9.67a	124a	203	9.75d	89	5.16a	4.5	159.75a	0.69b	1b	23.1
<i>16555</i>	9c	109b	186	12.2c	87	5a	4.8	113ab	0.67b	1.27b	13.2
<i>Belabas-16527</i>	9.6b	114ab	162	13.4b	52.5	3.14b	4.3	78.36b	0.74b	1.078b	13.8
Mean	9	110	185.7	14	68.9	4.2	4.2	112.57	0.7	1.34	19.77
CV	1	9.54	25	0	60.7	19	39.6	37.9	35	31.35	36.2
LSD (5%)	*	*	NS	**	Ns	*	NS	*	*	*	Ns

^{a,b,c}=Mean in a column within the same category having different superscripts differ significantly ($p < 0.05$), DE= Days to emergency, DF=days to flowering, DM= days to maturity, Nbpp= number of branches per plant, Ppp=Pod per plant, Pl=Pod length, Spp=seed per pod, Ph=plant height, LSR=leaf to stem ratio, Bmy= biomass yield, SY=seed yield, Cv=Coefficient of variation, LSD= Least significant difference, *=significant, **=highly significant, Ns= None significant.

Table 3. Mean chemical compositions of four pigeon pea cultivars.

Cultivars	DM (%)	TASH (%)	OM (%)	NDF (%)	ADF (%)	ADL (%)	CP (%)
Belabas(16527)	91.7	6.7	85.1	64.4	40.1	8.5	24.4
Tsigas	89.2	8.4	80.8	47.3	45.9	17.4	30.4
Degagsa(11575)	89.3	9.3	80.1	64.4	37.5	8.1	23.5
16555	87.95	8.9	79.2	68.6	42.9	5.2	23.1

DM=Dry matter, TASH=Total Ash, OM =Organic Matter, NDF = Neutral Detergent Fiber ADF= Acid Detergent Fiber; ADL= Acid Detergent Lignin; CP = Crude Protein

4. Conclusion and recommendations

The result of this study indicated that Tsigas variety showed best performance in most agronomic and yield parameters including biomass yield, leaf to stem ratio and seed yield as compared to the other varieties/genotype. Moreover, Tsigas variety contains the highest crude protein value. Thus, based on its performance, Tsigas variety is recommended for further promotion in the midland of Guji zone and similar agro-ecologies.

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Release and Registration of “Bareda” Oat (*Avena sativa* L.) Variety for Mid and Highland of West Hararghe Zone, Oromia, Ethiopia

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Abstract:

The name ‘Bareda’ is given to the newly released oat (*Avena sativa* L.) variety. This variety was developed by Mechara Agricultural Research Center and was released in 2020. The pedigree name of Bareda was ILRI Acc.5450 and Bareda was evaluated together with ten genotypes including standard checks (Bonsa and SRCPX80Ab2806) for two years (2017 and 2018) in regional variety trial at four testing sites of Mechara, Chiro, Habro and Tulo. The combined analysis of yield data across years and locations showed that, Bareda had mean herbage dry matter yield of 9.25 t/ha) with (24.16 and 15.77)% yield advantages over the corresponding checks Bonsa (7.45t/ha) and SRCPX80Ab2806 (7.99t/ha), respectively. From forage quality parameters, on the other hand, the quality data indicated that Bareda had mean crude protein and IVDMD of 10.33% and 57.75%, respectively, which are higher than the standard checks by 7.2 and 6.9% crude protein over Bonsa and SRCPX80Ab2806, respectively but lower than both standard checks in IVDMD. It was also better resistant to major oat diseases: leaf blight and stem borer which had scored a scale of 2 % and 1 %, respectively. Analysis using GGE bi-plot method had showed that Bareda variety was more stable than the standard check (Bonsa). Therefore, this new variety was recommended for Chiro, Habro, Tulo and Mechara areas and other areas with similar agro-ecologies

Keywords: bi-plot, herbage dry matter yield, Mechara, Oat, Variety

1. Introduction

The success and prosperity of livestock production is determined by adequate, quality and timely availability of feed. The green forages are major and the most economical source to fulfill the dietary needs of livestock. The insufficient fodder supply is characterized as major constrain of low animal performance for milk and meat production (Rana *et al.*, 2014, Ahmad *et al.*, 2014). The national feed resources potential includes natural pasture (fodder, forage), cultivated forage, concentrates, crop residues, stubble grazing, brewery and winery by-products, oilseed by-products, molasses, sugarcane tops, other feed resources, such as foliage and pods, maize or sorghum thinning, cactus pear, etc (Shapiro *et al.*, 2017).

In Ethiopia, there are large potential areas with diverse altitude, climate, soil type and farming systems for the production of diverse forage species seeds (Alemayehu *et al.*, 2016). Among the different forage crops recommended for various agro-ecological zones of Ethiopia, common oats (*Avena sativa* L.) is abundantly grown in the central highlands of Selale and some parts of west Shewa like Meta-Robi and

Galessa areas of Dendiworeda, Arsi, Bale and Gojjam . Production of oats in different parts of the country dates back at least three decades. The study conducted in north Shewa evident that, farmers are highly interested in growing oats for various reasons among which, feed, roof thatching (straw), source of income and its better performance on poor soils without any inputs. Oats on the other hand, it has been well acknowledged for its ability to grow on wider range of soil types and resistance to biotic and abiotic stresses (Gezahagnet *et al.*, 2016).

Although, west Hararghe is one of the most icons of livestock production areas of the county, there is lack of animal feed technologies especially production of improved forage crops and limited information on nutritional quality of existing feed resources due to the remoteness of the area. The zone has a great opportunity for cattle production due to the availability of good fattening weather, good indigenous knowledge of fattening and the popularity of fattened Harar bull in the country

Even though, cattle fattening requires both quality and quantity of feeds. feed shortage accounts for 75.7%, Animal health (4.8%) and feed cost (3%) (Abdi *et al.*, 2013). Previous studies in various parts of West Hararghe zone (Fekedeet *et al.*, 2016; Fikadu and Asfaw, 2017 and Muleta *et al.*, 2017) revealed that the major constraints of cattle keepers are feed shortage which ranked first. The major feed resources of livestock in west Hararghe such as grasses, trees, and shrubs obtained from enclosed forest area and backyard forage production (Fikadu and Asfaw (2017). However, nowadays its share per household gradually decreasing because of increasing human population and expansion of crop production.

Such and other feed resources related problems became initiating forces for the need of improved forage germplasm introduction and evaluation (like *Avena sativa*).) It is favorite feed of animals and its straw is soft and superior to wheat and barley. The oat grain is valuable feed for almost all categories of animals (Zamanet *et al.*, 2006). Oat is a fast growing and produces a significant amount of fresh fodder within short period (55 to 65 days) with adequate nutritional qualities.

Thus, the breeding efforts are mostly focused on yield stabilization, resistance to biotic and abiotic stresses, biochemical structure associated with feed quality. It has to be noted that modern cultivar, mostly of Bareda produced high biomass yield and better feed quality as compared with other genotypes. Therefore, the objective of this study was to evaluate and identify the best performing oat variety for wide production in the tested environments and other areas with similar agro-ecologies.

2. Materials and Methods

Eight oats (*Avena sativa L.*) genotypes including two standard checks (Bonsa and SRCPX80Ab2806) for two years (2016/17 and 2017/18) in regional variety trial at four testing sites of Mechara, Chiro, Habro and Tulo districts of west Hararghe zone was evaluated. The tested genotypes were ILRI (5442, 5450, 5445, 5447, 5457, 5467, 5478, and 7251), bonsa and SRCPX80Ab2806. Out of the eight tested genotypes during RVT trial, two candidate of oat accessions (ILRI Acc. 5442 and Bareda) were promoted to the variety verification trial (VVT) stage and evaluated at multi locations (DaroLebu, Habro, ChiroZuria and Tulo) of West Haraghe zone during 2018/19 cropping season against two best standard checks (Bonsa and Bate). To verify the genotypes, 10 m x 10 m plot size at a total of ten locations on both farmers' field and research station were used.

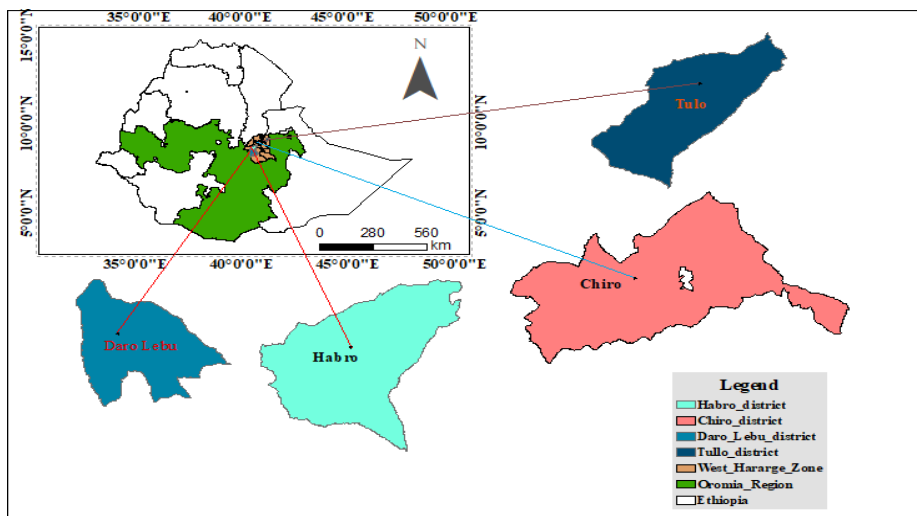


Figure 11 Map of the study area (Source: Ethio-GIS shape file, 2016)

3. Result and Discussion

3.1. Varietal Origin/Pedigree and Evaluation

The pedigree name of the new oat variety namely Bareda was ILRIAcc. 5450. Bareda together with other eight oat genotypes was evaluated against the standard checks (Bonsa and SRCPX80Ab2806) at four testing sites (Mechara, Chiro, Habro and Tulo) districts of west Hararghe zone for two years (2016/17 and 2017/18) in regional variety trial. Bareda was planted both on stations and on farms of all testing sites and were verified in year 2018/19.

The verified variety, Bareda is characterized by growth habit of erect and moderate tuft at basal. Seed color of Bareda is pale brown. Bareda variety on average has a grain yield of 33.4 quintal/ha under research condition and 24 quintal/ha at farmers field. On the average, Bareda needs 72 days to reach 50% of heading/flowering and 126 days to reach seed maturity stage. Bareda variety had plant height on average of 108 cm at physical maturity of harvest. The variety was released for the highlands and mid lands of West Hararghe and performed well within an altitude from 1550-2400 meters above mean sea level.

3.2. Herbage DM yield performances

Bareda produced high mean herbage DM yield of 9.25 t/ha with (24.16 and 15.77) % yield advantage over the standard checks (Bonsa and SRCPX80Ab2806) (7.45 and 7.99) t/ha, respectively (Table 1).

Table 1. Mean Herbage Dry matter yields (t/ha) of different oat genotypes across environments and years

Accession No	2016/17				2017/18					
	Mechara	Chiro	Habro	Tulo	Mechara	Chiro	Habro	Tulo	Comb mean (2017-2018)	Yield Adv %
Bareda (ILRI 5450)	6.29	13.62	7.9	9.94	10.53	9.06	6.06	10.53	9.25	15.77
ILRI 5442	4.82	13.3	8.91	10.41	7.47	11.64	5.52	7.47	8.70	8.89
SRCPX80Ab2806 (check)	5.31	10.91	11.66	9.56	6.52	7.72	5.29	6.94	7.99	
Bonsa (Check)	5.27	7.64	11.63	8.52	6.94	7.94	5.03	6.62	7.45	
<i>ILRI 5457</i>	2.57	15.47	9.41	8.78	6.93	6.67	5.38	6.93	7.77	
<i>ILRI 5467</i>	4.57	11.23	6.69	8.16	6.81	8.23	4.78	6.81	7.16	
<i>ILRI 5478</i>	4.53	7.93	5.98	7.25	5.68	7.94	4.71	5.65	6.21	
<i>ILRI 7251</i>	2.96	10.42	7.82	7.43	6.17	6.44	5.5	6.07	6.61	
<i>ILRI 5445</i>	3.91	12.76	8.59	9.34	5.29	5.08	4.43	5.29	6.84	
<i>ILRI 5447</i>	4.86	15.53	9.82	11.19	6.15	6.97	5.73	6.15	8.30	
Mean	4.51	11.88	8.75	9.09	6.83	7.83	5.34	6.83	7.60	
CV %	29.18	30.48	26.5	16.32	30.53	22.29	32.74	30.53	25.9	
LSD (0.05)	2.19	6.21	3.98	2.54	3.56	3	3	5.58	1.97	
P-Value	0.1031	0.147	0.087	0.059	0.2363	0.0252	0.995	0.2367	0.0691	

3.3. Herbage Quality Parameters

The chemical composition of the oats samples are presented in Table 2. The Crude protein content averaged (10%) ranging from (9.8%) for accession ILRI 5442 to (10.33 %) for Bareda (ILRI 5450) variety that have 6.94 and 7.16% yield advantage over SRCPX80Ab2806 and bonsa respectively. The organic matter content was higher for accessions variety SRCPX80Ab2806 with mean values of (83.16%) followed by Bonsa (82.9 %). The IVDMD was higher for variety/acc. ILRI 5467 (57.88%) followed by Bareda (ILRI 5450 (57.75%) and the lowest for variety Bonsa (57.14%). As regards to fiber quality, the lowest ADF and NDF were displayed by Bonsa and ILRI 5445 accession and (62.83% and 63.85%), respectively and the highest were recorded from ILRI 5467 and ILRI 5457 Accession (68.66 % and 75.62%) in that order.

Table 2: Mean nutritive value of different accessions of *oat* genotypes at regional variety trial of at multi locations.

Genotypes	% DM							
	DM %	CP	NDF	ADF	ADL	IVDMD	Ash	OM
Bareda (ILRI 5450)	92.99	10.33	74.15	65.38	7.82	57.75	10.58	82.13
ILRI 5442	92.84	9.8	74.83	65.79	7.08	57.32	10.57	82.16
SRCPX80Ab2806	92.63	9.66	73.35	63.04	7.97	57.53	9.47	83.16
Bonsa	92.71	9.64	73.35	62.83	7.82	57.14	9.81	82.9
ILRI 5457	92.89	10.03	75.62	65.62	7.36	57.68	11.4	81.5
ILRI 5467	92.85	10.2	68.66	68.66	7.38	57.88	11.22	81.63
ILRI 5478	92.8	10	74.7	68.11	7.37	57.62	10.71	82.09
ILRI 7251	92.82	10.02	75.1	64.09	7.89	57.72	10.89	81.93
ILRI 5445	92.8	10.35	63.85	63.8	7.3	57.43	10.44	82.37
ILRI 5447	92.76	10	74.57	66.36	7.44	57.62	10.6	82.16
Grand Mean	92.8	10	74.57	65.17	7.54	57.53	10.57	82.21
CV %	0.2	7.62	1.73	4.31	12.06	1.66	14.56	1.83
LSD (0.05)	0.26	1.11	1.88	4.07	1.32	1.39	2.23	2.18
Genotype (trt)	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns
Location	***	***	***	***	***	***	***	***

Bonsa and SRCPX80Ab2806 (Check), DM= Dry matter, CP=Crud protein, NDF= Neutral detergent fiber, ADF= Acid detergent fiber, IVDMD= Invitro dry matter digestibility

3.4. Farmers and DA's Preferences

Results from experimental farmers and developmental agents were collected and analyzed through Henry Garrett ranking method during variety verification trials. The result indicates that the perceived degree of importance of Bareda variety was ranked first based on the criteria's like leafiness, green fodder yield, standing vigor, plot cover, absence of lodging, disease tolerant, softness and fast growth. The visual and hand evaluation of the farmers and developmental agents were very critical for ranking of this cultivars. Accordingly, the average rank showed that farmers and developmental agents gave the first score for Bareda variety followed by Bonsa (standard check) (table 3).

Table 3: Garret ranking result for farmer selection

Accessions/Varieties	Total Score	Average Score	Rank
5442	848	84.8	3
5450	920	92	1
Bate	840	84	4
Bonsa	864	86.4	2

3.5. Stability Performance

Analysis using the GGE bi-plot confirmed that Bareda (ILRI 5450) variety was fell into the center of concentric circles indicating that it is ideal variety in terms of higher yielding ability and stability (Fig 1). Likewise, ILRI Acc.5442 genotype was located on the next circle. Genotypes that fall in the central (concentric) circle are considered as ideal environments and stable genotypes (Yan *et al.*, 2001).

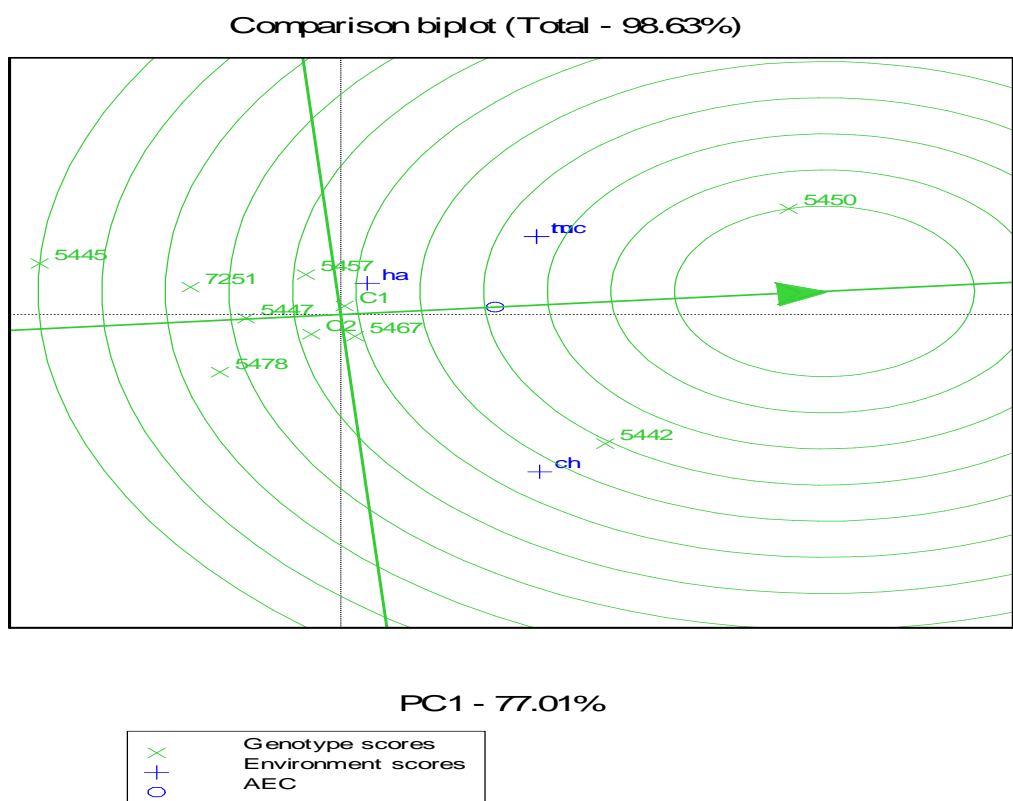


Fig 1. GGE bi-plot analysis of genotypes and environments for Dry matter yield

Table 4. Mean forage yield, agronomic traits and disease reaction of Bareda *and* adapted checks in multi-location tested

Variety	Sdyldqu/ha)		FBYt/ha		DM (t/ha)	DS	LSR	FP
	ReF	FF	Re	FF				
Bareda (Acc.5450)	33.4	24	47.47	44.5	9.25	1.3	0.66	1
Acce No 5442	40.42	27	45.13	28	7.99	1.5	0.71	3
Bonsa	29	22	36.19	34.6	6.62	1.3	0.61	2.7
Bate	23	17	34.88	32.1	*	1.5	0.44	3.3
Means	31.46	22.5	40.92	34.8		1.4	0.61	2.5

* = data is not available, Sdyldqu/ha = seed yield quintal per hectare, ReF = at research field, FF = at farmers field, FBYt/ha= fresh biomass yield tone per hectare, DS = DS = diseases score, LSR = leaf stem ratio, FP = Farmers preferences

4. Reaction to Disease and Pest

The most common diseases of oats are leaf rust, stem rest and crown rust. On 1-5 rating scale, *Bareda and Bonsa* scored a mean of 1.3 and *Bate* scored 1.5 for leaf rust diseases. Hence, the released varieties are characterized by more tolerant to the major diseases at all sites. The disease score results for the varieties and the checks are summarized in Table 4.

5. Conclusion and Recommendation

The released variety, Bareda 'Acc. 5450' has better herbage dry matter yield performance, good general adaptability, stability, disease and pesttolerant from the tested as compared to other rested genotypes. The released variety also has better nutritional quality, especially dry matter, crude protein and organic matter. Therefore, smallholder farmers and other stakeholders who have engaged in animal production can utilize the Bareda variety as energy supplements for low quality feed resources.

6. Acknowledgement

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Agronomic/morphological characteristics of Oat variety, Bareda (Acc.5450)

Characteristics	
Variety Name	Bareda
Adaptation area	Mechara, Gelemso, Chiro, Tulo and similar agro ecologies
Altitude(m.a.s.l)	1550 - 2400
Rainfall(mm)	-
Fertilizer rate	
Nitrogen(kg N ha ⁻¹)	46
NPS(kg P ₂ O ₅ ha ⁻¹)	19
Fertilizer application time	At sowing stage
Fertilizer application method	Row drilling
Planting or seeding	Row drilling
Planting date	Early July
Seed rate(kg ha ⁻¹)	100
Row spacing(cm)	30
Plant spacing(cm)	Drilling
Weeding frequency	Two week after sowing then each three weeks to 100% flowering
Days to flowering (days)	56-90
Days to Maturity (days)	87-155
Plant height(cm)	78-138
Inflorescence compactness	-
Seed color	pale brown
Crop pest reaction(1-5 scale)	
Leaf blight(Leaf rust)	2
Stem borer	1
Grain mold	1
Yield(Qt ha⁻¹)	
Research field	27-39
Farmers' field	16-32
Year of release	2020
Breeder seed maintainer	Mechara Agricultural Research Center

Evaluation of Grass-legume Mixed Silage for Enhancement of Silage Quality under Sub-humid Climatic Condition of Western Oromia, Ethiopia

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Abstract

Feed conservation practices in the form of silage could be an option to increase dry season feed availability. Such technology would ensure satisfaction of the high demand for nutrients for livestock production operations during dry period. Grass and legume silage makes it possible to increase the home-grown protein supply, so that with good grass-legume silage, the requirement for other protein supplements can be reduced. The study was conducted at Bako Agricultural Research Center during the year 2019 and 2020 to investigate the nutritional property of silage made from mixtures of grass/maize and a legume (Lablab purpureus). This food-feed mixture is composed of BH-543 variety of maize, Bako-04 variety of Napier grass and Gebis-17 variety of Lablab purpureus (LP). The treatments were: T1 (sole Napier grass), T2 (sole maize), T3 (Maize + 20 % LP), T4 (maize + 30 % LP), T5 (maize + 40 % LP), T6 (Napier grass + 20 % LP), T7 (Napier grass + 30 % LP), T8 (Napier grass + 40 % LP). The experimental treatments were replicated three times in a Randomized Complete Block Design (RCBD). The results demonstrated that inclusion of lablab forage to grass-legume mixture silage increased its nutrient content in terms of CP, OM and IVOMD c. The CP, OM (and IVOMD contents of T5, followed by those of T4 were higher than the silage from sole maize with respective values of 10.9 %, 92.94 % and 63.1 % for T5 and 10.3 %, 91.89 % and 61.5 % for T4 and 7.06, 91.24 and 58.61 % for sole maize silage. Increasing lablab levels didn't affect silage pH, which is one of the determinants that influence the extent of fermentation and quality of silage. In the current study the mean values of silage pH was 4.63, which is considered to be an optimum PH value for a good quality silages. In conclusion, inclusion of lablab purpureus at 30-40% in Napier grass/maize-lablab mixture silage is recommended over the silage made from sole grass or maize in terms of improving the silage nutrient content and quality.

Key words: Grass-legume mixtures, *lablab purpureus*, Napier grass, silage quality

1. Introduction

Livestock is recognized as being an integral component of the mixed farming systems. Animal manure and traction make the land more productive than would be the case in their absence. Yet, it has been recognized with equal force that livestock caused a devastating effect on the environment through overgrazing the natural vegetation which leads to soil erosion and ultimately desertification (Steinfeld *et al.*, 1998). Technologies aimed at achieving a balance whereby livestock can increase productivity, while resource degradation is minimized, must be developed (Steinfeld, 1998). In the dry period the grazing

areas are usually very poor; consequently animals lose weight, thereby resulting in declined productivity (Dube, 1995). Feed conservation practices in the form of silage could be an option to increase dry season feed availability (Alemayehu *et al.*, 1997; Steinshamn, 2008). Such technology would ensure satisfaction of the high demand for nutrients for livestock production operations (Dube *et al.*, 1995)

Plant species is one of the factors that affect silage quality (McEniry *et al.*, 2013). Different grass species have different ensiling abilities, depending on the content of water soluble carbohydrates (WSC) and buffering capacity. Legumes as an important source of protein are very difficult to ensilage due to the high buffering capacity and low concentrations of easily soluble sugars (Wyss, 2004; Bijelić.Z. *et al.*, 2015). In mixture with grasses, their fermentable characteristics are improved so that silage of adequate quality can be obtained. Grass and legume mixture silage makes it possible to increase the home-grown protein supply. With good legume-grass mixture silage, the requirement for other protein supplements can be reduced (Bijelić.Z. *et al.*, 2015). The fodder crops, such as maize, sorghum, oats, millet, and Napier grass are rich in soluble carbohydrates and are most suitable for fodder ensiling (Steinshamn *et al.* 2008). Quality of silage can also be improved with the use of suitable additives such as molasses, urea, salt, formic acid, etc (Benjamin F. *et al.*, 2018). Therefore, the aim of this study was to investigate the contribution of protein rich forage legume on nutritional property of silage made from grass-legume mixtures.

2. Materials and Methods

2.1. Experimental Materials and their Managements

In this experiment two forage species and one cereal crop were involved: Napier grass of variety 'Bako-04', *Lablab purpureus* of variety 'Gebis-17' and maize of 'BH-543' variety. These experimental fodder crops were planted as per their relevant agronomic recommendations. The crops were managed as commercial crops in terms of fertilizer application, weeding and pest and disease control. They were harvested at their respective recommended harvesting times. Finally they were mechanically chopped into small pieces (6-10 cm length) and the chopped materials were ensiled in plastic bag and kept in a cool dry place until samples were taken for laboratory analysis.

2.2. Experimental Design and Treatments

The experimental treatments were laid out in a Randomized Complete Block Design-(RCBD) with 3 replications.

The treatments were:

1. Sole Napier Grass
2. Sole Maize
3. Maize + *Lablab purpureus* (20 %)
4. Maize + *Lablab purpureus* (30 %)
5. Maize + *Lablab purpureus* (40 %)
6. Napier grass + *Lablab purpureus* (20 %)
7. Napier grass + *Lablab purpureus* (30 %)
8. Napier grass + *Lablab purpureus* (40 %)

2.3. Ensiling process:

Freshly chopped grasses and LP were mixed by hand. Furthermore, to facilitate silage fermentation process, 5 % molasses were mixed across the treatments. The mixed forages were ensiled in 25 kg capacity plastic bags and then packed and compacted by hand to exclude air. The materials were incubated in a room for four weeks before samples were taken for laboratory analysis.

2.4. Samples and Sample preparation:

Representative samples were collected from each treatment and dried in an oven at 65 °C for 72 hours. The oven-dried samples were ground to pass through 1 mm sieve screen size and stored at room temperature until sub-samples were taken for laboratory analysis. The ground samples were kept in sealed plastic bags pending for chemical analysis.



Figure 1. Overview of the silage preparation process

Key: A=silage being packed in plastic bags and ready for ensiling, B= packed and sealed silage ready for incubation in a dry and free room, C= Fermented silage being observed for its physical characteristics (flavor, odor and color), D= data collection and sampling

2.5. Laboratory Analyses procedures:

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)

2.6. Statistical Analysis : Collected data were analyzed by ‘R’ software (R, 2017, version 3.4.1). Significantly different means were separated by using Tukey’s LSD.

3. Results and Discussion

Summary of analysis of variance for most of the tested variables of the treatments are presented in Table 1. The treatment mean squares varied in DM yield ($p < 0.05$), Organic matter (OM), Crude protein (CP), in vitro organic matter digestibility (IVOMD), Ash and acid detergent fiber (ADF) at the level of significance shown in the Table while they were not significantly different for NDF and ADL ($p > 0.05$).

Table 1: Mean squares of chemical composition of grasses and legume silage mixtures

Source of variation	DF	Mean squares								
		DM	CP	OM	Ash	NDF	ADF	ADL	IVOMD	pH
Rep	2	5.334	1.607	0.389	0.591**	88.88	33.02*	0.58	2.615	0.280
Treatment	7	4.868*	10.259***	10.97***	10.588***	26.56	5.89**	1.06	44.16***	0.067
Residual	14	1.670	0.679	0.204	0.081	27.34	7.54	0.864	1.268	0.047
Total	23									

Where, *, **: Significant at 5% and 1%, respectively. DF: Degree of freedom, DM: Dry matter, CP: crude protein, OM: organic matter, NDF: neutral detergent fiber, ADF: acid detergent fiber, ADL: acid detergent lignin, IVOMD: in vitro organic matter digestibility.

3.1. Chemical Composition of Silage Mixtures

The chemical composition of grass-legume silage mixtures and their descriptive statistics are shown in Table 2. The mean silage DM content was 89.2 % with values ranging from 87.39% for T3 (maize 20% LP) to 91.16% for T6 (Napier grass + 20 % LP). The dry matter content of silage is one of the important parameters that determine the success of fermentation of the ensiling material. The ash contents were significantly higher ($p < 0.001$) for T1 (sole Napier grass), T6 (Napier grass + 20 % LP) and T7 (Napier grass + 30 % LP) with mean respective values of 11.84, 11.46 and 11.21% , whereas it was lower for T5 (maize + 40 % LP), T4 (maize + 30 % LP) and T2 (sole maize) with mean values of 8.11 %, 8.39 % and 7.06 %, respectively. The crude protein (CP) content ranged from 4.98 % for T1 to 10.9 % for T5, with overall mean of 8.56 %. Similarly, the OM content was significantly ($p < 0.001$) higher for T5 (92.94 %) and T4 (91.89). Lower value of OM was recorded for T1 (88.16 %).

The concentration of ADF varied significantly ($p < 0.002$) among the treatments ranging from 33.59 % for T5. to 45.5 % for T1. The Neutral detergent fiber (NDF) and Acid detergent lignin (ADL) concentrations did not differ ($p > 0.05$) among treatments. However, numeric advantages in both NDF and ADL were observed. Increases in legume (lablab) inclusion levels significantly increased the concentrations of CP and decreased the concentration of ADF in the ensiled materials. This indicates a good quality silage and suggests that a grass-legume forage mixtures improved nutritive value of the silage. Similar to the current study, increasing the legume (alfalfa) proportion in the grass-legume silage mixtures increased CP and decreased ADF concentrations (ZhulinXue *et al.*, 2020).

Table 2. Chemical composition and *in vitro* DM digestibility of the of grass-legume mixture silage

Treatments	DM%	Silage quality traits (%DM)					
		Ash	OM	CP	NDF	ADF	ADL
1. Sole Napier grass	90.85 ^a	11.84 ^a	88.16 ^d	4.98 ^d	64.15	45.50 ^a	9.29
2. Sole Maize	88.59 ^{bc}	7.06 ^e	91.24 ^{bc}	7.33 ^c	65.45	37.18 ^{cd}	8.32
3. Maize + 20 % LP	87.39 ^c	8.82 ^c	91.61 ^b	8.79 ^b	60.78	35.89 ^{cd}	7.91
4. Maize + 30 % LP	90.55 ^{ab}	8.39 ^{cd}	91.89 ^{ab}	10.3 ^a	61.82	36.03 ^{cd}	8.47
5. Maize + 40 % LP	90.38 ^{ab}	8.11 ^d	92.94 ^a	10.9 ^a	57.10	33.59 ^d	7.83
6. Napier grass + 20 % LP	91.16 ^a	11.46 ^{ab}	88.54 ^{cd}	8.43 ^{bc}	66.76	42.82 ^{ab}	9.50
7. Napier grass + 30 % LP	90.25 ^{ab}	11.21 ^b	88.78 ^{cd}	8.21 ^{bc}	62.62	39.16 ^{bc}	8.70
8. Napier grass + 40 % LP	89.2 ^{abc}	9.38 ^{bc}	89.28 ^c	9.53 ^{ab}	62.76	39.26 ^{bc}	8.65
Grand mean	89.68	9.79	90.42	8.56	62.68	38.68	8.58
CV%	1.4	2.9	0.5	9.6	8.3	7.1	10.8
LSD (0.05)	2.263	0.499	0.791	1.443	9.157	4.81	1.628
<i>P-value</i>	0.042	<0.001	<0.001	<0.001	0.448	0.002	0.352

Key: CV=coefficients of variation, LSD= Least significant differences, LP= lablab purpureus; DM=dry matter; OM=organic matter; CP=crude protein; NDF=Neutral detergent fiber; ADF=acid detergent fiber; ADL=acid detergent lignin;

a,b,c,d,e=Means with different superscript letter in a column are significantly different at a significance level given in the table

4.2. *In vitro* Degradation and Ensiling Characteristics of the Silage

Averaged over the evaluated treatments, the IVDMD content was 57.96 %, with the highest value obtained from T5 (63.1 %) and lower value from T1 (52.76) followed by T6 (52.95 %). In the present study, the increase of legume (lablab) in the mixtures increased the IVOMD and OMD compared with that for both sole grasses. This trend is in agreement with previous studies (Dal Pizzol, J.G. *et al.*, 2017; ZhulinXue *et al.*, 2020), in which quadratic increase in IVDMD was reported when the proportion of alfalfa increased in axonopus-alfalfa and tall fescue-alfalfa mixtures.

This may be because of the balanced digestible nutrients from grass components (Napier grass and maize) and lablab silage mixtures set off a ruminal synergistic effect on the fractional rate of degradation and the extent of fermentation, followed by better nutrient availability and utilization efficiency for rumen microorganisms (Kiran, D. *et al.*, 2007). The association between fermented nutrients from grasses and legume mixtures may also lead to synergistic effects on the dominant microbial populations and shifts in the microbial community composition (Pandit, R.J. *et al.*, 2018).

Silage pH is one of the major determinants that influence the extent of fermentation and quality of ensiled crops. A well preserved silage usually has a low pH but high acetic acid concentration (ZhulinXue *et al.*, 2020). In the current study, an increase in the lablab proportion in the mixtures did not statistically affect ($P>0.05$) the silage pH concentration but numerical differences were observed (Table 3). This is in disagreement with the previous study from Contreras-Govea *et al.* (2011) in which legumes such as alfalfa, pea, and red clover did not easily make good silage because of the high buffering effects of both

plant and silage microbial enzymes, which compromised the ensiling quality compared with that associated with grasses.

Table 3. Silage pH and *in vitro* degradation characteristics of grass-legume silage

Treatments	Silage quality traits	
	pH	IVDMD
1. Sole Napier grass	4.42	52.76 ^c
2. Sole Maize	4.57	58.61 ^c
3. Maize + 20 % LP	4.82	58.58 ^c
4. Maize + 30 % LP	4.64	61.5 ^{ab}
5. Maize + 40 % LP	4.47	63.1 ^a
6. Napier grass + 20 % LP	4.81	52.95 ^c
7. Napier grass + 30 % LP	4.60	55.71 ^d
8. Napier grass + 40 % LP	4.75	60.4 ^{bc}
Grand mean	4.63	57.96
CV%	4.7	1.9
LSD (0.05)	0.381	1.971
P-value	0.271	<0.001

Note: IVDMD=*in vitro* dry matter digestibility; LP=*Lablab purpureus*; CV=coefficients of variation, LSD= Least significant differences.

a,b,c,d,e=Means with different superscript letter in a column are significantly different at a significance level given in the table

Conclusions and Recommendation

From this study of silage preparation from mixtures of either Maize or Napier grass with different levels of LP, inclusion of *Lablab purpureus* (LP) at different proportions in the grass-legume silage had better nutritional quality, but didn't affect fiber parameters like NDF and ADL. The increase in the LP proportion in the mixtures increased OM, CP and IVDMD of the silage compared with the silage made from the sole plants (Napier grass and maize). These results were helpful for the effective production, preservation, and utilization of superior forages without causing environmental concerns. Grass-legume mixtures (Napier grass/maize with *lablab purpureus*) can be made into good silage with **certain** additives. Napier and *Lablab* can be ensiled together to make a reasonably good quality silage, but the quality can be improved with **certain** additives (e.g. 5% molasses) before ensiling. Therefore, compared with sole grasses or maize silages, addition of LP at 30 % and 40 % in maize and 40 % in Napier grass silage were recommended for favorable ensiling and higher quality, thus resulting in increased forage use efficiency.

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Fishery Research Results

Chemical preservation of fish offal and odor elimination during fish meal processing

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Abstract

Fish waste product cannot be used to their full potential due to their foul smell which is considered as massive environmental pollutant. The objective of this study was to preserve fish offal using chemicals and eliminate odor during fish meal processing using Effective microorganisms. Fish offal was collected from Zeway-Batu Fishermen Cooperative fish processing shade. 10kg offal was preserved by spraying 1% by weight of Anhydrous Ammonia, 6% Hypochlorite, 1% Benzoic acid and 1% Sodium Nitrite onto the fish offal in 10 L capacity bucket and sealed air tightly for 15 days. 10 kg of fish meal was separately spread thinly on 1m² and sprayed with EM at the rate of 0 lit/m² (control), 0.2 lit/m², 0.4 lit/m² and 0.6 lit/m² drying. Fish offal which is not treated with chemical was strongly odorous, fly larva were invaded the lid of bucket and 50 flies were present on the second day. The offal has under gone putrefaction and it was discarded after 48 hrs. After 15 days of preservation, fish offal preserved using 1% Sodium Nitrite was odorless and no flies and its larva were present on the lid. Fish meal sprayed with 0.4 lit/m² EM had a specific odor of fish meal, light brown color to dark brown, not greasy, not clumping and not indicating physical changes and rancidity after four months of storage. EM could be potentially useful as a tool for reducing odor induced during fish meal processing.

Keywords: Fish offal, fish meal, chemical preservative, Effective microorganisms

1. Introduction

Fish offal is wasted fish that is not destined for direct human consumption, but which is processed into feeding-stuffs used in the production of foods such as chicken, eggs and pork. Approximately 75% of world fish production is used for human consumption and the remaining 25% is used to produce fish meal and oil. The process of making fish meal from fish and fish offal generates odors (<http://www.fao.org/wairdocs/tan/x5943e/x5943e01.htm>).

Fish waste product cannot be used to their full potential due to their foul smell which is considered as massive environmental pollutant. This pollution is immense in the towns where small scale fish processing private irresponsibly discharges thousand liters of waste to the environment. Small scale fish processing operations produce wastewater, which contains active organic contaminating organisms in soluble, colloidal and particulate form. There is an increasing number of citizens' complaints about odor nuisance due to production or service activity. High social awareness imposes pressure on entrepreneurs and service providers forcing them to undertake effective steps aimed at minimization of the effects of their activity, also with respect to emission of malodorous substances.

The environmental authority around Batu town has closed two small scale fish meal processing individuals. They were forced to move to Meki and Bochessa rural areas. Although there is limited evidence that serious risks to physical health occur through odorous emissions from fish meal plants; some research report that odor source substances can cause health effects such as eye, nose and throat irritation, headache and drowsiness; and possibly aggravate allergies, asthma and bronchitis (Odor Control Task Force, 1998). The odor of spoiled fish may be eliminated with application of effective microorganism (EM) technology. Effective Microorganisms (EM), a fermented medium developed by Professor Higa at the University of the Ryukyus, is a mixed culture containing dozens of microorganisms which are beneficial to nature including people, animals, plants and many microbial species in environment. EM is known to contain more than 80 kinds of anaerobic or aerobic microbes including photosynthetic bacteria, lactic acid bacteria, yeast, actinomycetes, fungi and so on, with yeast, lactic acid bacteria and photosynthetic bacteria as the main species of EMs. The use of effective microorganism technology has broadened in the last two decades from agriculture to water treatment, odor control, animal husbandry, human health, and other numerous industrial treatments (Higa, 1996).

EMs are environmentally friendly, not-naturally organized, not chemically synthesized, not dangerous and not pathogenic. EM technology proved effective in odor control and sanitation management (Abdullah *et. al.*, 2011). EM also has the ability to absorb toxic gases (Hydrogen sulphide and Ammonia) and convert them into organic acids, thereby eliminating their foul odor. EM has been thoroughly tested and proven safe for human and other animals. The United States Department of Agriculture categorizes all of the microorganisms in EM as GRAS (Generally Recognized As Safe). The United States FDA categorizes most of the organisms in EM as food grade microorganisms. EM eliminates odors by dominating the microbial ecology with organisms that exploit a fermentative pathway and therefore do not produce odorous gases.

EM is used in agriculture, animal husbandry, aquaculture, waste water and solid waste management to increase the quantity and to improve the quality of products and the treating of certain polluting elements. The application of EM to the environment including garbage and sewage reduces and virtually eliminates smells and also the populations of flies. This makes the environment a better place for all living organisms. In addition to EM, there are several chemical agents which are nontoxic, not impart odors, not impair nutritive value, not accelerate rancidity and noncorrosive which preserve and extend the shelf life of fish offal's. Malodours produced during the production of fish meal tend to be very much worse when spoiled fish are processed; odours from the deteriorating stored raw material waiting processing can also cause offence. Therefore, it is necessary to preserve the offal before processing into fish meal. The objective of this study was to preserve fish offal using chemicals and eliminate odor during fish meal processing.

2. Materials and Methods

2.1. Preservation of fish offal using chemicals

Fish offal was collected from Zeway-Batu Fishermen Cooperative fish processing shade. Four chemicals (anhydrous ammonia, Hypochlorite, Benzoic acid and Sodium Nitrite) have been purchased from local markets. 10kg offal was preserved by spraying 1% by weight of Anhydrous Ammonia, 6% Hypochlorite,

1% Benzoic acid and 1% Sodium Nitrite onto the fish offal in 10 L capacity bucket and sealed air tightly for 15 days. The variables observed were sensory properties like aroma, color and texture.



Fig.1 Fish offal preserved using four different chemicals

2.2. Application of Effective Microorganism to fish meal

Effective microorganism (EM) used in this study was supplied by EM Technology as liquid culture contained a mixture of lactic acid bacteria *Lactobacillus planetarium* (1.0×10^4 CFU/ml), Yeast with 1.0×10^5 CFU/ml *Candida utilis*, actinomycetes *Streptomyces albus* (3.0×10^3 CFU/ml), fermenting fungi *Aspergillus oryzae* (1.1×10^5 CFU/ml). EM solution is a brownish liquid with a pleasant odor and sweet sour taste with a pH of 3 and stored in cool place without refrigeration. Fish meal was processed following six steps: cooking, pressing, separating, evaporation, drying and grinding (Abera *et al.*, 2008). 10 kg of fish meal was separately spread thinly on 1m^2 and sprayed with EM at the rate of 0 lit/ m^2 (control), 0.2 lit/ m^2 , 0.4 lit/ m^2 and 0.6 lit/ m^2 drying.

2.3 Foul smell evaluation

Sensory evaluations of the odor of all collected air-samples in the bucket were performed by panels of people. The collected odor samples (odorants were adsorbed onto cotton fabric media) were presented to an odor panel (a group of people trained to detect odor). Panel members with normal senses of smell, were selected in advance. Panel members were not permitted to eat or smoke for one hour before the session to ensure the quality of their results. Panel members were in the odor room for 15 minutes before the measurements were made. They did not use perfumes, after-shave lotions or any other fragrant essence before the session. Panel members were not allowed to attend a session if he/she had a cold, influenza or any other health problem that could affect his/her nose. The panelist rated the treated fish meal as odorless, slightly odorous, moderately odorous and strongly odorous.

3. Results and discussion

3.1. Effects of chemical preservation on fish offal

Chemicals which are nontoxic, not impart odors, not impair nutritive value, not accelerate rancidity and non-corrosive are very important for preserving fish offal. The variables which were observed during offal preservation were physical characteristics (texture and odor) of fish offal and fish meal. From the study it was observed that, 1 % Sodium nitrite is effective in preserving fish offal. Some study indicate that fish offal containing more than about 2 % sodium nitrite may cause death in some animals, eg Mink, since toxic nitrosamines can be produced during drying (Windsor and Thoma, 1974).

Table 1. Physical characteristics of fish offal preserved with different chemicals

Treatment	Physical characteristics	
	Texture	Odor
1% Ammonia solution	Not greasy, not clump	Moderately odorous
6% Hypochlorite	Moderate clumping	Slightly odorous
1% Benzoic acid	Not greasy, not clump	Moderately odorous
1% Sodium Nitrite	Not greasy, not clump	Odorless
No chemical added	Greasy putrid	Strongly odorous

Fish offal which is not treated with chemical was strongly odorous, fly larva were invaded the lid of bucket and 50 flies were present on the second day. The offal has under gone putrefaction and it was discarded after 48 hrs. After 15 days of preservation, fish offal preserved using 1% Sodium Nitrite was odorless and no flies and its larva were present on the lid. Although the preservative action of anhydrous ammonia seems very effective the cost of the treatment would be high and there might be difficulties in handling ammonia solutions. However, the work showed that fish meal, made from fish preserved with ammonia and then washed, retained its nutritive value and that there were no toxic residues. Anhydrous ammonia is, however, highly toxic and handling it would require special equipment and precautions (Windsor and Thoma, 1974).

3.2. Effects of EM application on fish meal

Effective Microorganisms was sprayed on fish meal to reduce the odor specifically the concentration of NH_3 , H_2S and CH_3SH . Fish meal sprayed with 0.4 lit/m^2 EM had a specific odor of fish meal, light brown color to dark brown, not greasy, not clumping and not indicating physical changes and rancidity after four months of storage. Major odorous gases from fish meal processing sites consist primarily of hydrogen sulfide (H_2S), tri-methylamine (TMA) and dimethyl sulfide (DMS) (Neil, 1993). EM also has the ability to absorb toxic gases (Hydrogen sulphide and Ammonia) and convert them into organic acids, thereby eliminating their foul odor (Eduardo, 2007).

Table 2. Physical Characteristics of Fishmeal sprayed with EM at different rates of during drying

Treatment	Physical characteristics		
	Color	Texture	Odor
0 lit/m^2	Dark brown	Greasy/clumping	Strongly odorous
0.2 lit/m^2	Moderate brown	Moderate clumping	Slightly odorous
0.4 lit/m^2	Light brown	Not greasy, not clump	Moderately odorous
0.6 lit/m^2	Brown	Not greasy, not clump	Odourless

EM works by overcoming the odour producing microbes (such as sulphide producing bacteria), and replacing them with the beneficial microbes that are contained in EM. They then change the process of breakdown so that the odour producing microbes are displaced. The effect of spraying an odorous environment can be very rapid, the odour can be reduced to 80% within 8 hours. Fish meal sprayed with 0.4 lit/m^2 EM had a specific odor of fish meal, light brown color to dark brown, not greasy, not clumping and not indicating physical changes and rancidity after four months of storage.

4. Conclusion and recommendations

Odor measurement is difficult because no instrument has been found to successfully measure an odor and all its components. Currently, the human nose is the only device that can really measure odor, and even then personal preference affects what is considered acceptable or offensive. The multiple functions of EM is closely related to the actions of the dominant species in EM. The EM could be potentially useful as a tool for reducing odor induced from the fish meal processing. From the study it was concluded that, 1 % Sodium nitrite is effective in preserving fish offal. Fish meal sprayed with 0.4 lit/m² EM had a specific odor of fish meal, light brown color to dark brown, not greasy, not clumping and not indicating physical changes and rancidity after four months of storage. The present result indicate that EM use in fish meal processing has great potential for suppressing malodors, improving sustainable production, and protecting the environment, all on a cost-effective basis. The use of Effective Microorganisms is certainly a technology that deserves considerable and serious attention. It is beneficial potential for creating a sustainable world is too promising. It is recommended that, any small scale fish processing individuals or company can use 1 % Sodium nitrite to preserve fish offals for 15 days and spraying of 0.4 lit/m² EM remove noxious odor during fish processing.

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Habitat suitability and fish production potential of GenaleDawa 3 and Gidabo Reservoir

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Abstract

Many reservoirs in Ethiopia have been built for hydroelectric power, water supply, or irrigation purposes, which are also serving as fish production sites. Establishment of Fisheries in the newly built Reservoirs GenaleDawa 3 and Gidabo were studied during the initial impoundment. Production potential of the reservoirs was estimated based on empirical methods. Six fish species were recorded in both GenaleDawa 3 and Gidabo Reservoir dominated by *Labeobarbus intermedius* (Rüppell, 1835). Length-weight relationships were highly significant with high coefficients of determination ranging from 0.872 for *Varicorhinus jubae* up to 0.990 for *Labeobarbus intermedius*. 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 28.9 cm TL at GenaleDawa 3 reservoir while it was 33.4 at Gidabo. Based on the morphometric characteristics of the reservoirs, an average fish production of 133 t/year and 789.6 t/year was estimated for Gidabo and GenaleDawa 3 reservoirs respectively. Our results show a clear adaptation and fish colonization pattern during initial impoundment. This helps in establishment of successful fisheries in both Reservoirs. The results also highlight the importance of considering stocking of *Oreochromis niloticus* as successional patterns of fish colonization in order to improve the fisheries in the future.

Keywords: fish biodiversity, fishery management, impoundment, reservoir creation,

Introduction

Many reservoirs in Ethiopia have been built for hydroelectric power, water supply, or irrigation purposes. Yet despite the initial prescribed reservoir purpose, reservoirs are perceived as multiuse infrastructures with the ability to provide several essential services to nearby communities (Degefu *et al.* 2015, Jembere and Yihdego 2016). In Ethiopia, freshwater fish production is important for nutritional security in riparian communities. Being landlocked, lakes, rivers, and reservoirs of the country are an important source of protein and micronutrients (Tesfaye and Wolff 2014).

Reservoirs are managed ecosystems that share several common features with natural lakes (Kolding and Van Zwieten 2006). The presence of the pelagic zone and processes such as primary production or predator-prey interactions are the same in reservoirs as in natural lakes (Thornton *et al.* 1990). Nevertheless, Dams impact 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 (Thornton *et al.* 1990, Forsberg *et al.* 2017). Some migratory fish species reproduce in upstream or headwater locations before returning to their downstream feeding sites (Esguícero and Arcifa 2010).

Fish production potential of a reservoir depends on multiple abiotic environmental factors (e.g. discharge, geomorphology, surface area, depth, and habitat structural complexity), biotic factors (e.g. species pool,

dispersal ability), and the design, age and operational features of dams (Agostinho *et al.* 2016). The combined effects of altered environmental conditions and fragmentation of riverscapes result in significant changes in fish assemblage structure and functional diversity in rivers (Arantes *et al.* 2019). Such shifts are associated with traits that convey either tolerance or vulnerability to the new ecological conditions of the impacted system.

Empirical models have been used to predict fish yields in reservoirs. (Henderson *et al.* 1973, Tesfaye *et al.* 2011, Tesfaye and Wolff 2014, Teame *et al.* 2016). The theoretical basis for such models is that fish production is largely determined by the level of primary production in an aquatic system. Statistical models (mostly linear regressions) relating morphometric and edaphic factors to fish yields in reservoirs were widely used to predict fish production in data limited fisheries, and also for prediction of newly constructed reservoirs (Moreau and De Silva 1991, Esguícero and Arcifa 2010). These models seek to predict complex biological effects from simple environmental parameters (MRAG 1995). Yield predictions have been based on primary production (Downing *et al.* 1990), total phosphorus (Yurk and Ney 1989, Nissanka *et al.* 2000) or its surface area (Crul 1992).

This study aimed at identifying the retained fish species during initial impoundment of Genale-Dawa 3 and Gidabo reservoirs, in Ethiopia and estimating their fisheries production potential in relation to their morpho edaphic parameters.

Materials and Methods

Study area

Genale Dawa 3 hydroelectric reservoir (GD-3) is located on the Genale River, Ethiopia (5°38'N, 39°43'E). Administratively the Reservoir falls under the jurisdiction of the Liben and Mede Welabu woredas in the Guji and Bale zones of the Oromia National Regional administration respectively. At full supply level the reservoir covers an area of 98 km² (Lahmeyer International and Yeshe-Ber Consult 2007). Gidabo irrigation Dam is located in Gidabo river (6°20' N, 38° 05'), within the boundary of Abaya district of Oromia Regional state and Dale district of Sidama Regional state, near Dilla town to the east of Lake Abaya. At full supply level the reservoir covers 27 km² (WWDSE 2009).

Table 11 Physical properties of Gidabo and Genale Dawa 3 reservoir

Physical characteristics	Genale Dawa 3	Gidabo
Catchment Area (km ²)	10,445	2532
Reservoir full supply level (m a.s.l.)	1120	1217
Surface area at full supply level (km ²)	98	27
Total storage volume at full supply level (km ³)	2.57	0.063
Maximum depth (m)	110	21
Catchment area (km ²)	10445	3,302
Annual rainfall (mm)	1,550	745

Fish sampling

The fish communities were sampled by multifilament gill net having (60, 80, 100 and 120 mm) stretched mesh size. GD-3 reservoir was sampled from May to June 2019, While Gidabo Reservoir was sampled from January to June 2019. Immediately after capture Fish Species were identified, and

the total (T_L) and total weight (T_w) were measured and weighted to the nearest 1mm and 0.1g respectively. Sex was determined from gonads of the specimens.

Data analysis

Length-weight relationships parameters were estimated by a linear model fitted to logtransformed data using the following formula:

$$\log_{10} W \text{ (g)} = \log_{10} a + b * \log_{10} TL \text{ (cm)}$$

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 (L_{50}) was determined from the percentages of mature fish that were grouped in 1 cm length classes and fitted to logistic equation described by (Echeverria 1987).

$$P_L = (\exp(\alpha + \beta L)) / (1 + \exp(\alpha + \beta L))$$

Where P_L is proportion of mature fish at length (L) and L , is total length (cm), and α (the intercept) and β (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 "gillnetfunctions" 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. 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 a number of models developed by MARG, 1995 the 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^2), Catchment area (km^2) and Rainfall (mm), (based on 32 Lakes and Reservoirs, $r^2 = 0.844$)

$$\ln(\text{Catch}) = -10.502 + 0.484 * \ln(\text{Area}) + 0.45 * \ln(\text{Catchment}) + 1.57 * \ln(\text{Rainfall})$$

Model 2: Area (km^2), Altitude (m a.s.l.), (based on 132 Lakes and Reservoirs, $r^2 = 0.711$)

$$\ln(\text{Catch}) = 3.844 + 0.891 * \ln(\text{Area}) + -0.342 * \ln(\text{Altitude})$$

Model 3: Area (km²), Maximum depth (M), (based on 83 reservoirs, r² = 0.820)
 $\ln(\text{Catch}) = 2.625 + 0.879 * \ln(\text{Area}) + -0.121 * \ln(\text{Zmax})$

Results

Species composition: Six fish species (3 families) were recorded in GD 3 Reservoir (Table 12). In the present study, *Labeo barbus intermedius* (Rüppell, 1835) dominated the catch. Six fish species (5 families) were recorded in Gidabo reservoir (Table 12). *Labeo barbus intermedius* was also the dominant fish in Gidabo Reservoir followed by *Schilbemystus* (Linnaeus, 1758).

Length-weight relationships: LWRs were highly significant with high coefficients of determination ranging from 0.872 for *Varicorhinus jubae* up to 0.990 for *Labeo barbu sintermedius* (

Table 13). The exponent b ranged from 2.5673 for *Labeocylindricus* to 3.1296 for *Labeobarbusintermedius* in GD-3 Reservoir (Figure 12).

Table 12. Fish species composition of GenaleDawa 3 and Gidabo Reservoir

Family/ Species	GD-3	Gidabo
Bagridae		
<i>Bagrusdocmak</i> (Forsskal, 1775)	-	+
Cichlidae		
<i>Oreochromisniloticus</i> (Linnaeus, 1758)	+	-
Claridae		
<i>Clariasgaripepinus</i> (Burchell, 1822)	-	+
Cyprinidae		
<i>Labeobarbusintermedius</i> (Rüppell, 1835)	+	+
<i>Labeocylindricus</i> (Peters, 1852)	-	+
<i>Labeobarbusgananensis</i> (Vinciguerra, 1895)	+	-
<i>Labeobarbusjubae</i> (Banister, 1984)/	+	-
<i>Varicorhinusjubae</i> <i>Labeobarbusbeso</i> (Rüppell, 1835)/	-	+
<i>Varicorhinusbeso</i>		
Mormyridae		
<i>Mormyruskannume</i> (Forsskål, 1775)	+	+
Schilbeidae		
<i>Schilbemystus</i> (Linnaeus, 1758)	-	+

Table 13. Length Weight relationship of fishes in GenaleDawa 3 Reservoir

Species	TL (cm)		Regression parameters				
	Min	Max	a	b	b CI 95 %	p	R ²
<i>Bagrusdocmak</i>	43	62	0.0050	3.0957	2.6410 - 3.5502	<0.001	0.919
<i>Labeocylindricus</i>	25	52.5	0.0515	2.5673	2.1867 - 2.9478	<0.001	0.899
<i>Labeobarbusganaanensis</i>	29	61	0.0102	3.0706	2.7117 - 3.4295	<0.001	0.954
<i>Labeobarbusintermedius</i>	18	58.8	0.0084	3.1296	3.0771-3.1821	<0.001	0.990
<i>Mormyruskannume</i>	27	52	0.0073	2.9757	2.7477 - 3.2037	<0.001	0.945
<i>Varicorhinusjubae</i>	21.5	55	0.0159	2.9576	2.6181 - 3.2972	<0.001	0.872

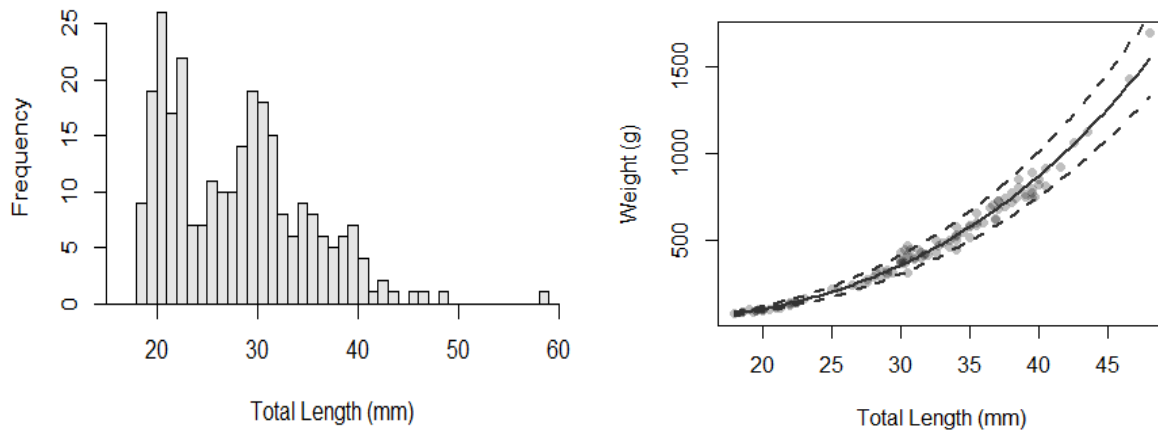


Figure 12 Length frequency distribution (a) and Length Weight relationship with 95% CI for *Labeobarbus intermedius* in GenaleDawa

Sexual maturity:The smallest fully ripe female captured during the sampling period was 27.5 cm for *Labeobarbusintermedius*, the length at first sexual maturity (L_{50}) was 28.9 cm TL at GenaleDawa 3 reservoir while it was 33.4 at Gidabo(Figure 13).

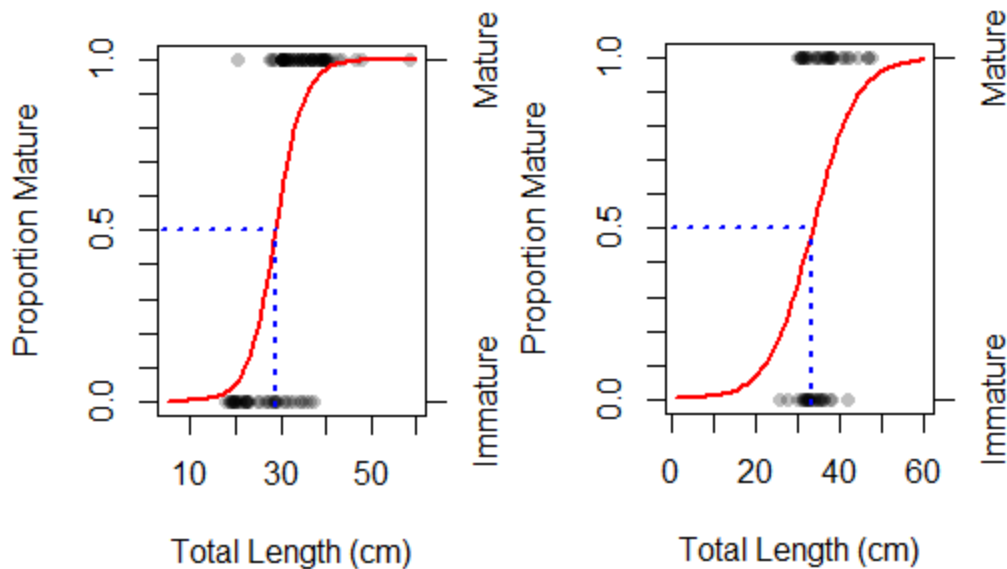


Figure 13. Length at first Maturity for *L. intermedius* of GenaleDawa 3 Reservoir (a)Gidabo Reservoir (b)

Gear selectivity: The results of the SELECT method fitted for *L. intermedius*, the dominant fish species in GD-3 Reservoir are given in (Table 14). 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. The fitted selectivity curves and deviance residuals for *L. intermedius* is shown in (Figure 14).

Table 14 Gear Selectivity parameters of GenaleDawa 3

Model	Equal fishing power		Fishing power relative to mesh size		dof
	Model deviance	null_dev	Model deviance	null_dev	
Normal	64.58	752.66	64.01	1718.86	88
Normal_sca	55.62	752.66	73.38	1718.86	88
Gamma	51.51	53.55	53.55	1718.86	88
Lognormal	50.72	50.68	50.68	1718.86	88

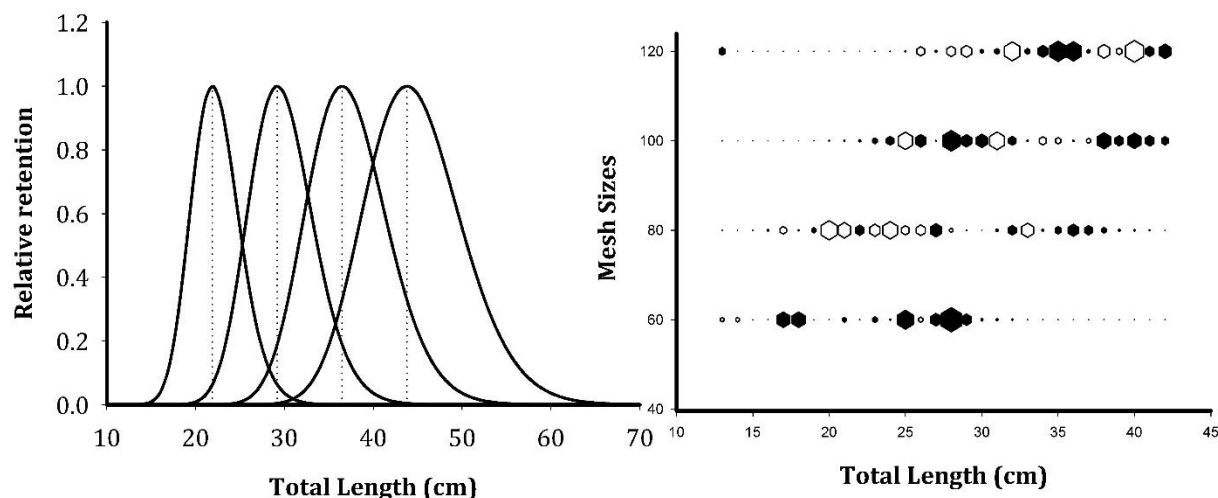


Figure 14 Gillnet selectivity curve and deviance residuals for *L. intermedius* in GD-3 Reservoir Fish production estimation:

Based on the morphometric characteristics of the reservoirs, an average fish production of 133 t/year and 789.6 t/year was estimated for Gidabo and GenaleDawa 3 reservoirs respectively (Table 15).

Table 15 Estimated annual fish production potential of GenaleDawa 3 and Gidabo Reservoir

Model	Total yield (t/year)	
	GenaleDawa 3	Gidabo Reservoir
1	1660.0	148.8
2	251.6	77.6
3	457.1	173.1
Average	789.6	133.1

Discussion

Dams influence spatio-temporal models of fish community structure and fish production by clogging migration pathways, modifying sediment transport and water quality, encouraging invasive alien species and biotic homogenization, and favoring generalists over specialized species (Monaghan *et al.* 2020). From the riverine fish species in Genale River described by Golubtsov *et al.* (2012), *Glossogobiusgiuris* and *Anguilla bengalensis* where not captured in GD-3 reservoir. Due to the absence of fish passage in the Dam, the migratory route of these fishes is blocked and their fate is restricted to the river below the Dam only (Larinier and Marmulla 2004).

In newly built reservoir in tropics *Labeobarbus* spp. tend to dominate the initial years after impoundment (Fernando and Holčík 1991), however, they tend to decline in the presence of active fishery, and stocking of fish better adapted to Reservoir environment i.e. *Oreochromis niloticus* is worth considering in the coming years. From the 25 fish species in Lake Abaya, that were perceived to colonize Gidabo Reservoir, according to the EIA report (WWDSE 2009), only seven species were retained in the Reservoir. It is known that all the fish in lotic environment might not adapt to the lentic environment. Fish that can easily

adapt in lotic environment, *Oreochromis niloticus* and *Clarias gariepinus* has established in Gidabo reservoir, which can help in establishing sustainable fishery if managed properly.

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 *Labeocylindricus* 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 FishBase were compared (Froese and Pauly 2000). Parameters a and b were within 95% of observed parameter values based on Bayesian LWR estimates calculated in FishBase (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 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).

Size at first maturity can be used to determine the appropriate fish to be harvested, in conjunction with fishing gear. Maturity can also be influenced by both genes and the environment. The current estimated maturity values can be used as a reference point when intensification of fishery occurs in the reservoirs, to see the dynamics of maturation and adjust the fishing gear and fishing intensity accordingly.

Conclusion

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 establishment of a successful fisheries in both Reservoir. The results also highlight the importance of considering stocking of *Oreochromis niloticus* as successional patterns of fish colonization in order to improve the fisheries in the future.

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Study on the effect of some medicinal plants on managing aquatic fish disease

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Abstract:

In aquaculture, infections that have both economic and zoonotic importance are considered to be the major cause of mortality. This study was undertaken to evaluate the effect of some medicinal plants (namely Moringa Olifera, Rosmarinus Officinale and Azaridica Indica, claimed to be traditionally used against infection) on managing aquatic fish disease. Healthy Oreochromis niloticus (weight 19 ± 3.8 g, Ns: 288) obtained from our center was maintained in 24 water tank (150L) in recirculation aerated system. Each water tank was stocked with twelve fish and experiment was conducted with duplicate. Fish in each tank was fed with 0% (control), 2.0%, 3.0% and 4% crude extracts of medicinal plants at the rate of 5% of their body weight twice a day for sixty days. After the feeding trial, fish were challenged by pathogenic Aeromonas hydrophila, which was given by intraperitoneal (IP) injection and they were kept under observation for fifteen days to record any abnormal clinical signs and the daily mortality rate. The evaluation of the antimicrobial activity of the individual crude extracts revealed that the plant extracts produced a dose dependent increase in the zone of inhibition from 100 μ l concentration and MIC was recorded as 19.38mg/ml, 67.5mg/ml and 67.5mg/ml for Moringa, Neem and Rosemary respectively. Hematocrit and RBCs counts were significantly increased in fish on 2.0% and 4% supplementation diet with Azaridica Indica and Moringa Olifera respectively as compared to other group. Fish fed on diets containing 4% of Rosemary Officinale, Azaridica Indica and Moringa Olifera incorporated diet exhibited 4.37 ± 0.643 , 4.21 ± 0.712 and 5.91 ± 0.0362 WBCs counts respectively which shows fish fed on diets containing 4% of Moringa Olifera incorporated diet shows significant increment of WBCs counts. Relative Percentage of Survival also clearly indicated that fish supplemented diet containing 4% moringa diet showed high protection (87%) over the control group. The lowest bacterial count was obtained in fish fed 4% Moringa Olifera, whereas the highest one was obtained when fish fed control diet. growth rate increased significantly on fish fed with 2.0% supplementation diet with Azaridica Indica and 4% of Moringa olifera as compared with similar concentration. These results indicate that herbal supplementation is promising for disease prevention in tilapia culture.

Key Words: medicinal plants, Oreochromis niloticus, Immune system, Pathogenic Bacteria

Introduction

Aquaculture has been a growing activity for the last 20 years world wide and this impressive development has been attended by some practices potentially damaging to animal health. The bacterial infections that have both economic and zoonotic importance are considered to be the major cause of mortality in aquaculture (Castro *et al.*, 2008). Fish are susceptible to severe bacterial infections, mainly when reared

in high density conditions. Disease out breaks elevated the mortality rate and decreased the productivity efficiency, causing high economic loss of the fish farmers (Christy bapita *et al.*, 2007).

Due to the use of a wide variety of antibiotics, aquaculture has been implicated as potential environment to the development and selection of resistant bacteria and a source of these pathogens to other animals and humans (Ahilanet *al.*, 2010). Some bacterial fish pathogens are also associated to diseases in humans, making the aquaculture products a potential risk to the customers (zoonotic or food borne diseases) (Turkeret *al.*, 2009).

Herbal drugs can be used not only as remedies but even more so, as growth promoters, stress resistance boosters and preventatives of infections (Ravikumaret *al.*, 2011). Hence, herbal drugs in disease management are gaining success, because they are cost effective, eco-friendly and have minimal side effects (Pandey and Madhuri, 2010; Ravikumaret *al.*, 2011). Although herbal remedies have been with us for human therapy for millennia, there has been relatively little research on the medicinal plants to be used against fish diseases (Abdul Kader Mydeen and Haniffa, 2011; Turkeret *al.*, 2009). There is now a fast growing interest in screening antiparasitic substances from plants to replace chemical and antibiotic alternatives (Nargiset *al.*, 2011).

Oromia is one of the leading producers of freshwater fish in the country. In spite of the tremendous potential production, fishery sector has been suffering from outbreak of several diseases. Fish farmers of the region have been experiencing about huge production loss due to diseases and poor management in freshwater aquaculture sector (Unpublished data, 2018). The aquaculture sector in Ethiopia in general and Oromia in particular is still deprived of the developments in fish health and disease management practices. Thus searching for best alternative to control the disease of cultured fish is very crucial. This study was conducted with the objective of evaluating some medicinal plants to manage fish diseases

Materials and Methods

Plant Collection:

Leaves of plants namely *Moringa Olifera*, *Rosmarinus officinale* and *Azadiridica indica*, claimed to be traditionally used against infection were collected from their natural habitat with the guide of traditional healers. Voucher specimens were deposited in the laboratory of the center.

Crude Extract Preparation:

Fresh plant materials were separately washed with tap water, shade dried, grinded and coarsely powdered using grinding machine. The pounded specimens were then be subjected to extraction using absolute methanol by maceration technique (Harikrishnanet *al.*, 2010). Methanol was used as solvent of choice for extraction because methanol is considered as the best solvent for the extraction of antimicrobial substances, it mimics the property of water which is commonly used as extraction solvent by traditional healers and the methanolic extracts contain diverse chemical compounds with biological activity (Nya and Austin 2009).

A total of 250g of the pounded materials were separately soaked in the extraction solvent (100 g of powder in 1000 ml of solvent proportion) followed by agitation periodically for three days and then filtered. The mixtures were first filtered using gauze and then the filtrate was passed through sterile filter paper (Whatmann number 1). This was repeated three times to allow the solvent extract substantial quantities of the chemical constituents from the pounded plant materials. Then the filtered extract was dried in hot air oven at a temperature of 65⁰C. The resulting extracts were then be transferred into well labeled vials and kept in 4⁰C until required for use.

In vitro antibacterial activity test of crude extracts:

To evaluate the antimicrobial activity of the individual crude extracts, the antibacterial agar well diffusion assay were employed following the methods described and used by different works (Winkaler *et al.*, 2007, Sharma *et al.*, 2010, Ravikumar *et al.*, 2010, Oliveira and Figueiredo, 2008). The standardized bacterial broth culture was streaked evenly on sterile Muller Hinton agar (MHA) plates with cotton swab. After thirty minutes, on each plate, equidistant wells were made with a 6mm diameter sterilized cork-borer. The labeled wells were filled with 100 μ l of 500, 250, and 125 mg/ml of test extracts. For comparison, gentamycin (25 μ g/ml) and sterile distilled water (100 μ l/well) was used as a positive and negative control respectively. Then, the plates were allowed to stand on laboratory bench for 2hrs to allow proper diffusion of the extracts into the media. Finally, the plates were incubated at 37⁰C for 24 hrs. After incubation, the resulting diameters of zones of inhibition, including the diameter of the well, was measured and reported in millimeter (mm) and the mean of zones of inhibition was calculated for each test extract and the standard antibiotics.

Determination of Minimum Inhibitory Concentration: The minimum inhibition concentration (MIC) of individual plant extracts were determined according to the method described by Chung *et al.*, (2011). MIC was determined for extracts that showed growth inhibition diameter of ≥ 10 mm at 500 mg/ml concentration. Two-fold serial dilutions of the extracts were made with nutrient broth. Extract solution of 250 mg/ml was serially diluted in ten test tubes to the concentrations of 250, 125, 67.5, 38.75, 19.38, 9.69, 4.84, 2.42, 1.21 and 0.60 mg/ml. The tubes were added with 1ml of the microbial suspension and incubated at 37⁰C for 24 hr. The control tubes did not have test extract, but contained the test bacteria and the sterile distilled water used to dissolve the extracts. After incubation, the visual turbidity was observed and recorded. The lowest concentration in which the turbidity was observed was measured as an MIC of the individual extracts.

Fish and experimental design:

Healthy Nile tilapia, *Oreochromis niloticus* (weight 19 \pm 3.8 g, Ns: 288) obtained from our research center was maintained in 24 water tank (150L) aerated recirculation fresh water system. Each water tank was stocked with twelve fish and experiment was conducted with duplicate. Fish in each tank was fed with 0% (control), 2.0%, 3.0% and 4% crude extracts of medicinal plants at the rate of 5% of their body weight twice a day till the end of experiment.

Immunological and Hematological examination:

Blood was collected from four fish in each experimental group at first and second weeks of post infections for hematological and immunological assays following the method of Sarder *et al.* 2001.

Challenge test and bactericidal activity:

At the end of the study, (On 60th day of feeding), all fish was injected intraperitoneally (i.p.) with 100µl phosphate buffered saline (PBS) containing *A. hydrophila* at 3.7×10^7 cfu ml⁻¹ according to Yin *et al.*, 2008. All groups were kept under observation for 15 days to record any abnormal clinical signs and the daily mortality rate. Bactericidal activity in fish samples were analyzed according to the Miles–Misra technique (Okada *et al.*, 1999) to check viable counts by agar plate-spread method on Tryptic Soy Agar (TSA) 24hr after incubation.

Growth performance: Growth performance of treated fish was assessed by determining percentage weight gain (WG), specific growth rate (SGR) and feed conversion ratio (FCR) according to Choudhury *et al.* (2005). Growth response parameters were calculated as follows:

- **Mean Weight gain (MWG, %)** = $100 * [(final\ mean\ body\ weight - initial\ mean\ body\ weight) / initial\ mean\ body\ weight]$;
- **Specific Growth Rate (SGR, % /day)** = $100 * [(In\ W_t - In\ W_i) / T]$, where W_t is the weight of fish at time t , W_i is the weight of fish at time 0 and T is the rearing period in days;
- **Feed Conversion Rate (FCR)** = Total dry feed fed g/ fish / total wet weight gain g/fish.

Statistical analysis:

Values for each parameter measured were expressed as the arithmetic mean \pm standard error (SE) by using SPSS software. Effect of herbal diets on hematological, immunological and growth parameters were tested using two-way ANOVA and the mean values were compared at the 5% level of significance.

RESULTS

In vitro antibacterial activity and MIC test:

The evaluation of the antimicrobial activity of the individual crude extracts revealed that the plant extracts produced a dose dependent increase in the zone of inhibition from 100µl concentration and MIC was recorded as 19.38mg/ml, 67.5mg/ml and 67.5mg/ml for *M. olifera*, *A. indica* and *R. officinale* respectively (Table 1).

Table 1: Antibacterial activity and Minimum Inhibition Concentration test of crude extracts

Bacteria Used	Extract Conc.(mg/ml)	Plant Spp.	Inhibition zone(mm)	MIC (mg/ml)
<i>A. hydrophila</i> (3.7×10^7 cfu)	0	<i>M. olifera</i>	0.00 \pm 0.00*	-
		<i>A. indica</i>	0.00 \pm 0.00*	-
		<i>R. officinale</i>	0.00 \pm 0.00*	-
	125	<i>M. olifera</i>	17.31 \pm 0.70**	-
		<i>A. indica</i>	12.00 \pm 0.31*	-
		<i>R. officinale</i>	10.11 \pm 0.43*	-
	250	<i>M. olifera</i>	18.23 \pm 0.30**	-
		<i>A. indica</i>	14.21 \pm 0.10(**)(*)	-
		<i>R. officinale</i>	11.67 \pm 0.47*	-
	500	<i>M. olifera</i>	37.20 \pm 1.43**	19.38
		<i>A. indica</i>	17.10 \pm 0.00*	67.5
		<i>R. officinale</i>	16.33 \pm 1.52*	67.5

Note: The result with ** in the same column of single row is significantly different at CI of 95%.

Immunological and Hematological examination:

Hematological assays indicated that hematocrit and RBCs counts were significantly increased in fish on 2.0% supplementation diet with *Azardica Indica* compared to other group. On the other hand fish fed on diets containing 4% of *Moringa Olifera* incorporated diet show significant increment of hematocrit and RBCs counts as compared with *Rosemary Officinale*, *Azardica Indica* incorporated diet. The low counts of RBCs were obtained at the control diet (Table 2).

Table 2: Hematological parameters of Nile tilapia fed diets containing different levels of experimental plants

Parameters	Doses	Plant Species		
		<i>Rosemary Officinale</i>	<i>Azardica Indica</i>	<i>Moringa Olifera</i>
Haematocrit	0%	22.5±1.2a	23.4±1.7	23.9±1.0a
	2.0%	25.3±0.4	29.9±2.9**	25.2±1.3
	3.0%	26.2±1.9	24.4±0.4	25.7±1.8
	4.0%	27.5±0.60	24.6±2.2	30.7±1.1**
RBC (x10 ⁶ /μL)	0%	1.66±0.087	1.62±0.079	1.59±0.127
	2.0%	1.96±0.181	2.78±0.158**	1.99±0.721
	3.0%	2.22±0.153	2.18±0.000	2.54±0.127
	4.0%	2.37±0.127	1.92±0.043	2.94±0.138**

Note: The result within the row with ** shows significant different at CI of 95%.

Similarly immunological analyses indicated that lymphocytes and neutrophils were significantly increased in fish on 2.0% supplementation diet with *Azardica Indica* as compared to other group. Fish fed on diets containing 4% of *Rosemary Officinale*, *Azardica Indica* and *Moringa Olifera* incorporated diet exhibited 4.37±0.643, 4.21±0.712 and 5.91±0.0362 WBCs counts respectively which shows fish fed on diets containing 4% of *Moringa Olifera* incorporated diet shows significant increment of WBCs counts as compared with *Rosemary Officinale* and *Azardica Indica* incorporated diet. The low counts of WBCs were obtained at the control diet (Table 3). Perversely, the control diet produced the highest monocytes and granulocytes, which decreased with the increase of herbal levels in fish diet ($p < 0.05$).

Table 3: Immunological parameters of Nile tilapia fed diets containing different levels of experimental plants

Parameters	Doses	Plant Species		
		<i>Rosemary Officinale</i>	<i>Azardica Indica</i>	<i>Moringa Olifera</i>
Neutrophils	0%	0.6±0.04	0.6±0.03	0.7±0.04
	2.0%	1.1±0.04	1.5±0.05**	0.8±0.06
	3.0%	0.7±0.02	0.8±0.04	1.4±0.05
	4.0%	0.9±0.06	1.1±0.04	1.8±0.03**
Lymphocytes	0%	21.3±1.2	22.4±1.1	22.6±1.2
	2.0%	27.5±1.5	31.8±1.2**	24.1±1.4
	3.0%	22.6±1.0	24.1±1.4	29.5±1.4
	4.0%	24.7±1.4	27.8±1.3	36.4±1.2b**
WBC (x10 ⁵ /μL)	0%	2.94±0.124	2.71±0.133	3.29±0.176
	2.0%	3.39±0.098	4.97±0.102**	3.52±0.133
	3.0%	3.94±0.136	3.73±0.127	4.17±0.700
	4.0%	4.37±0.643	4.21±0.712	5.91±0.0362**
Monocytes (%)	0%	11.2±0.18	9.9±0.16	10.6±0.23
	2.0%	5.6±0.15	6.6±0.15	6.5±0.19
	3.0%	3.8±0.06	4.2±0.80	3.9±0.08
	4.0%	1.4±0.61	1.3±0.71	1.2±0.82
Granulocytes (%)	0%	7.4±0.13	5.4±0.13	4.4±0.35
	2.0%	4.8±0.03	4.1±0.00	2.8±0.39
	3.0%	3.2±0.16	1.3±0.13	0.9±0.06
	4.0%	1.3±0.06	1.1±0.26	0.3±0.05

Note: The result within the row with ** shows significant different at CI of 95%.

Challenge test and bactericidal activity:

The serum bactericidal activity was significantly high in fish fed with 2.0% supplementation diet with *AzardicaIndica* and 4% of *Moringaolifera* against pathogen as compared with similar concentration. The lowest number of bacterial colonies indicated the efficiency of immune cells in serum to kill the pathogen. The bacterial count after incubation with fish sera decreased with increase of herbal level in fish diets. The lowest bacterial count was obtained in fish fed 4% *MoringaOlifera*, whereas the highest one was obtained when fish fed control diet (Table 4).

Table 4. The total counts of *bacteria* 24hr after incubation with serum of Nile tilapia.

Parameters	Doses	Plant Species		
		<i>Rosemary Officinale</i>	<i>AzardicaIndica</i>	<i>MoringaOlifera</i>
Bacterial count (x10 ⁵ cell)	0%	87.7±3.28	85.7±2.32	86.3±3.57
	2.0%	68.0±1.73	43.7±3.32**	51.1±0.73
	3.0%	61.2±1.73	57.5±2.31	39.1±2.32
	4.0%	44.6±1.65	43.3±2.03	33.2±2.03**

Note: The **in the same row is significantly different at CI of 95%.

The result of the study indicated that after challenged with *A. hydrophila* the mortality was 13% and 27% in fish fed with diet supplemented with 4% and 3% *MoringaOlifera* respectively. 4% dose diet showed the highest relative percentage of survivability (RPS) 87% compared to other dose diet in the experimental study. The mortality increased to 37% with 2% dose diet. The lowest mortality of 13% and a higher survivability rate of 87% were observed in fish fed with 4% dose diet supplemented with *MoringaOlifera* (Figure 1).

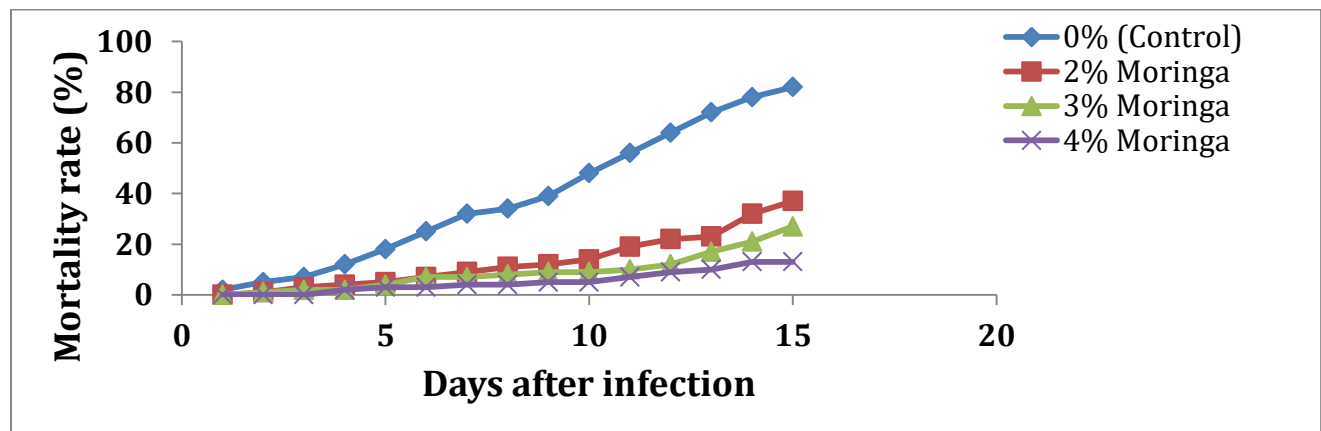


Figure 1. Mortality rate of Nile tilapia fed with different levels of *MoringaOlifera* in water tank

Similarly, fish fed with 4% and 3% supplementation diet with *Rosemary Officinale* showed the mortality rate of 25% and 29% respectively. 4% dose diet showed the highest relative percentage of survivability (RPS) 75% compared to other dose diet in the experimental study (**Figure 2**).

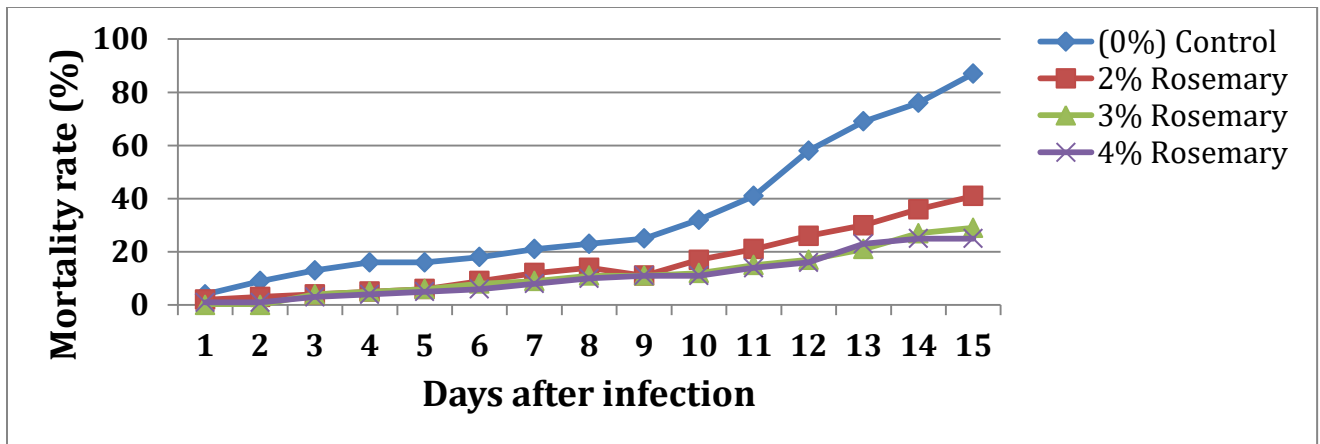


Figure 2. Mortality rate of Nile tilapia fed with different levels of Rosemary *Officinale* in water tank

On the other hand, relative percentage of survivability (RPS) was assessed for fish fed with different level of Azaridica *Indica* (Neem). Unlike the other 2% supplementation diets with Azaridica *Indica* showed the least mortality rate of 17%. This showed 2% dose diet showed the highest relative percentage of survivability (RPS) 83% compared to other dose diet in the experimental study. The mortality increased to 33% with 3% dose diet supplemented with Azaridica *Indica* (Figure3).

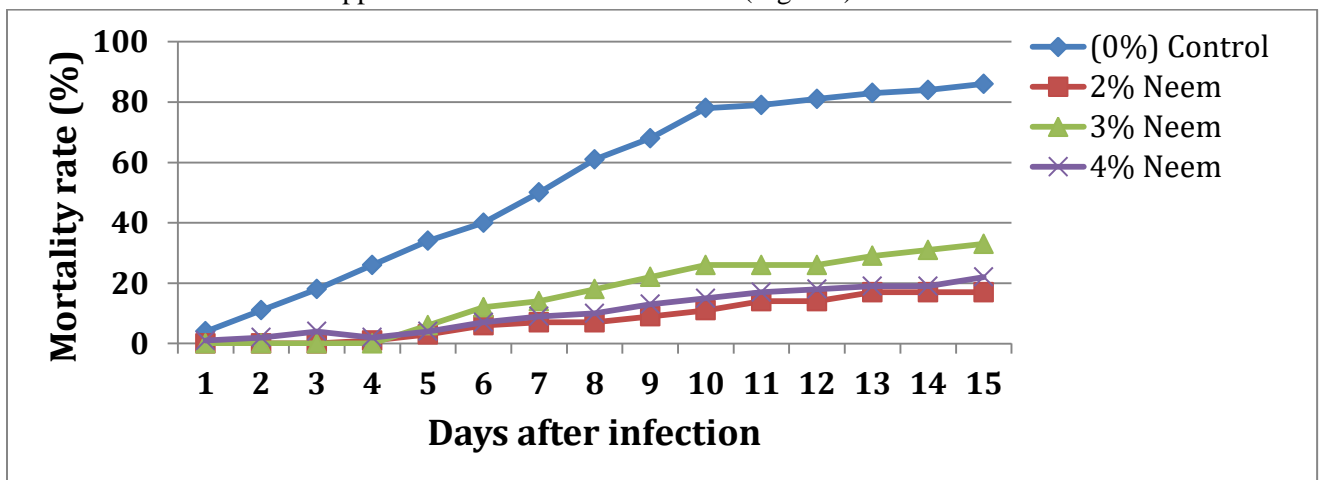


Figure 3. Mortality rate of Nile tilapia fed with different levels of Azaridica *Indica* in water tank

The effect of supplementation of different medicinal plant on relative percentage of survivability (RPS) was also assessed for fish fed with 4% of Rosemary *Officinale*, Moringa *Olifera* and Azaridica *Indica* to compare their effect at the highest dose. The finding revealed that Moringa *Olifera* at the dose of 4% supplementation with fish diet showed the least mortality rate of 13% followed by Azaridica *Indica* 22%. This showed 4% dose diet supplemented with Moringa *Olifera* showed the highest relative percentage of survivability (RPS) 87% compared to other incorporated diet in the experimental study (Figure4).

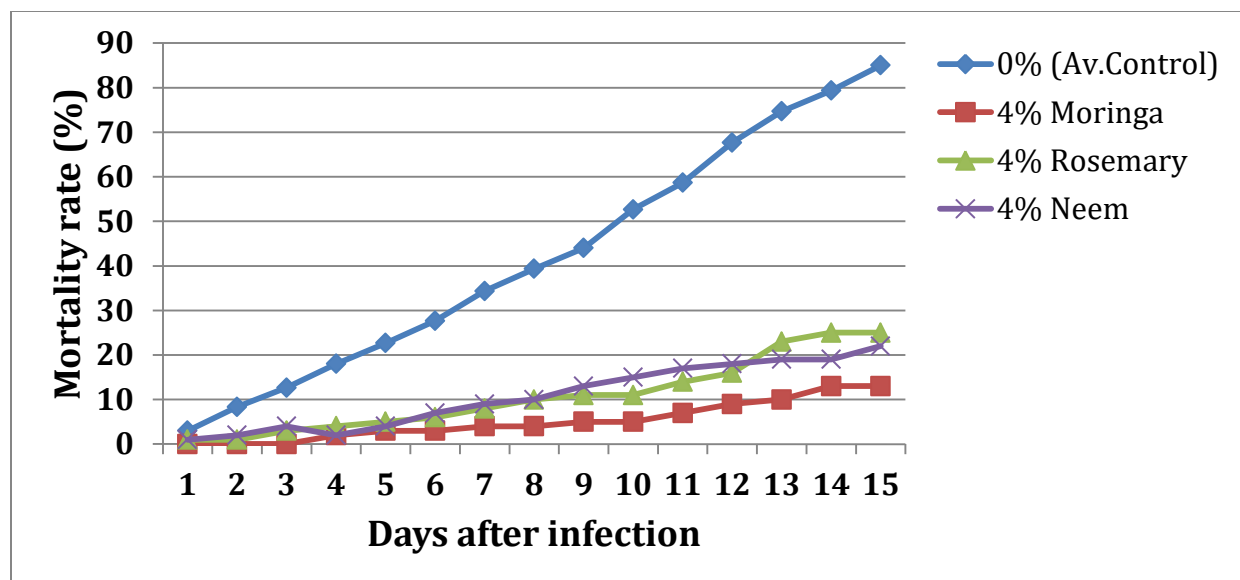


Figure 4. Mortality rate of Nile tilapia fed with 4% of experimental plants in water tank

Growth performance:

Growth performance of treated fish was assessed by determining percentage weight gain (WG), specific growth rate (SGR) and feed conversion ratio (FCR) and hence growth rate increased significantly on fish fed with 2.0% supplementation diet with *AzardicaIndica* and 4% of *Moringaolifera* as compared with similar concentration (Table 5).

Table 2: Growth parameters of Nile tilapia fed diets containing different levels of experimental plants

Variables	Doses	Rosemary <i>Officinale</i>	<i>AzardicaIndica</i>	<i>MoringaOlifera</i>
MWG	0%	28.3±1.3	29.1±1.2	30.4±1.5
	2.0%	30.4±1.4	46.3±1.3**	36.4±1.9
	3.0%	36.7±2.2	34.1±1.1	45.8±1.2
	4.0%	41.6±2.0	39.7±1.5	51.3±1.0**
SGR	0%	1.3±0.1	1.4±0.5	1.4±0.3
	2.0%	1.5±0.3	1.6±0.7	1.7±0.2
	3.0%	1.4±0.2	1.6±0.2	1.6±0.5
	4.0%	1.4±0.2	1.7±0.1	1.8±0.3
FCR	0%	1.6±0.4	1.8±0.3	1.5±0.2
	2.0%	1.5±0.4	1.7±0.4	1.7±0.3
	3.0%	1.6±0.2	1.7±0.2	1.8±0.2
	4.0%	1.1±0.3	1.3±0.2	2.0±0.4

DISCUSSION

Immunological approaches to prevent fish diseases have been normally used antibiotics, chemicals or vaccination against specific pathogens, while the use of immune stimulants is relatively a new and developing area (Jong et al., 1993). In the recent years, there is increasing interest in the use of herbal

extracts as dietary and therapeutic supplements indicate that modulate immune function in fish (Harikrishnan *et al.*, 2011). The present study indicates *Oreochromis niloticus* fed with all doses diet had significantly increased growth rate when compared to the control. However, SGR and FCR did not significantly increase with any diet. The Hematocrit, lymphocytes and neutrophils levels significantly increased with 2.0% supplementation diet with *Azadirachta indica* and 4% of *Moringa olifera* against pathogen as compared with similar concentration. The results are in agreement with Sumana *et al.*, 2015 which indicate after dietary administration with *Azadirachta indica* Chinese carp infected with *Aeromonas hydrophila* had shown similar effects.

In the present study, the finding revealed that *Moringa olifera* at the dose of 4% supplementation with fish diet showed the least mortality rate of 13% followed by *Azadirachta indica* 22%. This showed 4% dose diet supplemented with *Moringa olifera* showed the highest relative percentage of survivability (RPS) 87% compared to other incorporated diet in the experimental study. Labeorohita and *O. mykiss* fed with *Achyranthes aspera* or garlic (Nya *et al.*, 2011) and *Ocimum sanctum* leaf extracts administered through intraperitoneally injection in *Oreochromis mossambicus* (Logambalet *et al.*, 2000) also reduced the mortality rate against *A. hydrophila* infection. *L. rohita* fed with garlic enriched diet exhibited increased number of serum bactericidal activity (Sahu *et al.*, 2007). Sumana *et al.*, 2015 indicate after dietary administration with *Azadirachta indica* Chinese carp infected with *Aeromonas hydrophila* had shown similar effects.

According to the finding the serum bactericidal activity was significantly high in fish fed with 2.0% supplementation diet with *Azadirachta indica* and 4% of *Moringa olifera* against pathogen. These results indicate that these herbal supplementation diets might have induced other antimicrobial mechanisms, which include release of lysosomal enzymes, cationic peptides, complement components, and production of reactive oxygen species (Adams and Hamilton, 1984). Enhanced or elevated phagocytic activity has been reported in goldfish and rainbow trout after treatment with different herbals against *A. hydrophila* (Awad and Austin, 2008, Dugenciet *et al.*, 2003). The enhancement of phagocytosis observed in this study may be due to *Azadirachta indica* and *Moringa olifera*. In the present study the serum bactericidal activity significantly increased with any enriched diets at any time. Such an increase in the bactericidal properties in fish serum could be attributed to the reduced mortality (%) as was observed for 15 days post-infection.

In the present study the stimulation of specific immune defense were observed and this might be due to the presence of one or more than one components present in *Azadirachta indica* and *Moringa olifera*. The present result has confirmed the maximum response with 2% *Azadirachta indica* and 4% *Moringa olifera* enriched diet against *A. hydrophila* which might suffice to activate the receptors and the corresponding genes responsible for the secretion of immune defense factors.

CONCLUSION AND RECOMMENDATIONS

In the light of the overall results obtained in the current study, it is clearly indicated that extracts of 2% *Azadirachta indica* and 4% *Moringa olifera* has great potential as antibacterial compound. It is anticipated that this herb can be potentially used in combating fish bacterial diseases especially for *haemorrhagic septicemia* infection. However, further study on safety and toxicity are warranted. Nowadays, most pathogenic organisms are becoming resistant to antibiotics. The present study also suggested the

potential of some herbs as an alternative to commercial and synthetic antibiotics, which could be used in aquaculture industries.

The present study also opens up new vistas of research to assess the most effective dose under field conditions, experimentation with a recommended dose of purified extract of *Azadirachta indica* and *Moringa olifera*, degree, and duration of the resistance offered, an administrative regime for different age group of fish and time of application to ensure improved harvest in culture ponds.

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Characterization of Four Local *Oreochromis niloticus* Strains for their Better Production Traits in Concrete Tanks at Batu, Oromia, Ethiopia

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Abstract

*This experiment was conducted to characterize four Nile tilapia (*Oreochromis niloticus*) strains collected from Chamo, Koka, Zeway and Hawassa lakes for their better production traits under concrete tank culture system. Mixed-sex fish fry from age of yolk sac completion to 90 days were fed 40% CP formulated feed at 10% body weight twice a day, 5 days a week. After the age of 90 days to 240 experimental days, the fish were provided with formulated feed of 30% CP at 3% body weight. Final body weight and Daily Growth Rate (DGRg/d) of Chamo, Koka, Zeway and Hawassa *O. niloticus* strains were 136.27±6.23 g and 0.56 g/d; 86.00±4.61 g and 0.35 g/d; 78.05±4.29 g and 0.32 g/d; 75.26±4.20 g and 0.30 g/d respectively. This study revealed that, the mean final body weight of Chamo strain was significantly ($P < 0.05$) higher than that of Koka, Zeway and Hawassa strains by 36.90%, 42.72% and 43.85% respectively but no significant ($P > 0.05$) difference between the later three strains. Chamo, Koka, Ziway and Hawassa strains attained survival rate of 81.4%, 80.0%, 72.3% and 74.3% and FCR of 2.02, 2.18, 2.24 and 2.27 respectively. Finally, it was concluded that mixed-sex of *Oreochromis niloticus* of Chamo strain attained best growth performance, survival rate and feed conversion ratio followed by Koka.*

Key Words: Concrete tank, growth performance, mixed-sex, Nile tilapia.

Introduction

Nile tilapia (*Oreochromis niloticus*, Linnaeus) is one of the tilapia species which widespread in the world as a result of tilapia farming (Pauly, 1976, Balarin, 1979, Witte and Wim van Densen, 1996, Berg, 2005). This native fish species to Africa is also found in various Ethiopian water bodies lakes, rivers, reservoirs and ponds (Abebe Getahun, 2017). Tilapia species are hardy, high yield potential and able to survive at low oxygen tension and wide range of water temperature for optimal growth and reproduction (Wohlfarth and Hulata, 1981; CTA, 1996; Berg, 2005). Mouth brooders, the Nile tilapia has low fecundity, high parental care to ensure that the majority of eggs will survive to the juvenile stage (CTA, 1996; Mark *et al.*, 2004). Tilapia culture provides home-grown animal protein, diversified income, job opportunity and compensating the fish supply in depleted wild capture (Sosa *et al.*, 2005; Mandal *et al.*, 2009; Palipoch *et al.*, 2011). As a result, *Oreochromis niloticus* accounted more than 81% of the total cultured tilapias globally (Suresh, 2003). To realize the potential of Nile tilapia culture, development of adapted farm races of this species should be part of the genetic improvement programs (Eknath *et al.*, 1991; Bentsen and Gjerde, 1994). Identification of relatively fast growing individuals early in their life cycle is an important consideration in genetic selection programs in aquaculture management (Wickins, 1987). Selection and

characterization of best performing breed is required for establishment of hatchery which enhance aquaculture development in Ethiopia. There is no comparative data available on characterization of different local strains of *Oreochromis niloticus* under controlled condition in Ethiopia.

Therefore, the study was aimed to characterize, *Oreochromis niloticus* strains of four lakes (Chamo, Hawassa, Koka and Zeway) for their desired quality traits, such as better growth performance, survival rate and food conversion efficiency under concrete tank culture system on station at Batu Fish and Other Aquatic Life Research Center.

Materials and Methods

Description of the Study Area

The characterization study of four local strains of *Oreochromis niloticus* was carried out between December 2015 and October 2018, particularly the fish growth performance was conducted in between March 2018 and October 2018 on station at Batu Fish and Other Aquatic Life Research Center located at 7.918⁰N and 38.727⁰E at an altitude of 1650 m.a.s.l. The center is situated in Batu town on South-West shore of Lake Ziway/Danbal within the Ethiopian rift valley system characterized by semi-arid agro ecology. The site is about 160 km far from capital city Addis Ababa to south direction in Batu town at the side of Addis- Hawassa asphalt road in Adami Tulu district, Oromia regional state.

Through experimental period from December 2015 to October 2018, positive and negative important characters and effects were observed. Physical, biological and behavioral data such as air and water temperature, strains growth performance, survival rate, sex ratio, feed conversion ratio, brood spawning behavior were recorded.

Temperature

During the experimental period, assessing climate conditions which could have influences on fish growth, reproduction and survival rate is a crucial issue. To evaluate and realize its effects on fish biological parameters, atmospheric and pond water temperature data were collected for 3 to 5 days every month, morning from 8:30 - 9:00 AM. and afternoon from 3:30 - 4:00 PM throughout the experimental period.

Brooders fish collecting, stocking and feeding

Total of 12 concrete experimental tanks each having 35 m² surface area and 1.20 m water depth were prepared and filled with well water. Wild *Oreochromis niloticus* juveniles were collected from four different Lakes (Chamo, Hawassa, Koka and Zeway) in December 2015 and stocked separately according their origin into the prepared experimental tanks to use them as parent stocks in the following experiment. The acclimatized and stocked juveniles were reared until they became sexually mature for breeding. Ground/well water after exposed to open air and settled in reservoir was used to fill and exchange water in experimental tanks. The fish tanks were regularly recharged with water every 2 to 3 days.

After the fish became adult, six males and six females with good condition were selected from each of the four strains and kept strain and sex separate in suspended hapa/mosquito nets in the concrete tanks for conditioning period of 2-3 weeks. The fish in the conditioning were fed with various ingredients in powder form (grinded mechanically using mortar and pestle and uniformly mixed manually). The prepared feed of 22.5% CP was comprised of wheat bran and Noug cake (1:1ratio) kept in plastic sac and provided for brooders at 2% body weight twice a day morning at 9.00 AM and afternoon at 3.30 -4.00

PM). The conditioned fish were checked for spawning readiness, specially by checking the female's genital papilla, by visual examination of morphological characteristics, choosing pink to red and yellow color with protruding (GIFT Technology Manual, 2004).

Following single pair mating method (GIFT, 2017), the selected and conditioned fish in each of the four strains were stocked as a single mating pair of one male and one female into the prepared breeding hapa of dimensions 1.60 X 1.80 X 1.50 m= 4.32 m³ each.

Progenies production and rearing

The present growth performance data of *Oreochromis niloticus* strains was collected from the experiment conducted in between March 2018 and October 2018 under concrete tank culture system. During spawning period, each hapa stocked with the single pair brooders were frequently checked for newly born fry in five days interval. During checking period, if individual females observed with fertilized eggs/larvae in mouth or swimming fry, it was recorded with clear description and situation. Based on observed report, when the fries completed yolk sac absorption, feeding fries with external feed was started with formulated feed made of 70% fish meal, 20% Wheat bran and 10% Noug cake containing 40% CP.

The mixed powder feed was moisten with water and provided two times a day, morning at 9:00 AM and afternoon at 3:00 PM for 5 days in a week. Broods/parents were removed and transferred to their original tanks 21 days after fry observed. Then, at the age of 23 - 25 experimental days, the fry were sampled from hapa nets and their total length (TL in cm) and total weight (TW in g) measured. After the sampling, the fry were transferred to the rearing tanks each having 35 m² surface area and 1.20 m water depth at the stocking density rate of 2 fish/m². In rearing tanks, fries were fed with the feed quality of 40% CP to 65 rearing days at 10% body weight two times a day morning at 9:00 AM and afternoon at 3:00 PM for 5 days in a week.

The daily ration was calculated and adjusted regularly according to the weight gain of the fish every month. After the age of 90 to 240 experimental days the feed protein content of the fish reduced to 30% CP, prepared from 30% fish meal, 39.5% wheat bran and 30% Noug cake was offered at 3% fish body weight 5 days a week morning at 9:00AM and afternoon at 3.30PM.

Fish sampling and data collection

Monthly fish data of Total Length (TL) in cm using measuring board to 1 mm and Total Weight (TW) in gram using sensitive balance to a minimum 0.01 g were recorded by collecting the fish from the experimental tanks sampled using beach seine having 1 cm mesh size from each tank. After the fish were caught and kept in bucket filled with water, 25 -35 random fish samples were measured from each strain and replication, after which they are returned back to their corresponding tanks. At the end of the experiment, fish of all tanks respecting their strains and replication were totally harvested and their data separately treated for each individual.

Fish growth analysis

The monthly collected fish data for the parameters of Daily Growth Rate (DGRg/d), Specific Growth rate (SGR%/d), survival rate (%), sex ratio (%) and feed conversion ratio were fed in to computer in SPSS and Microsoft excel programs in order to perform statistical analysis. The mean weights of the fish were

analyzed using one way analysis of variance (ANOVA). The difference in mean of the fish weight among the populations was analyzed using one way ANOVA and further mean separation subjected to significance testing using Tukey test. Statistical significance was determined at $P < 0.05$. The following parameters were analyzed using the appropriate designed formulas:

$$\text{Daily Growth Rate (g /day)} = \frac{\text{Mean Final Weight (g)} - \text{Mean Initial Weight (g)}}{\text{Experimental Days}}$$

$$\text{Specific Growth Rate (SGR \% /day)} = \frac{\ln(\text{Final weight (g)}) - \ln(\text{Initial weight (g)})}{\text{Culturing days}} \times 100$$

$$\text{Feed Conversion Ratio (FCR)} = \frac{\text{Feed intake}}{\text{Weight gain}}$$

$$\text{Survival rate (\%)} = \frac{\text{Number of fish harvested}}{\text{Number of fish stocked}} \times 100$$

RESULTS

Temperature:

As the result illustrated, the atmospheric temperature ranged from 16-30°C while the pond water temperature ranged from 16-26 °C (Fig. 1) with a great temperature fluctuation due to season (cold or warm days/season, rainy, cloudy and windy days, early morning and at evening). The maximum temperatures were in April both for in air and pond water.

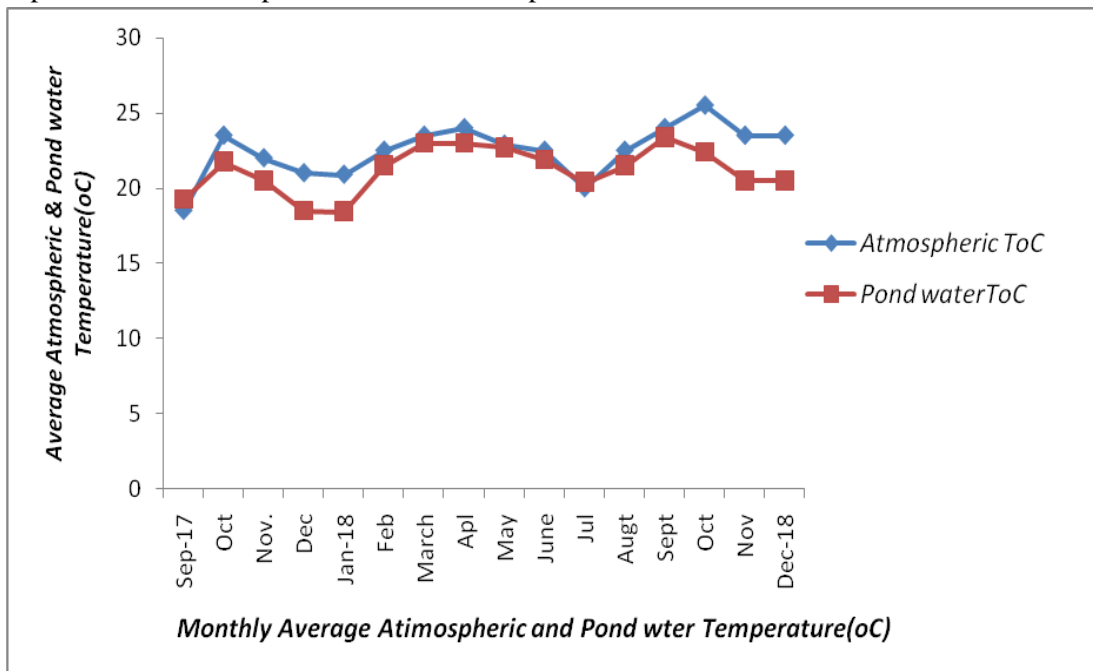


Fig. 1. Average air and pond water temperature at Batu during the experimental period..

Spawning behavior:

When single pair of male and female *O. niloticus* breeders were kept in hapa net for spawning, the males became aggressive to their female partners in Chamo, Koka, Zeway and Hawassa strains. The aggravation of males as 'sexual harassment' was deadly serious and frequently observed in Chamo strain. The 'sexual harassment' was frequently occurred in the single mating pairs in hapa regardless of the fish size and the female's ripeness where smaller sized males can attack bigger sized and ripe females. The attack on females was by biting, scratching, chasing/striking, crashing and pushing causing damages of females' body (skin, scales, fins and eyes) thereby finally the female fish wounded and die out (Fig. 2). Unless trimming/removing the upper part of the lip of the male, a single pair mating trial in narrow space is valueless. To reduce the aggravation of males over females in single pair mating within a limited space, stocking one or two additional unripe females along with the ripe female may be another remedy.



Fig. 2 Female *O. niloticus* killed by their male partners in hapa net during mating

Fish body deformity:

Aquaculture fish with body deformities can be problematic because they can affect fish marketability. Fish body deformity/growth abnormalities were observed in Koka and Chamo strains. The body deformity rarely observed in Koka *O. niloticus* strain was at the caudal part of the body (Fig. 3a & 3b). The fish sex organ and tail were clearly observed on each deformed fish. Sometimes the deformed females with fertilized egg in their mouth were observed (Fig. 3c).



Fig. 3 Body deformities observed in *O. niloticus* strains.

Growth abnormality/body deformity occurred in Chamo *O.niloticus* strain were on dorsal fin (Fig. 3e) and caudal peduncle (Fig. 3d). In this strain, individuals with dorsal fin's anterior spines missing and caudal peduncle part deformed and not developed were frequently observed. This problem was usually observed in the population during the four years characterization trials. Though heritability not experimentally checked, the body deformities which were observed in both strains can be genetically heritable because the type of deformity observed on one strain was not seen on other strain for four years. These body deformities have negative influence on fish market.

Growth performance:

After 240 experimental days in concrete tanks, growth performances of the four local *Oreochromis niloticus* strains (Chamo, Koka, Ziway and Hawassa) was presented as total length (cm), body weight (g), daily growth rate (DGR, g/d) specific growth rate (SGR, %/d) in table (Table 1). The mean of final total length, body weight, DGR and SGR were higher in Chamo strain followed by Koka..

Table 1. Growth performance of four local strains of mixed-sex *Oreochromis niloticus* in 240 experimental days at Batu.

Strain	Total length (cm)		Body weight (g)		DGR (gd ⁻¹)	SGR (%/d)	Sex ratio % (M:F)	Survival rate(%)
	Range	mean	Range	mean				
Chamo								
-Mixed sex	16.5-23.0	20.2	73.0-199.0	136.28±6.23b	0.56	3.11±0.05	1 : 0.78	81.43
-Male		21.5		161.32±4.07	0.66			
-Female		18.1		96.93±5.70	0.39			
Koka								
Mixed sex	14.0-20.0	17.8	37.0-123.0	86.00±4.61a	0.35	2.90±0.06	1 : 0.51	80.00
-Male		19.1		103.11±1.91	0.42			
-Female		15.3		51.82±3.68	0.21			
Ziway								
-Mixed sex	14.5-19.6	17.2	43.0-112.0	78.05±4.29a	0.32	2.89±0.06	1: 0.92	72.86
-Male		18,4		93.44±2.51	0.38			
-Female		15.3		51.90±2.90	0.21			
Hawassa								
-Mixed sex	13.8-19.5	16.8	35.0-109.0	75.26±4.02a	0.30	2.87±0.06	1: 0.92	74.28
-Male		18.1		90.96±2.47	0.37			
-Female		14.8		51.00±3.08	0.21			

The range of final fish body weight at harvest was wide for Chamo strain which ranged from 73-199 g; while it was relatively narrow for Ziway strain (Table 1). The mean final weight of Chamo *O. niloticus* strain was significant ($P < 0.05$) higher than that of Koka, Ziway and Hawassa strains. There was no significant difference ($P > 0.05$) in mean final weight among Koka, Ziway and Hawassa strains. The mean final body weight of Chamo strain at harvest was higher than that of Koka, Ziway and Hawassa strains by 36.90%, 42.72% and 43.85% respectively but no significant difference between the later three strains

There was no significant ($P > 0.05$) difference among all *O.niloticus* strains in survival rate and sex ratio of Chamo, Koka, Zeway and Hawassa (Table 1.).

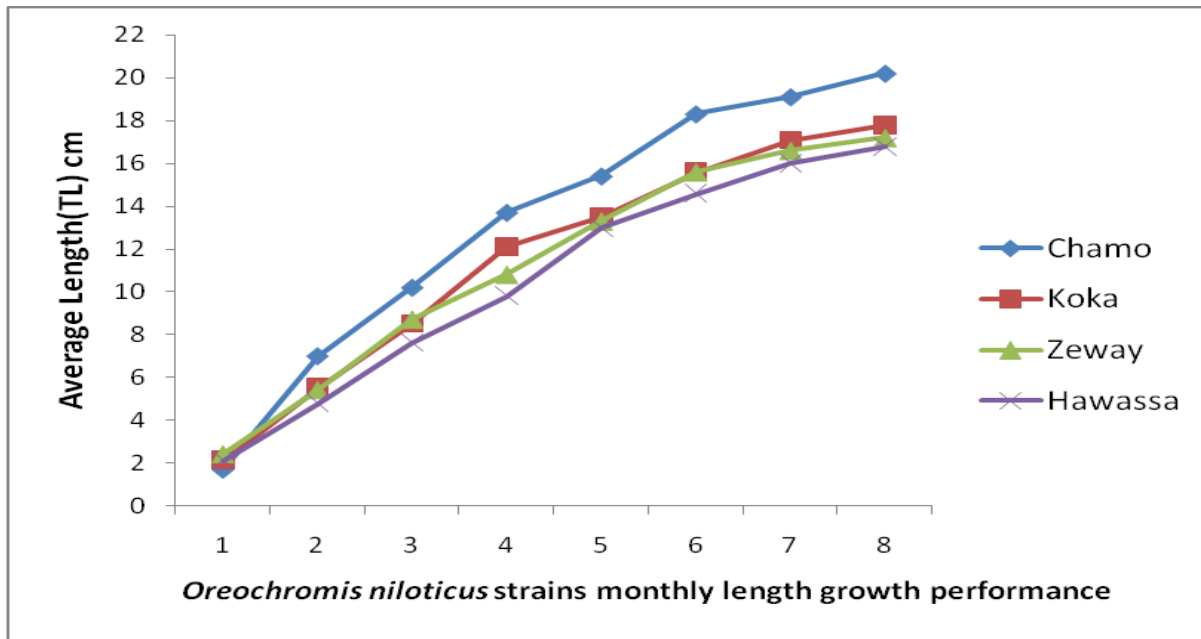


Fig. 2a. Monthly mean lengths of *Oreochromis niloticus* strains .

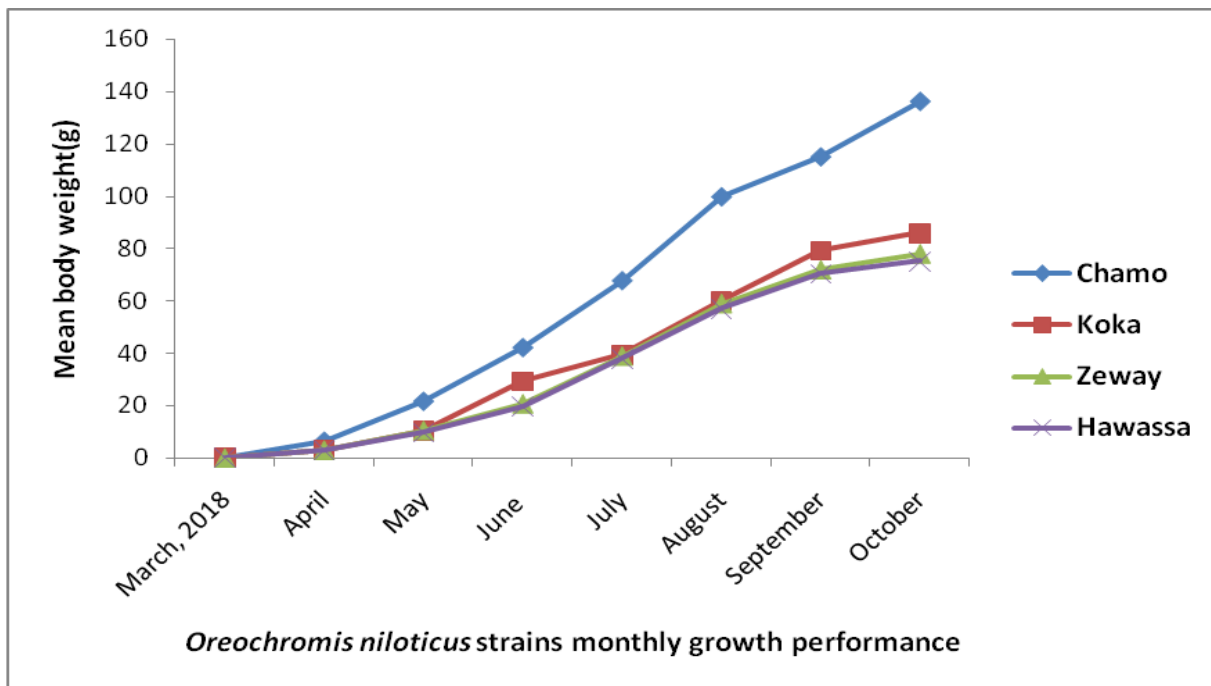


Fig. 2.b. Monthly mean weight of four local *Oreochromis niloticus* strains.

Feed conversion ratio (FCR) was analyzed for each strain. Chamo strain was more efficient in feed utilization (FCR of 2.02) whereas Koka, Ziway and Hawassa strains were lower in conversion in efficiency (higher FCR of 2.18; 2.24 and 2.27 respectively).

Discussion

In this study, characterization in terms of growth performance, survival rate, sex ratio, feed conversion efficiency and physical body deformity of local *Oreochromis niloticus* strains were the priority issues in order to select and recommend appropriate strains for fish culture development in the region.

Male *Oreochromis niloticus* aggravation during single pair (1M:1F) mating was observed in Chamo, Koka, Zeway and Hawassa strains while the aggravation was frequently observed on Chamo strain which was a challenging phenomenon during spawning period in hapa nets. As some authors have reported on *Oreochromis niloticus* aggressive behavior, it can be observed during spawning period due to atypical feminized males (AFM), which are normally characterized by having either abnormal genital papilla or male papilla and with ovaries (Morales *et al.*, 2015) which was not the case in Chamo males.

Body deformity/growth abnormality of *Oreochromis niloticus* was observed on both Koka and Chamo strains on different body parts. The body deformity rarely observed on Koka *O. niloticus* strain was on caudal part of the body. Though deformed, the fish's genital organ and their tail parts were differentiated and clearly observed on each deformed fish. These deformed fishes can breed that sometimes mature deformed females with fertilized egg in their mouth were observed. Growth abnormality in Nile tilapia can be caused by heredity, bacterial infection, fungus and/or polluted water and cause tailless, missing the caudal peduncle and the caudal fin (Tave *et al.*, 2011). The cause for fish body deformity observed in the current study was not clearly identified but it was more likely inherited genetically as the forms were specific to the strains.

The growth performance of *Oreochromis niloticus* depends on genetic materials, food quality, energy content of the food, stocking density and environmental factors (Abdel-Tawwab, 2004, Ashagrie Gibtan *et al.*, 2008, Kassaye Balkew *et al.*, 2012). The specific growth rate of mixed-sex *O. niloticus* under tank culture which ranged from 2.87 ± 0.06 to $3.11 \pm 0.05\%$ /day in the present study was comparably higher than the specific growth rate reported by Abdel-Tawwab (2004) with a range from 0.967% to 1.277% /day for strains of Abbassa, Aswan, Manzalah and Maryut in Egypt; Ashagrie Gibtan *et al.* (2008) with SGR range of 0.787% to 1.035% /day in cage culture system in Lake Kuriftu, Ethiopia; and Kassaye Balkew *et al.* (2012) with SGR range from 2.41 to 2.73%/day in pond culture, Sebeta, Ethiopia for strains of Koka, Ziway, Hawassa and Hora. The experimental fish in the current study were smaller in size and younger in age than those in the references that it contributed to the higher SGR results in the current study; the younger the fish the faster its growth rate. The variation in SGR results among the four strains in the present study was likely attributed to genetic differences among the strains because the experiment was run under similar culture system and similar management practices for all the strains. Similar observations were reported that growth performance of *Oreochromis niloticus* is affected by strain differences (Abdel-Tawwab, 2004; Ridha, 2006; Kassaye Balkew *et al.*, 2012). In general Chamo strain was with the best growth performance followed by Koka; while Zeway and Hawassa strains were ranked at the third and fourth steps. Survival rate of the fish was affected by internal and external factors whereby *O. niloticus* offspring below the age of three months at fry stage, when exposed to transfer to large tanks or ponds are susceptible to changes in water quality, temperature, presence of insect and other predators. Fry should be transferred to nursery ponds or tanks and stay there until they grow and pass the fry stage for better management.

In the case of the present study, the newly born fry at the age of 25 days were counted, sampled, measured (TL and TW) and stocked into concrete tanks at mean body size range from 2.08 cm and 0.074

g to 2.4 cm and 0.077 g, the stage at which mortality cases are relatively higher. The present study was exhibited the fish survival rate range from 72.86 to 81.43% which may be at least approach to the report by Tokuma Negisho *et al.* (2017). Survival rate was not fair to compare with report of Abdel-Tawwab (2004) where fingerlings with body size of 10 to 15 g gained 93.3 ± 2.04 to $100\pm 0.0\%$ survival; Kassaye Balkew *et al.* (2012) where fingerlings of mean length 9.18 cm and weight 12.48 g achieved 96 to 100% survival rate in concrete pond and Tokuma Negisho *et al.* (2017) where fingerlings of length 12.5 cm and weight 33.3 g achieved 80 ± 0.03 to $91\pm 0.03\%$ survival rates in concrete tanks. .

Feed conversion ratio (FCR) of the present study ranged from 2.02 to 2.27. This result partially agreed with the finding reported by Abdel-Tawwab (2004) which ranged from 1.80 ± 0.05 to 2.21 ± 0.09 . However, the FCR in the present study was higher than The feed conversion ratio range of 1.72 to 1.76, reported by Kassaye Balkew *et al.* (2012). . Relatively the lowest mean value of feed conversion ratio was 2.02 recorded in Chamo strain while the poorest (2.27) was observed in Hawassa strain. Kassaye Balkew *et al.*, (2012) also reported that the poorest feed conversion value was attained by the Hawassa strain. .

Conclusions

In the characterization of the four local *Oreochromis niloticus* strains (Chamo, Koka, Hawassa and Zewy) to assess their better production traits, the fish behavior in spawning, presence of body deformity, their growth rates, survival rates, feed conversion rate and their sex ratio were determined and documented. The Chamo strains were found to be better in growth parameters and feed conversion efficiency followed by Koka strains though some individuals in the two strains demonstrated body deformities.

Recommendations

The following recommendations are drawn based on the results of the study: Chamo *Oreochromis niloticus* strain is a potential tilapia for aquaculture development, possessing desired production qualities that the strain should be protected and conserved in its original site, Lake Chamo avoiding the current threat in genetic erosion (through continuous removal of vigorous fish from the population) and genetic dilution with other strains. The strain should also be considered as a most potential breed for aquaculture development in National breeding strategy of aquaculture fish. Chamo strain is appropriate for fry production in hatchery to expand sustainable aquaculture development.

The future research on culture of Chamo tilapia has to consider avoiding/reducing the aggressive behavior of males in spawning and the body deformity. It is also necessary to confirm whether the body deformity is heritable and whether the characters in the current study are maintained by the strains at different agro-ecologies and culture systems.

Acknowledgement

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Limnological Assessment of Didessa Reservoir for Fisheries Enhancement

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Abstract

Scientific studies on water quality and productivity can contribute to suitability assessment of the water for particular uses such as domestic, irrigation, sanitation and fish production. The primary productivity of different water bodies has been widely investigated to assess fish production capacity of the water body to formulate fishery management policies. This study was aimed at identifying the retained fish species during initial impoundment of Didessa reservoir and studying the basic limnological features of the newly built reservoir to estimate its fisheries production potential. The lowest pH mean value of 7.85 ± 0.22 was recorded at inlet site and highest pH value of 8.30 ± 0.32 at outlet. Seasonal variations showed that mean pH value in dry season (7.92 ± 0.46) was significantly lower ($P < 0.05$) than the mean value in wet season (8.30 ± 0.32). The value of surface water temperature of the reservoir ranged between 26.19 and 29.1 °C. The highest Electric conductivity value of 368.22 $\mu\text{S}/\text{cm}$ was obtained at inlet and the lowest value of 375.13 $\mu\text{S}/\text{cm}$ at outlet. Chlorophyll-a varied from a minimum of 14.2 in dry season and a maximum of 42.6 in Wet season with a mean value of 29 mg Chl-a m^{-3} . Three fish species of two families *Labeobarbus intermedius* (Rüppell, 1835) followed by *Oreochromis niloticus* (Linnaeus, 1758) and *Labeo cylindricus* (Peters, 1852) were recorded in Didessa reservoir.

Keywords: fish biodiversity, limnological parameters , reservoir creation,

Introduction

Although the definition of water quality depends on the purpose of the use of the water, it is mostly based on the biological, chemical, and physical elements of water and their interactions (Taner *et al.* 2011). Water quality becomes a critical factor affecting human health and wellbeing, with its deterioration and associated waterborne diseases which could lead to death (Patil *et al.* 2012). Therefore, it is essential and important to test the quality of water before using for drinking, domestic, agricultural or industrial purposes using different physicochemical and biological parameters. In Ethiopia, freshwater fish production is important for nutritional security in riparian communities. Being landlocked, lakes, rivers, and reservoirs of the country are important sources of aquatic protein and micronutrients (Tesfaye and Wolff 2014).

Limnological studies of tropical freshwater ecosystems are not extensive when compared to temperate ones. But East African lakes have been studied fairly well in comparison with other regions of Africa. This is true of Ethiopian lakes even though rift valley lakes have been relatively better studied (Zinabu *et al.* 2002, Gebrehiwot *et al.* 2017).

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, and other socioeconomic values based on its physical, chemical, and biological characteristics (Rim-Rukeh 2013). The quality and productivity of any water body are governed by its physicochemical factors and biological characteristics (Venkatesharaju *et al.* 2010). Distribution and productivity levels of organisms are largely determined by these factors (Pawar *et al.* 2006).

The primary productivity of different water bodies has been widely investigated to assess fish production capacity of the water body to formulate fishery management policies (Kumar *et al.* 2015). The study of energy transfer in lakes and reservoirs is based on the measurement of primary productivity of phytoplankton and the environmental variables, which limit or control this productivity. Primary productivity of aquatic ecosystems depends basically upon the photosynthetic activity of autotrophic organisms, like phytoplankton particularly in the open area of the water body (Tilahun and Ahlgren 2010). This study was aimed at identifying the retained fish species during initial impoundment of Didessa reservoir and studying the basic limnological features of the newly built reservoir for estimating its fisheries production potential.

Materials and methods

Study area

Didessa Reservoir is located along Didessa River, one of the largest tributaries of the Blue Nile River in Western Ethiopia. The major tributaries of 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 km², and it extends to area of 8 administrative districts of the two zones (Jima-Arjo and Bedele) of the National Regional State of Oromia. When its construction is completed, the earth and rock-fill Dam will have a height of 40.6 m.

Physicochemical parameters

Surface water temperature, pH, Dissolved oxygen (DO), Electric conductivity (EC), total dissolved solids (TDS) and transparency were measured in-situ. The water temperature, DO, EC and TDS were measured using portable digital water quality multi meter model CO 411, and pH was measured with portable pH meter model CP 401. Transparency was measured with 20 cm diameter secchi disk. Chlorophyll-a was determined spectrophotometrically at 665nm and 750nm after centrifugation using hot 90% acetone as the extracting agent from seasonal water samples in lab.

Fish sampling

The fish communities were sampled by multifilament gill net having (60, 80, 100 and 120 mm) stretched mesh sizes. Immediately after capture, species of the fish were identified, and the total length (T_L) and total weight (T_w) were measured and weighted to the nearest 1mm and 0.1g respectively.

Result and Discussion

Physical Parameters:

Seasonal variations showed that all the examined parameters were significant ($p < 0.05$) at wet and dry seasons. The physical water parameters of the reservoir were not significant ($p > 0.05$) between the sampling stations. The lowest pH mean value of 7.85 ± 0.22 was recorded at inlet site and highest pH value of 8.30 ± 0.32 at outlet. Seasonal variations showed that pH values in dry season (7.92 ± 0.46) were significantly lower ($P < 0.05$) than wet season (8.30 ± 0.32). The pH of the reservoir recorded during the period of the study falls within the recommended limits of 6.5 – 8.5 (Committee 1999) suitable for productive waters and falls within range of WHO standards of 6.5 to 8.5 for drinking water indicating that the reservoir in all indication is good for drinking and fish production. pH is one of the most important operational water quality parameters, supposing, pH is above 7, this will indicate that water is probably hard and contains calcium and magnesium (Zhu and Schwartz 2011).

The value of surface water temperature of the reservoir ranges between 26.19 and 29.1 °C. The record of water temperature showed that there was significant difference ($p < 0.05$) among the seasons. According to Jayalakshmi *et al.* (2011) report, water temperature is a vital factor affecting the chemistry and biochemical reactions in organisms. High temperature may increase toxicity of many substances such as heavy metals in water for domestic use (Committee 1999), however, high temperature is also favorable for optimal growth of aquatic animals. The values obtained in this study were slightly above the recommended WHO (2006) standard of 25 °C. High water temperature enhances the growth of micro-organisms and may increase problems related to taste, odor, color and corrosion (WHO, 2011).

The electrical conductivity (EC) of a water body is a very useful parameter for determining the water quality (Zhu and Schwartz 2011). Electric conductivity is a measurement of water's current and is directly related to the concentrations of ionized substance in the water and the levels affected by the EC of water are a direct function of its total dissolved solids, organic compounds and temperature. From the results it was evident that the highest value of 368.22 $\mu\text{S}/\text{cm}$ was obtained at inlet and the lowest value (375.13 $\mu\text{S}/\text{cm}$) at outlet. There was no significant difference ($p > 0.05$) of electric conductivity between the sampling sites. These values were below the recommended limits of 1000 $\mu\text{S}/\text{cm}$ for drinking water and fish production. Water transparency (ZSD) varied from a minimum value of 0.50 and 0.46 m in wet season to a maximum of 0.6 and 8.0 m in dry season at the inlet and outlet stations respectively. High values of water transparency coincided with dry periods (with low precipitation) whereas low values were recorded during rainy months. The difference in ZSD between the two stations was statistically significant ($P < 0.001$).

Chlorophyll a:

Phytoplankton biomass estimated as chlorophyll-a varied from a minimum of 14.2 in dry season and a maximum of 42.6 in wet season with a mean value of 29 mg Chl-a m^{-3} . The observed maximum concentration coincided with high light utilization efficiency of phytoplankton and high light-saturated rate of photosynthesis. Higher values above 29 mg Chl-a m^{-3} were estimated during wet months while lower values were in dry months.

Fish production potential:

Oglesby (1977) has derived relationships between fish yield ($Y_d = g \text{ dry wt m}^{-2} \text{ yr}^{-1}$; $Y_c = g \text{ C m}^{-2} \text{ yr}^{-1}$, assuming 25% dry wt: wet wt, and $1 \text{ g C} = 10 \text{ g wet wt}$ or 2.5 g wet wt) and summer standing crop of phytoplankton (CHLs, mg m^{-3} , of Chl-a) or primary production (P, in $\text{g C m}^{-2} \text{ yr}^{-1}$). $\log Y_d = -1.92 + 1.17 \log \text{CHLs}$; $n = 19$, $r = 0.92$. Based on this parameter fish production of a 7.5 kg fish can be produced per m^2 annually.

Species composition:

Three fish species of two families were recorded in Didessa Reservoir (Table 12). In the present study, *Labeobarbus intermedius* (Rüppell, 1835), Followed by *Oreochromis niloticus* (Linnaeus, 1758) and *Labeo cylindricus* (Peters, 1852).

Table 16. Fish species composition of Didessa Reservoir

Family/ Species
Cichlidae
<i>Oreochromis niloticus</i> (Linnaeus, 1758)
Cyprinidae
<i>Labeobarbus intermedius</i> (Rüppell, 1835)
<i>Labeo cylindricus</i> (Peters, 1852)

Conclusions and recommendations

Assessment of water quality is an important factor for assessing pollution levels and the productivity of the water body. This study revealed that the water in Didessa reservoir is suitable for fish production and other domestic purposes. The periodical assessment of physico- chemical analysis of the water body should be carried out, as this would be helpful in early detection of any future water quality. In all, the ranges of physico-chemical properties of Didessa Reservoir are comparable to those found in non-polluted African reservoirs, and are within the allowable limits recognized by WHO for drinking water supply as well as fish production.

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Assessment of the current fishery activities and some factors that affect the production of fish in Lake Koka

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Abstract

Lake Koka is an economically important lake in the country. However, the physico-chemical parameters of the lake seem to be threatened by anthropogenic and climatic factors, which in turn affect biotic factors as reflected in fish catch. This study was made to assess the status of fishes and factors that affect the production of the fish in the lake during 2018. Four fish species were identified. The composition of the fishes has undergone changes as compared to the last few decades. Clarias gariepinus attained the highest relative frequency (35%) next to Cyprinus carpio (36%), and then followed by Oreochromis niloticus (18%) and Labeobarbus intermedius (11%) in the catch proportion during this study. The fish catch of Lake Koka currently declined from 634 tons in 2007 to 542 tons in 2018. Most of the threats resulted from the anthropogenic impacts on the lake. Increased pressure in fishing was a problem in the lake. Currently, plenty of pumps in state and private commercial farms are abstracting fresh water from the lake throughout the year that they are critically affecting the water level of the lake. The lake ecosystem is also affected by catchment degradation which resulted in increased nutrient loading and siltation. The study suggested that if nutrient levels continue to increase and water levels continue to decline, further changes in fish composition can be expected in the lake, especially with a shift towards fish that are mainly turbidity-tolerant species such as C. carpio. The study showed that the fishery sector has been of critical importance to the economy and to the social well-being of the fishermen in the study area. However, current harvest trends and fishery conditions put these attributes of the production at risk. It is threatened with problems of pollutions, open access to the resources, lack of technology and marketing. Hence, appropriate management is an urgent requirement that could assist in sustainable exploitation of the resources, so that the resource could contribute to food security in the study area in particular and in the country in general.

Key words: Fish composition, fish yield, fishery constraints, Lake Koka.

1. Introduction

Lake Koka was constructed in early 1960s across Awash River mainly for hydroelectric power generation, and flood control (Halcrow and Pattern, 1989). It is located in an area with many agricultural activities and there are no soil conservation efforts in its catchment area. IBC (2005) also reported that the situation of the lake ecosystem is being affected by catchment degradation, siltation, imbalance between water inflow and outflow and uncontrolled fishing practices. Eventually, the lake started to support common carp, barbs, tilapia and catfish, most of which came from the river and the carp was very likely to have been introduced by early workers like Shibru Tedla, Feseha Haile Meskel and some FAO experts of the time. Koka has additional function of assimilating waste waters and industrial effluents from leather factories,

flower farms, etc., which has made the water highly eutrophic leading to the development of toxic Cyanobacteria (Elizabeth Kebede and Willen 1998). Such poisons coupled with pollution with heavy metals from the factories have started to cause serious health problems to local communities who directly drink the Koka water and bath in it (Brook Lemma, 2012). There are allegations of skin diseases, retarded developments of children and even deaths according to documentary films by renowned media such as Al Jazeera.

The commercial fish production in the lake was intensified through the support acquired from Lake Fisheries Development Project and onwards, the fish stocks of the lake have declined for a variety of reasons. Lack of appropriate management of the fishery of the lake result that average size of the fish caught have continued to show a declining trend and this implies that the fish stocks are being depleted and the long-term effect of all these on water quality and on food webs is complex and affects the fish production of the lake. Hence, based on the historical data available on environmental as well as water quality, it is possible to assign fishery changes in the lake to identifiable factors. Hence, the objectives of this paper is therefore, to assess the composition and production trends of the fishes in the lake, to assess the variation in physico-chemical factors that influences the fish community structure of the lake and finally to identify the constraints that affects the production of the fish in the lake.

2. Methodologies

2.1. Description of the study area and site selection

Lake Koka (Fig. 1c) is located in the Ethiopian Rift Valley (Fig. 1a & 1b) at 08° 23'22"N; 39° 5'15"E and altitude of 1,590 masl, which is about 120 km south of Addis Ababa. The lake had an initial volume of 1650 Mm³ (Abebe Mengistu, 2001), and surface area of 200 km² (Willén *et al.*, 2011). The morphometric characteristics of the lake have, however, been changing due to progressive siltation. According to a review by (Abebe Mengistu, 2001), the reservoir had lost 28.9 % of its water holding capacity by the year 1998 primarily due to siltation. The climate of the reservoir area is characterized by bimodal rainfall pattern, with a maximum peak in August and a smaller peak in April.

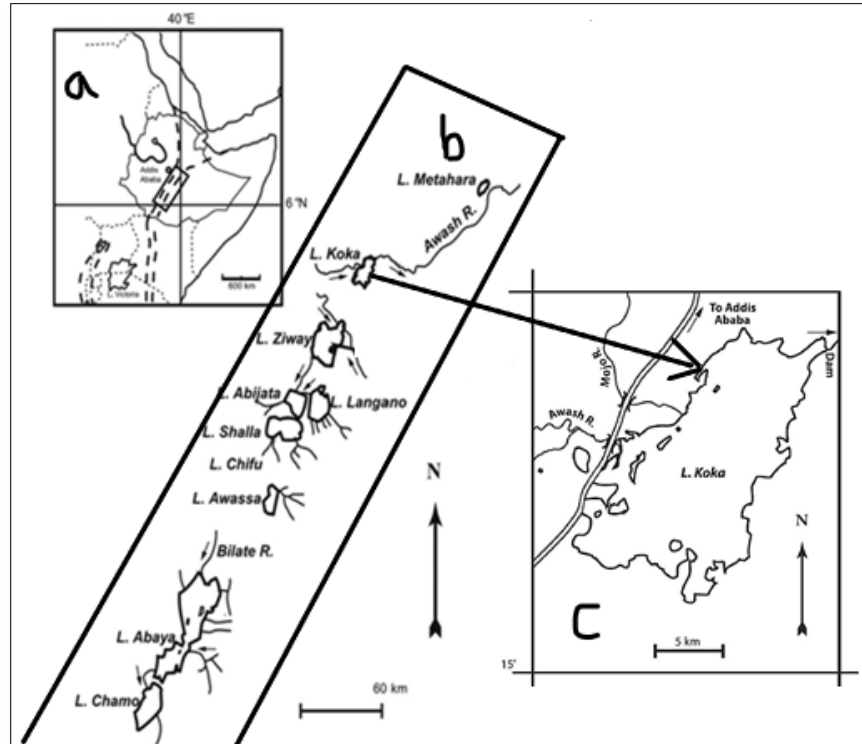


Fig. 1. (a) Location of Ethiopia in the Horn of Africa, (b) Location of Ethiopian Rift Valley Lakes and (c) Lake Koka

Before starting regular sampling program, a reconnaissance survey was conducted to fix the sites. Hence, three sampling sites were selected based on geographical proximity and/or habitat similarity (neighboring floodplains, depth and distance to shore), their distance from human settlements and anthropogenic effect. Global Positioning System (GPS) readings were taken to demarcate the locations of the sampling sites in the lake during the survey.

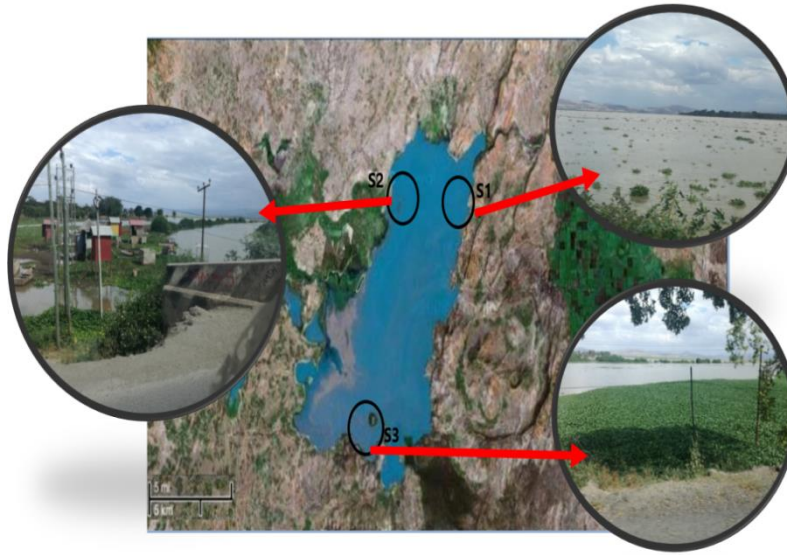


Fig. 2 Sampling sites

Site 1 is located around the hydroelectric power station and relatively the deepest area of the lake, with a distance of 1.5 km from the tip of the mouth of turbine of the hydroelectric power. The area is mostly composed of mud.

Site 2 is located in the western part of the lake and the shoreline is characterized by intensive cultivation with horticulture and field crops.

Site 3 is located around the leather industry and as is the case with site 1, the area was characterized as moderately cultivated (Fig. 2).

2.2. Field sampling and measurements

2.2.1. Physico-chemical parameters:

Physico-chemical parameters of the water were measured monthly from each site during the study periods. Temperature, pH and conductivity of the lake were measured *in situ* at each sampling site. Temperature and pH were measured using a portable digital pH meter (Hanna 9024) and conductivity was measured using conductivity meter (Elmetron-model of CC411). Transparency was measured with a Secchi disc of 20 cm diameter. Water samples were collected from each site in dark plastic bottles, washed with acid and rinsed with distilled water several times in duplicates for nutrient analysis. Water samples for Chlorophyll-*a* determination was taken by the Schindler sampler from all sites. Finally, the samples were transported on ice to the laboratory for further analysis.

2.2.2. Fish parameters

Parallel to the physico-chemical sampling, every month the fishes were collected from each site using different fishing gears, which includes gill nets of various mesh sizes (6, 8, 10 and 12 cm stretched mesh sizes) and multiple long-lines of different hook sizes (9, 10, 11 and 12). In addition, monofilament nets

with various stretched mesh sizes (5 mm to 55 mm stretched mesh size) were used. The gears were set in the afternoon (4:00 pm) and collected in the following morning (7:00 am).

2.2.3. Catch and effort

Catch and effort data were recorded from commercial fisheries at landing sites of the lake during the study period. Total landing by species in kg, the number and type of gears used for fishing and the number of settings for each gear were recorded. The number of gears in a community was monitored. Total effort per gear type was estimated based on the number of gears (gill net and hooks), settings (beach seine) and time (the number of days the gear was deployed in the month). The number of gears and settings in relation to fish catch in a gear was obtained through interviews with the fishermen. These data were also used as raising factor in the estimation of annual yield. A review was also made on the long-term trend of fish production from various published research results and unpublished data collected by different organizations.

2.3. Factors that affect the fish production

Some physico-chemical variables and fishing gears that directly related to the fishery were reviewed to assess its effects on the production of fish.

2.4. Constraints of the fishery in the lake

2.4.1. Selection of landing site and fishery cooperatives

Landing sites were selected on the shores of the lake that fishermen were used to land their fish catches. Totally three major landing sites were purposely selected for the survey.

2.4.2. Sample size determination of interviewee households

Arsham's (2005) mathematical formula, as indicated below, was used for sample size determination.

$$N = 0.25/SE^2;$$

Where N = Sample size, SE = Standard error; which was calculated by using confidence interval of 10% and confidence level of 95%, $SE = 0.1/1.95 = 0.05$, where 1.95 is constant

Totally 100 households were randomly selected and data were collected in the study area through semi-structured questionnaire interview and the content of the questionnaire includes main constraints of the fishery and the marketing system in the lake.

2.4.3. Focus group discussion and key informants

Focus group discussions were held in the study area after filling out the questionnaires by way of interview. Members of the focus group were purposely selected to make a total of four to six members. The members included committee members of fishery cooperative, peasant association executive committee members, development agents, and the districts livestock and fishery development desk officers and individuals who were believed to be knowledgeable about the past and present status of the fishery of Lake Koka.

The leading check list of issues to be discussed at the group discussions were prepared to guide the discussion with the focus group with special emphasis on policy issues, external support for the schemes, institutional and managerial issues, major problems and future plans to further development of the fishery of the lake.

In addition, secondary data were collected from published and unpublished sources. Bora, Lume and Adama Districts' Agricultural and Natural Resource Development Offices, Cooperative Promotion Offices and Livestock Development Agency Desks were the important sources of data.

3. Results and discussions

3.1 Fish species distribution and composition

A total of four species of fish in the Families Cyprinidae, Claridae and Cichilidae were identified from the different sites in the lake (Table 1 and Fig. 3). The species were *Labeobarbus intermedius* and *Cyprinus carpio* from the Family Cyprinidae, *Clarias gariepinus* from the Family Claridae and *Oreochromis niloticus* from the Family Cichilidae. The status (presence/absence) of the species from the sampling sites was provided in Table 1.

Table 1. Fish species identified from the study sites of Lake Koka (Present (+), Absent (-))

Family	Fish species	Sampling Sites		
		S1	S2	S3
Cyprinidae	<i>Labeobarbus intermedius</i>	+	+	+
	<i>Cyprinus carpio</i>	+	+	+
Claridae	<i>Clarias gariepinus</i>	+	+	+
Cichilidae	<i>Oreochromis niloticus</i>	+	+	+

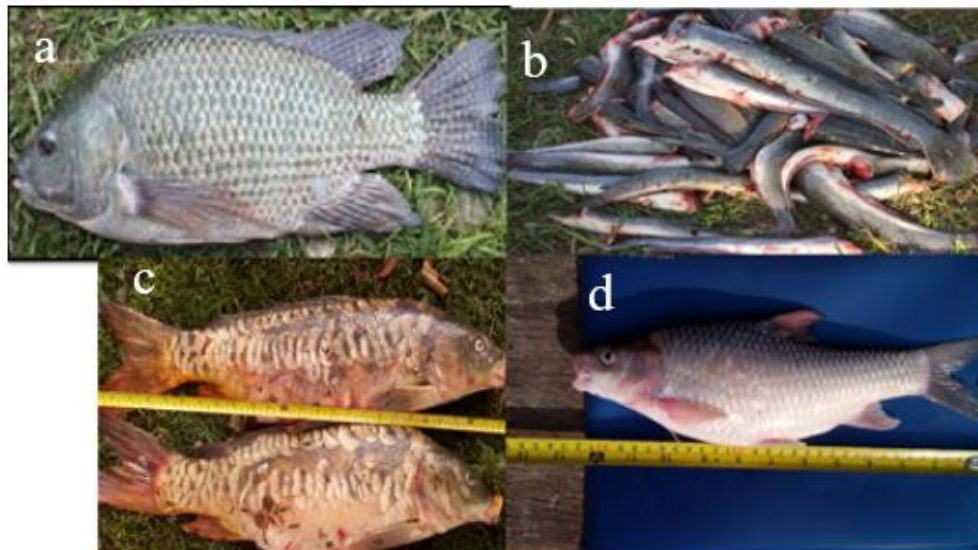


Fig. 3. Lateral view of sampled fish (a) *Oreochromis niloticus*, (b) *Clarias gariepinus*, (c) *Cyprinus carpio* and (d) *Labeobarbus intermedius*

A total of 2019 fish specimens were recorded from the three families during the study period. The species were *C. carpio* and *L. intermedius* from the family Cyprinidae (56%); *Oreochromis niloticus* from the family Cichilidae (18%); and *Clarias gariepinus* from the family Clariidae that accounts 26% (Fig. 4).

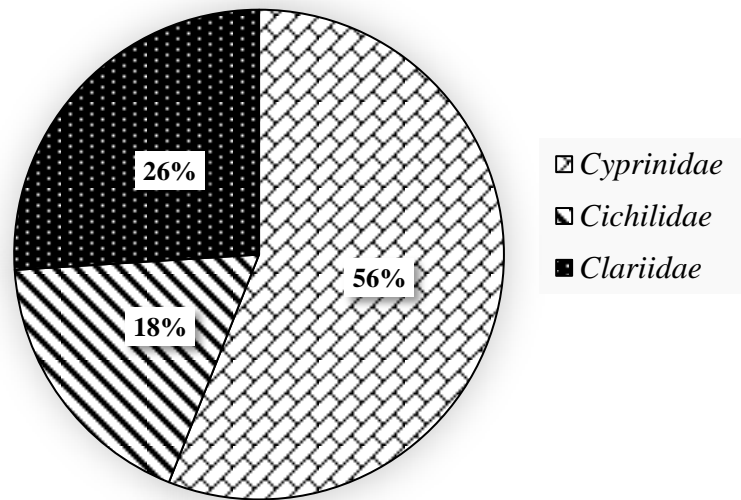


Fig. 4. Composition of fish at family level in the lake (%)

At species level, *C. carpio* was the dominant fish species from the Family Cyprinide as well as the whole species from the lake and it accounts to 36% of the total catch. *Clarias gariepinus* was the second dominant species that accounts to 35%. *O. niloticus* and *L. intermedius* were represented in the catch with 18% and 11 %, respectively (Fig. 5).

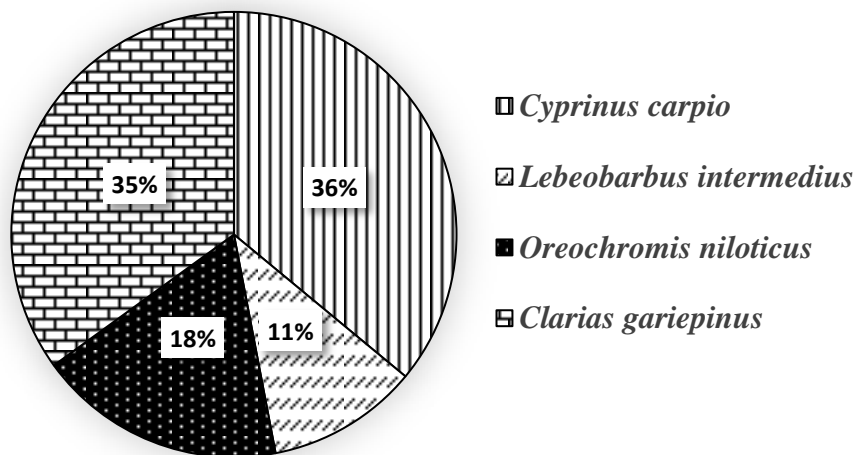


Fig. 5. The current fish species composition of the lake

3.2. Annual fish catch

Lake Koka fisheries have been developing over the past decades and the lake was part of the eight major lakes considered by the lake fisheries development project (LFDP) in 1990s (LFDP, 1997). Koka is among the most important lake for Ethiopian small-scale fisheries in general and riparian societies in particular (Gashaw and Wolff, 2014). At present, it provides about 542 tons of fish annually (Fig. 6). Currently *O.*

niloticus, *C. gariepinus* and *C. carpio* were commercially important fish species in the lake. Of these *C. carpio* and *C. gariepinus* comprise more than 70 % of the annual catch.

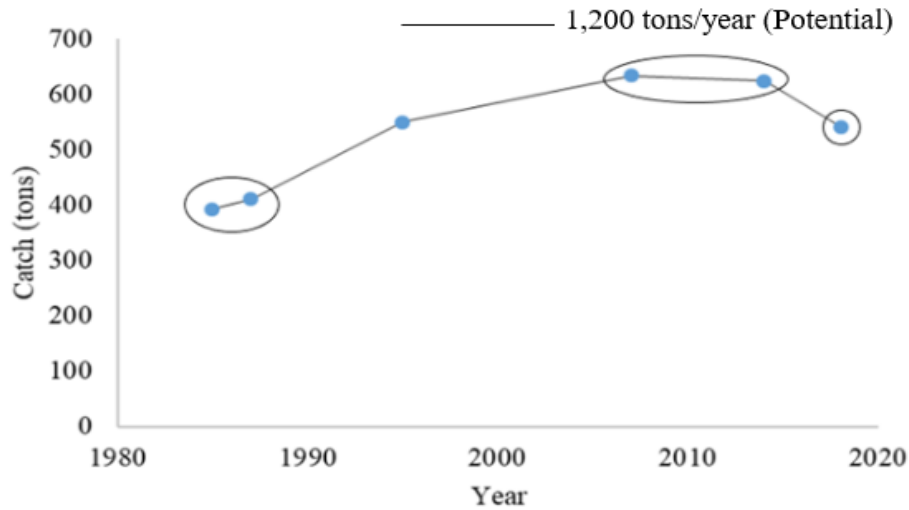


Fig. 6. Change in total annual fish catch from Lake Koka between 1985 and the present (Source: LFDP, 1985; Gashaw Tesfaye, 2014 and the present study).

The annual fish catch of the lake showed different trends with time (Fig. 6). The first fishing phase was around 1985, which was a starting period of fisheries in the lake where the catch was lower with a mean annual catch of 393 tons. The possible reason for the stability was low number of active fishing gears in the lake. The second phase was the growth period around 2007, which was a period when yield increased to 634 tons which has almost the recommended sustainable catch level for the lake. At this time additional and effective fishing gears were handed by fishermen. The third phase was the period in which fish catch declined to 542 tons during the study period.

3.3. Factors that affect the fish production of the lake

3.3.1. Fishing gears

Three types of gears were operated in the lake: beach seines, gillnets and long-lines (Table 2); while hook-and-lines were utilized by occasional fishermen along the shoreline. The fishermen utilize wooden boats for casting beach seines and rafts for gillnets and long-lines. Beach seine was the most common fishing gear used, whose presence increased from about 60 in 2002 to 68 in 2018 while, the numbers of gill nets and long-lines have been decreased from 23 and 17 in 2002 to 20 and 12 in 2008 respectively (Table 2). The increase in destructive type of fishing gear (beach seine) in Lake Koka may be to increase the catch of the fish in the lake.

Table 2. Summary of fishing gears trend involved in Lake Koka in different years (in percent)

Years	Gill nets	Beach seines	Long-line (Hooks)	Reference
2002	23	60	17	Gashaw Tesfaye,2014
2018	20	68	12	Current study

3.3.2. Water quality

Aside from problems related to decreasing water level of the lake, water abstraction started to induce salinity, due to losses of freshwater from the lake. Simultaneously, the application of agrochemicals and fertilizers due to intensification of irrigation practices around the lake has increased nutrient loads and affected the water chemistry of the lake (Table 3). With the clearing of the vegetation in the watershed for different purposes, silt loads during rainy season and the feeder river (Awash River) bringing impact on the limnological system and hydrological conditions of the lake (Table 3). Hence, currently, the high concentration of SRP in the lake could be due to influx of pollutants and organic matter from the lake shores (Fig 2). The farm lands located near the shore use fertilizers which are rich in nutrients, especially phosphates and nitrates, and hence can increase the concentration of SRP (Table 3).

Table 3. Trends in some physico-chemical factors of Lake Koka

Parameters	Unit	Amount		General trend
		Previous	Present study	
Secchi depth	cm	28 (Kebede <i>et al.</i> (1994)	19.1	Decreasing
Mean depth	m	9 (Wood and Tailing (1988)	3.2	Decrease
pH	-	8.3 (Wood and Tailing (1988)	8.1	Decreasing
Temperature	°C	22.56 (Yeshimebet Major,2016)	25.6	Increase
D. Oxygen	mg/L	7.7 (Yeshimebet Major,2016)	5.9	Decreasing
Chl- <i>a</i>	µg/L	22.4 (Elizabeth Kebede and Amha Belay,1994)	236	Increasing
Nitrate	µg/L	89.3 (Fasil Degefu <i>et al</i> ,2011)	392	Increasing
SRP	µg/L	167 (Yeshimebet Major, 2016)	424	Increasing
Ammonium	µg/L	44.4 (Fasil Degefu <i>et al</i> ,2011)	264	Increasing
Conductivity	µS/cm	200 (Melaku Mesfin <i>et al.</i> , 1988)	401.9	Increasing

The turbidity expressed as low Secchi depth measured during the study period was high as compared to previous records (Table 3). The lower Secchi depth can be partly caused by the rising levels of siltation which can also be a reason for increased conductivity. Also the major route of entry of nitrogen waste into the lake may be due to anthropogenic causes related to activities in the lake's catchment such as the leather industry, Blane flower farm wastewater, discharges from car exhausts, etc. All these directly or indirectly affect the water quality of the lake.

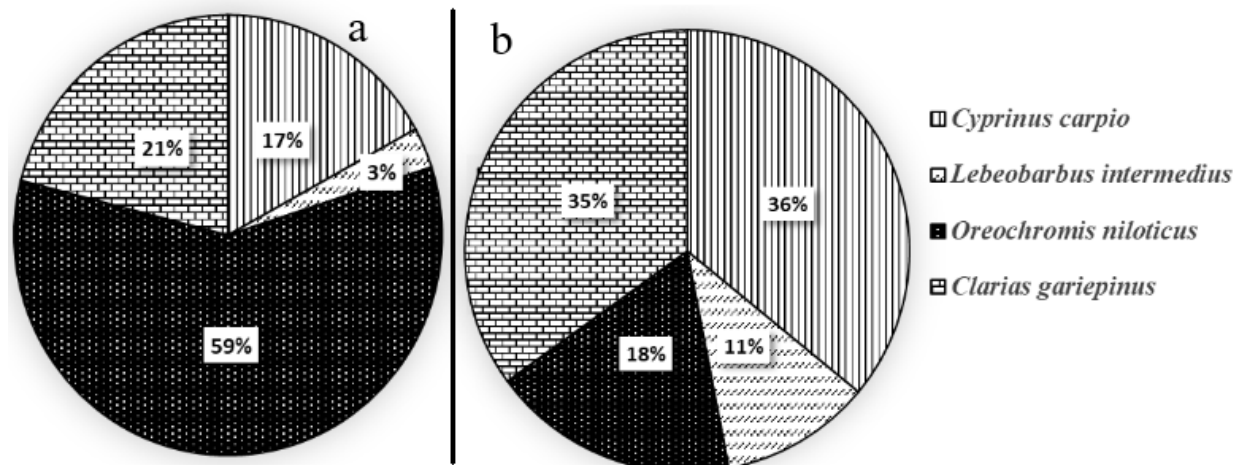


Fig. 7. Fish species composition by number (%) in 1987 (a) and the current study (b) of Lake Koka

Previously in Lake Koka, *O. niloticus* was one of the fish species that 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 Ziway in rift valley area and Lake Naivasha in Kenya (Mavuti, 1983). Currently, the littoral zone is being affected due to different anthropogenic factors and the composition of the fishes changed and as a result there is a progressive increase in the proportion of *C. carpio* (Fig. 7)

3.4. Major constraints of the fishery sector of Lake Koka

Pollution and open access to the resource were the most common problems in the lake according to the current survey result of respondent households (Fig. 8). Pollution of the lake was taking place due to improper farming methods and poor tillage systems, which contribute towards the erosion of top soils of the steep cultivated land around the catchment of western part of the lake. Urbanization and human settlement are amongst the most serious problems around the lake; associated industrial development was also problematic in Lake Koka that intensifies pollution. Farming along the lakeshore not only disturbs lakeshore ecology but also exacerbates siltation and increases turbidity as indicated in table 3.

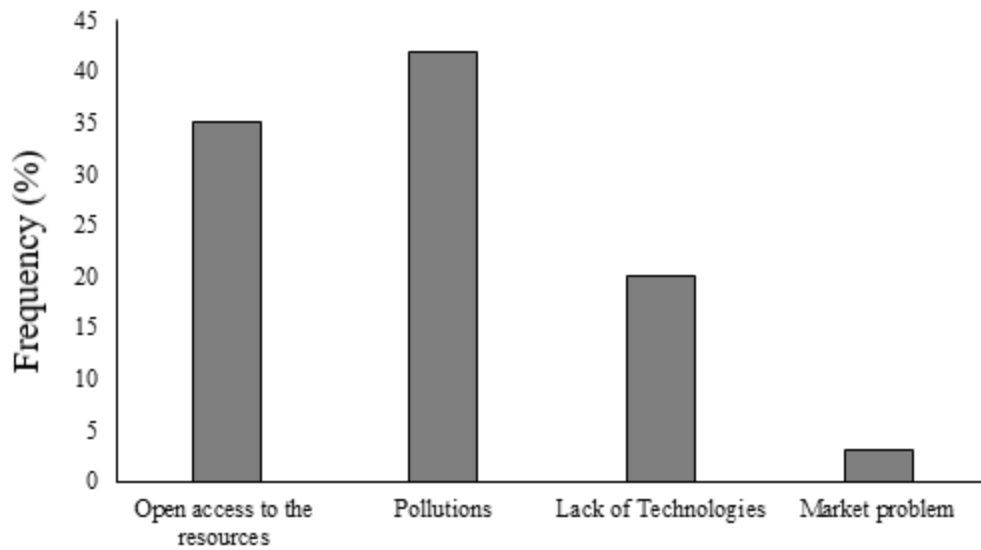


Fig.8. Major Constraints of Lake Koka fishery as indicated by respondent households

In the area, the market issues depend on physical access to landing points, numbers of retailers in the area and the amount of catch (personal observation). This issue was not faced as a major problem by the fishermen in the study area.

3.5. Marketing chain of the lake

A generic schematic fish market chain at the study site is presented in Fig. 9. Based on information gathered in the course of the field study there were no variation with the report of Lake Ziway in 2016. The key actors along the chains include cooperative and private fishermen at landing and their assistants (Fig. 9).

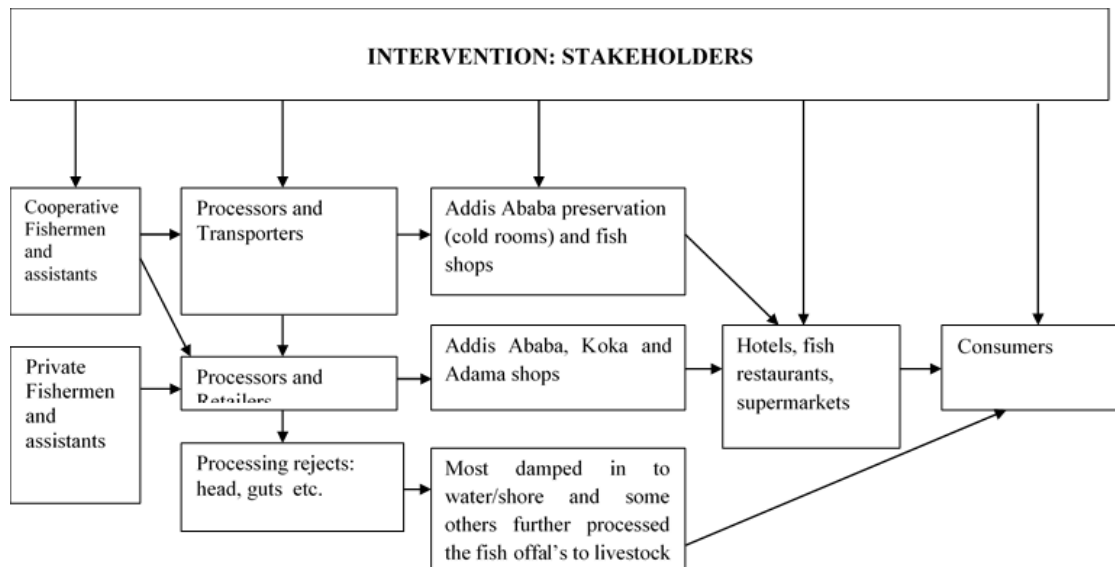


Fig. 9. Market chains of the fish of Lake Koka

4. Conclusions and recommendations

Lake Koka has shown some undesirable changes in terms of some physico-chemical factors and shift in catch composition of fish species. Soluble reactive phosphorus, nitrate level and phytoplankton biomass (as chl-*a*) of the lake increased in recent years. The Secchi depth reading of the lake have shown declining trend. The fish fauna of Lake Koka was dominated by Cyprinids. Based on percentage composition, *C. carpio* was relatively the most dominant fish in Lake Koka, which contributed to 36 % of the total catch. *Clarias gariepinus* contributed 35 % of the samples from the lake. The remaining fishes were each less than 18% of the total number of fishes encountered.

The status of Lake Koka fish yield has changed over time. Currently, the contribution of carp species (*C. carpio*) has increased and the contribution of *O. niloticus* has decreased. This is due to the increase in effort applied through time and other anthropogenic impacts exerted on the lake. Fishing technology on Lake Koka was not modern in its nature. Beach seine was predominant fishing gear on the lake. Even currently, the number of beach seines has increased as compared to other gears that operated on the lake.

The major cause for the accelerated water quality changes in the lake was identified as human impact in the catchment of the lake. This includes land use changes, particularly removal of vegetation cover (through deforestation, animal grazing, etc.), irrigation, diversion of inflows and industrial use of the water were taken as the cause for the decline in water quality and quantity that directly have an effect on the current change in fish catch composition.

Fishing plays an important role in the livelihood sources of the community through both direct consumption and other income generation in the study area. However, it is constrained by several challenges like, pollution, open access to the resources and lack of technology, among others. Thus, if some or all of the challenges are tackled, fishing can be a pillar of economic activity in the study area and appropriate management measures are urgent need to address the contribution of the fishery as a source of food, income and employment as ongoing activity. This can be done either by the government or by the fishing communities themselves or by both and may follow the following recommendations.

- With increase in nutrients and inorganic turbidity 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. The lakeshore should be restored with macrophytes and protected in order to control external nutrient loading.
- There are indications of severe degradations of the lake basin particularly the water shade areas. The most threats to the lake are related to deforestation, irrigation and overgrazing by domestic animals in the lake basin. Therefore, sustainable utilization of soil/aquatic resource? and conservation measures should be taken in and around the lake.
- Proper irrigation scheduling, focusing on high value crops, using limited area and water, and detailed crop-water requirements study has to be made in irrigation fields to protect the over abstraction of the lake before affecting the resource of the lake.
- In Lake Koka large number of small sized fish of all species is being exploited and proper management actions are required to protect the immature fish. Thus, management tools needs to practice like closed seasons, catch quota restriction, mesh size regulations, gear restrictions and limits on the number of fishers has to be for sustainable exploitation of the stocks.
- The development of aquaculture and other related alternative fisheries (Integrated-fish-horticulture-poultry farming) to reduce the pressure on the natural system should be encouraged.

- Promotion of credit arrangements should be given due attention to help fishermen access modern fishing equipment and maintain their fishing gears. Such credit can also be extended to complementary livelihood activities such as agriculture.
- Need to implement the existing regional as well as national fishery proclamation.
- Based on the lesson from Lake Koka, detailed studies and investigations are required on status and trends of fishes and fishery in rift valley basin in general due to high pressure of degradation of the area as well as ecological issues specially that of catchment of the lakes.

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