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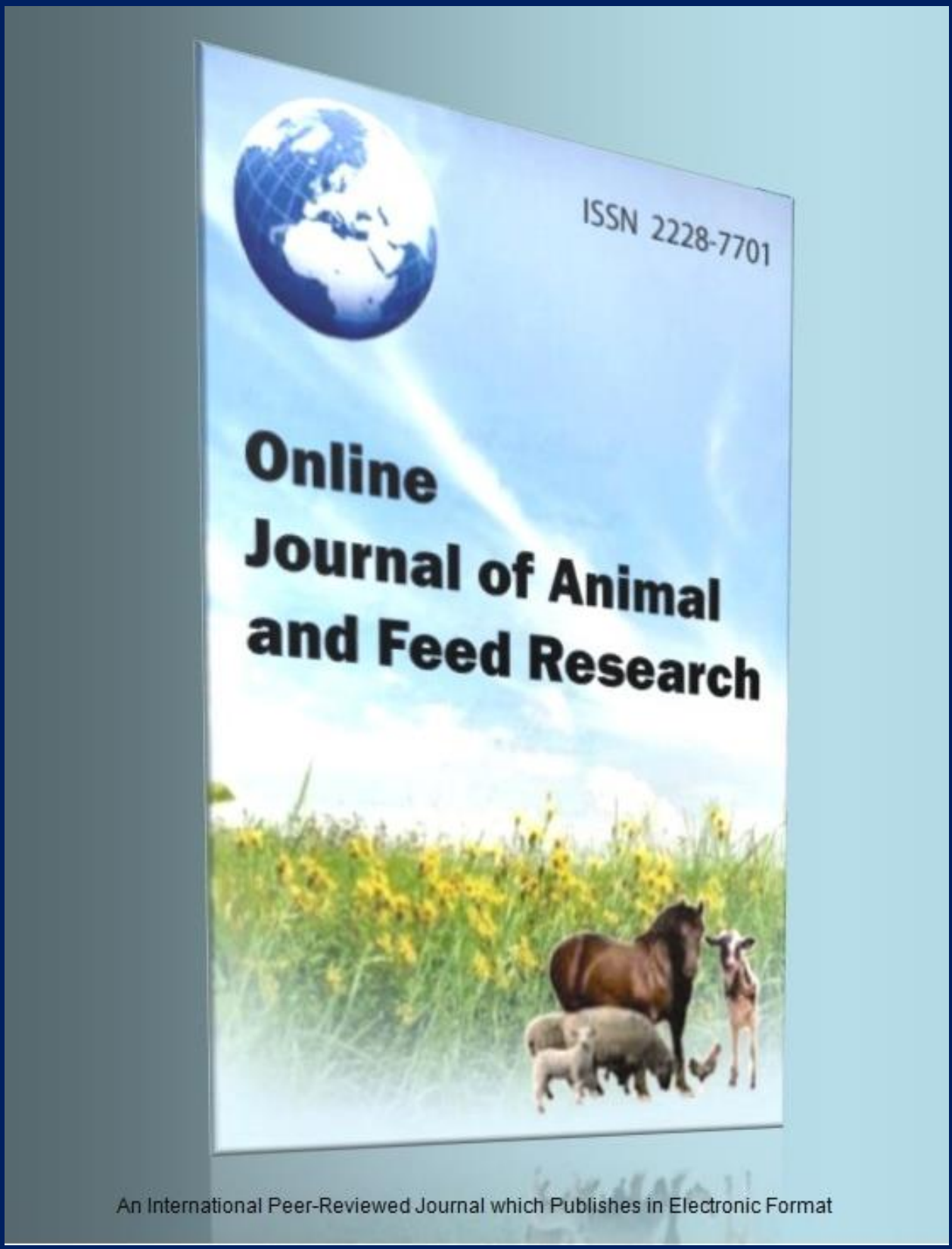

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Prevalence of hemorrhagic septicemia in cattle and buffaloes in Tandojam, Sindh, Pakistan.

Habib F, Jabbar A, Shahnawaz R, Memon A, Yousaf A, Bilal M, Jamil T, Khalil R and Sharif A.

Online J. Anim. Feed Res., 9(5): 187-190, 2019; pii: S222877011900026-9

Abstract
The current study was performed to evaluate the prevalence of hemorrhagic septicemia (HS) in cattle and buffaloes in Tandojam Pakistan. The average geometric mean titers (GMT) recorded against HS in diseased buffaloes and cattle were 5.7 and 6.1, respectively. The morbidity, mortality and case fatality rates were 57.58, 52.30 and 90.83% in young buffalo calves; and 3.17, 1.92 and 60.65% in adult buffaloes, respectively. In case of young cattle calves, morbidity, mortality and case fatality rates were 8.63, 5.27 and 61.11%, respectively, while in adult cattle, these values were 4.83, 2.18 and 45.23%, respectively. The present study revealed that the mortality, morbidity and case fatality rates due to HS were greater in young calves than the adults both in buffaloes and cattle. Furthermore, buffaloes were found to be more susceptible to the disease than the cattle.
Keywords: Buffaloes, Cattle, Hemorrhagic septicemia, Prevalence, Tandojam

[Full text-PDF]

Research Paper

Using khat (*Catha edulis*) leftover meal as feed for sheep: its implication on feed intake, digestibility and growth.

Brhanu A and Gebremariam T.

Online J. Anim. Feed Res., 9(5): 191-197, 2019; pii: S222877011900027-9

Abstract
Ninety days feeding trial was conducted with the aim to evaluate the impact of replacing concentrate mix with dried khat (*Catha edulis*) leftover meal on feed intake, body weight change and digestibility of Tigray Highland sheep fed a basal diet of mixed grass hay. The trial was carried out using 24 lambs (17.8±1.08 kg) with six blocks and four treatments in a randomized complete block design (RCBD). Concentrates mix was substituted with khat leftover meal at a ratio of 0% (T1), 15% (T2), 30% (T3) and 45% (T4) on DM basis. The dietary rations were formulated in iso-nitrogenous to meet the nutrient requirements of lambs. Data was analyzed by analyses of variance (ANOVA) using the General Linear Model (GLM) procedures of SAS (2008). Treatment means comparison was done using Tukey's HSD test at P< 0.05. Dried khat leftover meal had moderate crude protein (12.3%) and less NDF value (31.9%). Substituted concentrates mix with 15% khat leftover meal had better impact (P< 0.001) on lamb's total DM, OM, ME, ADL and ADF intake compared to the lambs dietary T3 and T4. Daily weight gain and feed conversion efficiency were significant (P< 0.001) till substituted concentrates mix with 30% khat leftover meal. Diet digestibility showed reducing as inclusion level of khat meal increased across treatment. It is concluded that khat leftover meal can be utilized up to 30% without deleterious effect on animal performance and health.
Keywords: Body Weight, Digestibility, Feed Intake, Khat Leftover.

[Full text-PDF]

Research Paper

Prevalence of parasitic honeybee diseases, pests and predators in North Gondar zone.

Nega T, Yayeh M, Mitiku T, Birhan M, Bogale B, Chanie M, and Kinubeh A.

Online J. Anim. Feed Res., 9(5): 198-205, 2019; pii: S2228770119000208-9

Abstract
A cross-sectional study was conducted on parasitic honeybee diseases, pests and predators Lay armachiho and Tach armachiho districts of North Gondar zone from October 2017 to May 2018. The objectives of this study were to assess the different parasitic honeybee diseases, pests and predators. The study mainly involves with the collection of adult honeybee abdominal suspension of wet mount for nosema and amoeba diseases and direct observation of varroa mite and bee louse from adult honey bee and brood cells. Questioner survey was used to know different honeybee pests, predators and agrochemicals in the study area. The SPSS version 20 was used for chi-square test and p-value < 0.05 was taken as statistical significant. During the study period a total of 384 honey bee sample in traditional, modern and transitional hives were examined. The overall prevalence 24.47% (94/384) of hives were found positive for *Nosema apis*, 17.2% (66/384) foramoeba (*malphighamoeba mellificae*), 30.5% (117/384) for *varroa destructor* and 37.5% (144/384) for bee louse (*braula cocae*). Bee louse was the predominant external parasitic disease in adult honeybees followed by *varroa destructor*. There was statistically significant variation between *Nosema Apis* and bee louse observed among the two selected districts and hive types (x^2= 23.5, p-value= 0.0001) for bee louse and (x^2= 5.3, p-value= 0.071) for *Nosema Apis*. About the 100 respondents are 46%, 42%, 39%, 35%, 28%, small hive beetle 26% and spider 24% were complaining on the impact of wax moth, chemical spray, skunk, birds, small hive beetle and spiders respectively. They have also responded that pests play a major role on the production loss, damaging of honey bee colony and absconding of the colony. In conclusion, the highest prevalence parasitic honeybee diseases and pests were recorded in the study areas that signify the occurrence of the parasitic burden has to be carried out and immediate intervention was implemented.
Keywords: Honeybees, Parasites, Pests, Traditional And Modern

[Full text-PDF]

Research Paper

The effects of different litter material on broiler performance and feet health.

Kuleile N, Metsing I, Tjala C Jobo T. and Phororo M.

Online J. Anim. Feed Res., 9(5): 206-211, 2019; pii: S222877011800029-9

Abstract
A The study was implemented at the National University of Lesotho with the aim to find a potential litter material for use in broiler production. The study followed a completely randomized design with four litter treatments replicated three times. A well ventilated house divided into 12 pans was used where each pan or a replicate contain 15 birds with a total of (n=180) birds. Feeding and watering were done on ad libitum basis while the normal routine for broiler production was followed. Litter treatments were made up of control represented by wood shaving and it was compared to dried pine leaves, decomposed kraal manure and sand. All litter materials were applied at the depth of 10 cm. Data was collected on the following parameters broiler production, feet health and chemical and physical properties. Litter material treatment had no significant influence on feed intake, body weight, body weight gain, and feed conversion ratio and mortality rate. Regarding broiler feet health litter treatment had significant influence on foot pad dermatitis while hock burns and broiler temperature were not statistically different amongst litter treatments. Litter evaluation results revealed that different litter sources were differing significantly in terms ammonia emissions, water holding capacity. pH and bulk density while litter temperature did not differ significantly between litter treatments. It was concluded that both dried pine leave and decomposed kraal manure are potential sources that could replace wood shavings in broiler production. Dried pine leaves ideal for improved production while decomposed kraal manure deemed fit for ensure good feet health. It is therefore recommended that farmers in Lesotho can use both decomposed kraal manure and dried pine leaves to replace wood shavings.

Keywords: Feet health, Kraal manure, Lesotho, Litter material, Wood shaving.

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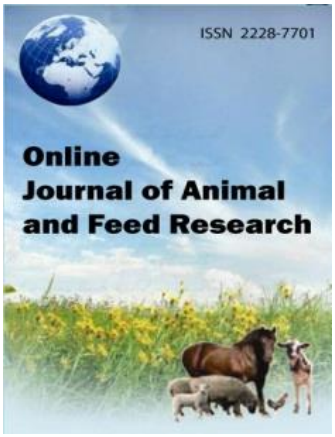

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PREVALENCE OF HEMORRHAGIC SEPTICEMIA IN CATTLE AND BUFFALOES IN TANDOJAM, SINDH, PAKISTAN

Faiza HABIB[1], Adnan JABBAR[2], Rehana SHAHNAWAZ[3], Azizullah MEMON[4], Adnan YOUSAF[2]✉, Muhammad BILAL[1], Tahseen JAMIL[5], Rabia KHALIL[2] and Adeela SHARIF[2]

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ABSTRACT: The current study was performed to evaluate the prevalence of hemorrhagic septicemia (HS) in cattle and buffaloes in Tandojam Pakistan. The average geometric mean titers (GMT) recorded against HS in diseased buffaloes and cattle were 5.7 and 6.1, respectively. The morbidity, mortality and case fatality rates were 57.58, 52.30 and 90.83% in young buffalo calves; and 3.17, 1.92 and 60.65% in adult buffaloes, respectively. In case of young cattle calves, morbidity, mortality and case fatality rates were 8.63, 5.27 and 61.11%, respectively, while in adult cattle, these values were 4.83, 2.18 and 45.23%, respectively. The present study revealed that the mortality, morbidity and case fatality rates due to HS were greater in young calves than the adults both in buffaloes and cattle. Furthermore, buffaloes were found to be more susceptible to the disease than the cattle.

Keywords: Buffaloes, Cattle, Hemorrhagic septicemia, Prevalence, Tandojam

SHORT COMMUNICATION

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INTRODUCTION

Hemorrhagic septicemia (HS) has a wide distribution particularly in tropical countries. In Asia, HS epidemics may occur as an alarming and devastating disease in cattle and buffaloes, jeopardizing the economic return of the animal to a dangerous extent (Benkirane and De Alwis, 2012). Buffaloes are considered to be more susceptible than the cattle. It is an acute pasteurellosis manifested by a highly fatal septicemia with the causative agent being *Pasteurella multocida* serotype B: 2 (Wijewardana, 2014). Radical changes in weather, including the advent of monsoon, debility caused by seasonal level of low nutrition and work pressure are some of the predisposing factors which ignite the occurrence of the disease in Pakistan (Farooq et al., 2007). Clinical manifestations include high rise in body temperature (104-108°F), respiratory distress, nasal discharge, salivation, tongue protrusion, reluctance to move, development of hot painful swelling and edema on throat, brisket or occasionally forelegs.

Studies on the prevalence of HS have been carried out in other regions of the world extensively (Zyambo et al., 2015; Dutta et al., 2016; Molina et al., 2014). However, keeping in view scanty work documented from Pakistan (Sheikh et al., 2016; Khan et al., 2006; Khan et al., 2011), the present study was executed to know prevalence of HS in buffaloes and cattle in Tandojam, Pakistan.

MATERIALS AND METHODS

Ethical approval

This experiment was performed considering to all animal rights (Society for Protection and Care of Animals. Sindh Agriculture University Tandojam, Pakistan)

Selection of site

An active surveillance was undertaken to understand the prevalence of HS in 10 villages of Tandojam, Pakistan The overall climate of the town is dry with scanty rain fall. The winter is not very cold and the climate is hot during the remaining part of the year, but it is very hot in summer. The temperature during summer is usually about 47^{0}C while during winter season the mercury goes down as far as 10°C. The summer season of the town usually touches the one of the highest point of temperature in Pakistan.

Citation: Habib F, Jabbar A, Shahnawaz R, Memon A, Yousaf A, Bilal M, Jamil T, Khalil R and Sharif A (2019). Prevalence of hemorrhagic septicemia in cattle and buffaloes in Tandojam, Sindh, Pakistan. *Online J. Anim. Feed Res*., 9(5): 187-190. www.ojafr.ir

Experimental animals

Ten villages with the infection/outbreak of HS in Tandojam were randomly selected to study surveillance in order to estimate the prevalence. A total of n=4248 animals (2963 buffaloes and 1285 cattle) from these villages were further divided into young (below 1 year) and adult (above 1 year) animals both for buffalo and cattle. Each homestead was visited from door to door in all villages and the relevant information regarding the affected animals (morbidity, mortality and case fatality rates) was recorded on a questionnaire. For comparison purposes, 10 villages (control) free of infection in the same district were also surveyed.

Table 1 - Comparative GMT values against HS in buffaloes and cattle

Species	Group	Distribution of animals on basis of HS titer										
		2^{-0}	2^{-1}	2^{-2}	2^{-3}	2^{-4}	2^{-5}	2^{-6}	2^{-7}	2^{-8}	2^{-9}	GMT
Buffaloes	Affected	11	15	19	31	17	5	2	0	0	0	5.7
	Healthy	0	0	0	9	23	28	32	5	3	0	34.3
Cattle	Affected	8	11	24	22	25	7	3	0	0	0	6.1
	Healthy	0	0	2	11	19	31	29	8	0	0	32.0

Blood collection and serological analysis

Blood samples (6 ml) without anticoagulant were collected from HS affected 100 buffaloes and cattle each. Matched samples from 100 healthy buffaloes and cattle each were also collected which served as control. Serum from each sample was decanted and stored at -20°C until analysis. Antibodies against *Pasteurella multocida* were measured by indirect haemagglutination test (IHA) using human blood group 'O' (Bain et al., 2002). Briefly, two fold dilutions of the test sera starting from 1:5 to 1:640 were made in normal saline using micro titer plates (96 wells) and added 25 µl amounts to all the wells of plate except those of column 11 and 12 which served as control. First four wells (A-D) of column 11 were added with known negative serum and last four wells (E-H) with the known positive serum. Sensitized RBC's (1%) were added in equal amounts (25 µl) to all the wells of the plate, so that column 12 served as control for the RBC's.

The plates were incubated at room temperature for two hours and the observations were recorded. Thereafter, the plates were kept re-incubated under refrigeration for overnight, shaken vigorously, allowed to resettle and were read again. Results were interpreted as positive (no bead formation) or negative (bead formation with sharp clear margins). The IHA antibody titers against HS were converted into geometric mean titers (GMT) for each group (Burgh, 2008).

Statistical analysis

All data were analyzed by using Statistical Analysis System package software (SAS version 9.2, SAS Institute Inc., Cary, NC, USA). The animals (buffalo and cattle) were divided into two age groups i.e. young and adult. Data thus collected regarding mortality, morbidity and case fatality were subjected to Chi square analysis.

RESULTS AND DISCUSSION

Hemorrhagic septicemia is a disease of utmost economic importance particularly in Asia where the susceptible animal population consists of 432 million cattle and 146 million buffaloes, which constitutes 30 and 95% of the world's cattle and buffalo population, respectively (FAO, 2016). In India, during the past four decades, HS is documented to be responsible for 45-55% of all bovine deaths. During the 18 years period from1998 to 2016, it accounted for 58.7% of the aggregate of deaths due to five endemic diseases, *viz*. foot-and-mouth disease (FMD), rinderpest, black quarter, anthrax and HS (Dutta et al., 2016). In an active surveillance study in Sri Lanka, it was shown that in the 1990's, around 15% buffaloes and 8% cattle died of HS annually (Alwis and Vipulasiri, 2001). Similarly, 34.4% of all deaths in susceptible stock (Sheikh et al., 2016) and 31.48% mortality have been reported in buffalo calves (Khan et al., 2011) in Pakistan. The results of this study also clearly indicate that HS is a vital hurdle in the economic uplift of the livestock sector with high incidence rates and alarming morbidity, mortality and case fatality rates. In the present study, the comparative values of GMT against HS, deducted through IHA test, both for buffaloes and cattle are presented in table 1. It was noticed that the GMT value was 5.7 for affected buffaloes in comparison to 34.3 in healthy ones. Similarly, it was 6.1 in affected cattle in contrast to 32.0 in healthy ones. Hence, in diseased buffaloes, the titer was lesser as compared to diseased cattle making them more susceptible to the disease. These results are in line with the findings of Alwis et al. (2001) who have reported a higher GMT values for cattle as compared to those for buffaloes.

Similarly, the mean GMT values of 4.12 and 64.41 for affected and recovered animals have been reported by Khan et al. (2006). In the total population of 4248 animals from 10 infected/outbreak villages, the overall morbidity, mortality and case fatality rates were 17.39, 14.66 and 84.30%, respectively with buffaloes having significantly higher values as compared to cattle (Table 2). In buffalo population, the morbidity, mortality and case fatality rates were 22.30, 19.64 and 88.04%, respectively; however, for the cattle population, these values were 6.07, 3.19 and 52.56%, respectively. These results are in accordance with those of Alwis (2001), who documented overall mortality rate of 45.2 and 15.8% for buffaloes and cattle, respectively. Similarly, Sheikh et al. (2016) have also documented 9% mortality and 78% case fatality rates of HS in buffaloes, whereas these values were 2.5 and 62% in cattle. A mortality rate of 31% has been reported in buffaloes by Suhail et al. (2003) in North Waziristan Agency, Pakistan. Radostits et al. (2005) have reported that the overall mean case fatality for buffaloes is nearly three times as

Citation: Habib F, Jabbar A, Shahnawaz R, Memon A, Yousaf A, Bilal M, Jamil T, Khalil R and Sharif A (2019). Prevalence of hemorrhagic septicemia in cattle and buffaloes in Tandojam, Sindh, Pakistan. *Online J. Anim. Feed Res*., 9(5): 187-190. www.ojafr.ir

high as in cattle. Buffalo has been considered the most susceptible animal to HS throughout the world with highest incidence, morbidity, mortality and case fatality rates. Perhaps, the genetic makeup of the buffalo makes it an ideal host for the causative parasite hence increasing its susceptibility to the disease. Young stock of both buffaloes and cattle was more affected as compared to the adult ones (Table 3). The morbidity, mortality and case fatality rates were significantly higher in young stock. In buffaloes, these values were 57.58, 52.30 and 90.83% for calves; and 3.17, 1.92 and 60.65%, for adult buffaloes, respectively.

Similarly, in case of cattle, the calves had the morbidity, mortality and case fatality rates of 8.63, 5.27 and 61.11%, respectively in comparison to the values of 4.83, 2.18 and 45.23% for adult cattle. These findings coincide with those of Khan et al. (2006), who have also reported that the young stock of both buffaloes and cattle have higher morbidity, mortality and case fatality rates as compared to the older ones. The exhaustion of the maternal immunity against HS after the 60th day of life and delayed vaccination might be attributed to the higher susceptibility of the young calves (Mahmood et al., 2007).

Table 2 - Overall morbidity, mortality and case fatality in buffaloes and cattle affected with HS

Species	Population (n)	Morbidity	Mortality	Case fatality
Buffalo	2963	661 (22.30%)	582 (19.64%)	79 (88.04%)
Cattle	1285	78 (6.07%)	41 (3.19%)	37 (52.56%)
Total	4248	739 (17.39%)	623 (14.66%)	116 (84.30%)
P Value		0.05	0.05	0.001

Table 3 - Morbidity, mortality and case fatality in buffaloes and cattle affected with HS according to their age

Species	Age	Population (n)	Morbidity	Mortality	Case Fatality
Buffalo	Young	1042	600 (57.58%)	545 (52.30%)	55 (90.83%)
	Adult	1921	61 (3.17%)	37 (1.92%)	24 (60.65%)
Cattle	Young	417	36 (8.63%)	22 (5.27%)	14 (61.11%)
	Adult	868	42 (4.83%)	19 (2.18%)	23 (45.23%)
P Value			0.20	0.70	0.50

CONCLUSIONS

It can be concluded that HS is endemically occurring in cattle and buffaloes. Buffaloes are more susceptible to the disease as compared to the cattle. However, the young stock of both is highly affected in terms of morbidity, mortality and case fatality of HS as compared to the adult ones. It is highly recommended that department of livestock should give attention toward such disease in this area to control such huge economical loss.

DECLARATIONS

Competing interests

The authors have declared that no competing interest exists.

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Author's contribution

Dr. Faiza Habib was the main researcher, Dr. Rehana Shahnawaz was research coordinator, Dr. Azizullah Memon was study supervisor, Dr. Adnan Yousaf did all correspondence of this article, Dr. Adnan Jabbar and Dr. Muhammad Bilal Muhammad revised the article, Dr. Tahseen Jamil contributions in statistics, Dr. Rabia Khalil and Dr. Adeela Sharif assisted in results analysis and other activities related to the research.

REFERENCES

Bain RVS, Alwis MCL De, Carter GR and Gupta BK (2002). Hemorrhagic septicemia. FAO Animal Production and Health Paper No. 33, Rome, Italy.

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Benkirane A and Alwis, MCL De (2012). Hemorrhagic septicemia, its significance, prevention and control in Asia. Vet Med-Czech, 47: 234-240.

Burgh MA, (2008). Simple method for recording and analyzing serological data. Avian Dis, 2: 362-365.

Alwis De MCL and Vipulasiri AA, (2001). An epizootiological study of hemorrhagic septicemia in Sri Lanka. Ceylon Vet J, 28: 24–35.

Alwis De MCL, Wijewardana TG, Sivaram A and Vipulasiri AA, (2001). The carrier antibodies status of cattle and buffaloes exposed to hemorrhagic septicemia. Ceylon Vet J, 34: 33-42.

Alwis MCL De, (2001). Mortality among cattle and buffaloes in Sri Lanka due to hemorrhagic septicemia. Trop Anim Health Prod, 13: 195-202.

Dutta J, Rathore BS, Mulick SG, Singh R and Sharma GC, (2016). Epidemiological studies on occurrence of hemorrhagic septicemia in India. Indian Vet J, 67: 893-899.

FAO, (2016). Production Yearbook (2016), FAO, Statistics Division, Rome, Italy.

Citation: Habib F, Jabbar A, Shahnawaz R, Memon A, Yousaf A, Bilal M, Jamil T, Khalil R and Sharif A (2019). Prevalence of hemorrhagic septicemia in cattle and buffaloes in Tandojam, Sindh, Pakistan. *Online J. Anim. Feed Res*., 9(5): 187-190. www.ojafr.ir

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USING KHAT (*Catha edulis*) LEFTOVER MEAL AS FEED FOR SHEEP: ITS IMPLICATION ON FEED INTAKE, DIGESTIBILITY AND GROWTH

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Supporting Information

RESEARCH ARTICLE

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ABSTRACT: Ninety days feeding trial was conducted with the aim to evaluate the impact of replacing concentrate mix with dried khat (*Catha edulis*) leftover meal on feed intake, body weight change and digestibility of Tigray Highland sheep fed a basal diet of mixed grass hay. The trial was carried out using 24 lambs (17.8±1.08 kg) with six blocks and four treatments in a randomized complete block design (RCBD). Concentrates mix was substituted with khat leftover meal at a ratio of 0% (T1), 15% (T2), 30% (T3) and 45% (T4) on DM basis. The dietary rations were formulated in iso-nitrogenous to meet the nutrient requirements of lambs. Data was analyzed by analyses of variance (ANOVA) using the General Linear Model (GLM) procedures of SAS (2008). Treatment means comparison was done using Tukey's HSD test at P<0.05. Dried khat leftover meal had moderate crude protein (12.3%) and less NDF value (31.9%). Substituted concentrates mix with 15% khat leftover meal had better impact (P<0.001) on lamb's total DM, OM, ME, ADL and ADF intake compared to the lambs dietary T3 and T4. Daily weight gain and feed conversion efficiency were significant (P<0.001) till substituted concentrates mix with 30% khat leftover meal. Diet digestibility showed reducing as inclusion level of khat meal increased across treatment. It is concluded that khat leftover meal can be utilized up to 30% without deleterious effect on animal performance and health.

Keywords: Body Weight, Digestibility, Feed Intake, Khat Leftover.

Abbreviations: ADF: Acid detergent fiber; ADL: Acid detergent lignin; ANOVA: Analyses of variance; AOAC: Association of Official Analytical Chemists; ATVET: Agricultural Technical Vocational Education and Training; CP: Crude protein; CSA: Central Statistical Agency; DC: Digestibility Coefficient; DM: Dry matter; FCE: Feed conversion efficiency; GLM: General Linear Model; HSD: Honest Significant Difference; m.a.s.l: Meter above sea level; ME: Metabolizable energy; MR: Mixed ration; MSE: Mean standard error; NDF: Neutral detergent fiber; NFE: Nitrogen free extract; NRC: National Research Council; OM: Organic matter; RCBD: Randomized complete block design; SAS: Statistical Analysis System; SL: Level of significance; TDMI: Total dry matter intake.

INTRODUCTION

The plant Khat (*Catha edulis*) is an evergreen perennial shrub plant that belongs to the *Celastraceae* family and is believed to be originated in Ethiopia (Lemessa, 2001). The plant is widely grown in different parts of Ethiopian including southern zone of Tigray. The total area of land under khat cultivation is estimated at 249, 358.02 hectare in Ethiopia where it has got the status of cash crop (CSA, 2015). A survey done in Raya-Azebo district reported that a household, on average, holds about 0.47 hectare lands covered by khat plantation as source of cash crop (Desalegn, 2017). Khat leftovers include the unused parts of the khat crop: hard leaves, branches and twigs that disposed by the producers, traders and consumers. The leftovers are largely utilized as non-conventional feed resources for livestock especially goats and sheep.

Many research evidences showed the importance of khat as animal feed source (Mekasha et al., 2008; Getinet and Yoseph, 2014; Woldu et al., 2015). The khat leftover leaves have high levels of ash, and nitrogen free extract (NFE) with fiber content expressed as neutral detergent fiber and acid detergent fiber and moderate crude protein (CP) content (Getinet and Yoseph, 2014). Furthermore, khat leftovers contain higher Ca and Mg but lower K and Mn levels than did other feeds (Mekasha et al., 2008). This implies the potential use of khat leftover as alternative feed source for livestock in replacing the expensive commercial feeds and thereby reducing production costs. Despite of its wide availability, however, little research efforts are so far done to see its impact on ruminant feed intake, digestibility and growth performance especially in its meal form. The objective of this study was to evaluate the impact of replacing concentrate mix with dried khat leave meal on feed intake, body weight change and digestibility of Tigray Highland sheep fed a basal diet of mixed grass hay.

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MATERIALS AND METHODS

Descriptions of the study area

The experiment was conducted at Maichew Agricultural Technical Vocational Education and Training (ATVET) College, located at 12º 47' N latitude and 39º 32' East longitude with altitude of 2432 m.a.s.l in southern zone of Tigray, northern Ethiopia. The mean annual rainfall ranges 600 to 800 mm and the average annual minimum and maximum temperature is 12º C and 24º C, respectively. Mixed crop-livestock farming system is the main feature of the study area (Moges, 2015).

Experimental animal managements

Twenty-four uncastrated Tigray Highland lambs (17.8±1.08 kg) were bought from local market. The experimental lambs were quarantined for two weeks to adapt them to the new environment and to observe their health condition before conducting the experiment. During this period, the lambs were drenched with broad spectrum Anthelmintic of 300 mg per lambs against internal parasites, sprayed with 12.5% diazinone against external parasites and were vaccinated against common infectious disease, with 1 ml anthrax vaccine and 1 ml PPR per lambs. The experimental animals were adapted to the feeds, feeding schedule and pen environment for about 14 days.

Feeds and feeding managements

The experimental feeds consisted of mixed grass hay, air dried khat leftover meal and concentrate mixture. The mixed grass hay was chopped, weighed and offered to the lamb's *ad libitum* allowing 20% refusal. Concentrate ingredients (noug seed cake, wheat short, maize grain and sesame seed cake) and salt were purchased from local market. Khat leftovers were collected from khat traders and producers and were air dried for five to six days under shade by spreading on plastic sheets. Further, to make easy for formulation, air dried khat leftovers were crushed and woody parts were removed at Bokra union feed processing. The supplement feeds were offered in two equal portions twice a day (08:00 and 16:00 hours) after the animals well fed hay and clean water. The management practices of all the animals irrespective of groups were similar. The feed supply to the sheep was adjusted every ten days on the basis of their body weight changes.

Experimental design and treatments

The feeding trial was undertaken using RCBD with four treatments and six replications. The lambs were allocated to blocks based on their initial basis of live body weight which was determined by two consecutive weighing after overnight fasting. Each treatment was randomly allocated to the lambs of a given block independently for each block. Dietary feed treatments were arranged in such a way that concentrates mix substituted with khat leftover meal at a ratio of 0% (T1), 15% (T2), 30% (T3), and 45% (T4) on DM basis (Table 1). The experimental treatment diets were offered as gram/sheep/day on DM basis. The dietary rations were formulated to be iso-nitrogenous to meet the nutrient requirements of lambs based on the recommendation of NRC (1984). The trial lasted 90 days with additional 14 days of adaptation periods.

Table 1 - Proportion of ingredients (%) used in formulating the experimental rations

Feed ingredients	Treatments			
	T1	T2	T3	T4
Mixed grass hay	*Ad libitum*	*Ad libitum*	*Ad libitum*	*Ad libitum*
Wheat short	30	13	10	5
Maize grain	17	20	10	2
Noug seed cake	28	11	17	13
Sesame seed cake	22	38	30	32
Mineral salt	3	3	3	3
Total	100	100	100	100

T1= 100% concentrate mix+mixed grass hay; T2 = mixed grass hay + 85 % concentrate mix + 15 % khat leftover meal;T3=mixed grass hay 70% concentrate mix + 30% kh at leftover meal; T4 = mixed grass hay + 55% concentrate mix + 45% khat leftover meal

Data measurements and observations ,Feed intake and feed conversion efficiency

The amount of feed offered and refused for each sheep was measured every day for the whole experimental period. The feed intake was calculated by subtracting the refusal from the offered feed. Nutrient intake was calculated as the difference between nutrients offered and refused. Feed conversion efficiency was calculated as the body weight gain divided by feed intake. Representative feed samples (hay, khat leftover and concentrates mix) were taken and kept for chemical analysis.

Live body weight change

Each sheep was weighed at the beginning of the experimental period and every 10 days throughout the trial. Body

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weight gain was calculated as the difference between final and initial body weight. Average daily gain (g/day) was calculated as the difference between final and initial body weights divided by feeding time.

Diet apparent digestibility measurement

The digestibility trial was conducted following the feeding trial using the same animals, dietary treatments and feeding schedule. The animals were adapted to the carrying fecal bags for three days followed by fecal collection for seven consecutive days. Feces voided was collected daily per animal and weighed every morning before feed offer. Out of the daily fecal excretion, 20% was sampled and pooled to make a composite sample for each animal over the collection period. The fecal samples were stored at -200 in between collections. Fecal samples were dried in an oven at 65C for 48 hours and ground to pass 1 mm screen sieve. The ground samples were stored in an airtight plastic bags pending chemical analysis. Feed intake was recorded daily. A weekly composite sample of each feed and refusal for each animal was taken during digestibility trial. Refusal samples were then pooled per treatment. Body weights at the beginning and end of the digestibility trial was taken for each animal. Apparent digestibility coefficient (DC) of each nutrient (OM, CP, NDF, ADF and ADL) was calculated using the general formula as below (McDonald et al., 2002).

$$\text{Digestibility coefficient (DC) (\%)} = \frac{\text{DM/nutrient consumed} - \text{DM/nutrients excreted in feces}}{\text{DM/nutrients consumed}} \times 100$$

Chemical analysis of samples

Representative samples of feed offered, refusals and feces were ground to pass a 1 mm sieve mesh after drying the samples at 65C for 48 hours. The DM, ash and nitrogen contents of feces were analyzed using the procedure of AOAC (2005) and crude protein (CP) was calculated as N*6.25. Organic matter (OM) was calculated as the difference between 100 and ash content. Neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) were determined through the procedures of Van Soest et al. (1994).

Statistical analysis

Data was analyzed by analyses of variance (ANOVA) using the General Linear Model (GLM) procedures of SAS (2008). Treatment means comparison was done using Tukey's HSD (Honest Significant Difference) test at P<0.001.

RESULTS

Chemical composition of ingredients and experimental rations

The chemical composition of feed ingredients and experimental rations is presented in Table 2. The nutrient content of experimental feeds varied as they came from different sources. The CP content of mixed grass hay (8.99%) was sufficient to meet the maintenance requirement, but it was relatively low to meet the growth demands of experimental animals. Khat leftover meal had medium CP content (12.3%) when measured in view of rumen microbial requirement but it was lower when compared to the commercial feeds: noug seed cake (33.1%), sesame seed cake (31.8%) and wheat short (18.8%). The result also showed that khat leftover meal had lowest NDF content (31.9%) as compared to hay (66.8%), noug seed cake (62.7%), sesame cake (62.8%) and wheat short (42.3%) but higher than that of maize grain (29.1%). The metabolizable energy content of khat leftover meal (7.18 MJ/kg DM) was higher compared to that of sesame seed cake (6.21 MJ/kg DM) but far lower than that of maize grain (13.52 MJ/kg DM) and wheat short (12.32 MJ/kg DM). The ash content of khat leftover was 7.36% which is lower than noug seed cake (11.4%) but higher than wheat short (4.16%).

The CP of the experimental rations was almost similar (21 to 24%) and this was due to the fact that the dietary treatments were made deliberately to have similar protein contents. The NDF content of dietary T1 (0% khat meal) was higher than dietary T2 (15% khat meal), T3 (30% khat meal) and T4 (45% khat meal) in that order showing decrements trend with increase of khat leftover meal along treatments. On the contrary, the ADF and ADL values showed increment with increasing inclusion level of khat meal. The ash amount (7.36% for T1 and 9.26% for T4) showed increment with level of khat meal. The energy concentration was decreased with khat meal level with the highest in T1 (9.45 ME MJ/kg DM) and the lowest in T4 (6.55 ME MJ/kg DM).

Dry matter and nutrient intake

The mean daily DM and nutrient intake of Tigray Highland sheep across the dietary treatments are presented in Table 3. The mixed grass hay DM intake expressed as gram per day was significantly different among the treatments (P<0.001). Sheep with dietary T1 (0% khat meal) and T2 (15% khat meal) had the highest (P<0.001) basal mixed grass hay DM intake than those sheep with T3 (30%) and T4 (45%). However, no significant differences were observed among the sheep in T1 and T2 (P>0.05). The highest total DM, OM and ME intake was recorded for sheep placed under T1 and T2 followed by sheep in T3 and T4 in descending order. But no significant difference was observed in sheep groups fed on T1 and T2 in this regard (P>0.05). The CP and NDF intake showed decreasing across treatments (P<0.001) with increasing inclusion level of khat.

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Body weight change

There was no initial body weight differences (P>0.05) among the experimental sheep. The dietary treatments had no significant effects (P>0.05) on the final live weight and body weight change of the experimental sheep among the T1 (0% Khat leftover meal), T2 (15% Khat leftover meal) and T3 (30% Khat leftover meal) but significantly (P<0.001) lower final weight was recorded in animals fed on T4 (45% Khat leftover meal). Similarly, daily body weight gain did not show significant difference (P>0.05) up to 30% khat meal inclusion level but it was lower in 45% (T1) (P<0.001). Feed conversion efficiency was not significantly different amongst T1, T2 and T3 (P>0.05) but it was lower for T4 (P<0.05).

Apparent nutrient digestibility

Dry matter and nutrient digestibility coefficients of the experimental diets are presented in Table 5. There was significant difference among the treatments in DM and nutrient digestibility (P<0.05). Sheep with dietary T1 and T2 had higher (P<0.001) DM, OM and CP digestibility coefficients than dietary T3 and T4. Likewise, sheep fed on dietary T1 and T2 had higher NDF and ADF apparent digestibility coefficient than T3 and T4. However, there was no significant difference among dietary T1 and T2 in DM, OM, CP, NDF and ADF apparent digestibility coefficient (P>0.05).

Table 1 - Chemical composition of feed ingredients and experimental diets

Feed type / Feed ingredients	DM%	OM	Ash	CP	NDF	ADF	ADL	ME(MJ/kg)
Mixed grass hay	94.67	90.67	9.33	8.99	66.8	40.5	5.38	8.16
Sesame cake	96.82	90.09	9.91	31.8	62.8	29.6	8.25	6.21
Maize grain	94.86	96.49	3.51	7.09	29.1	4.71	2.70	13.52
Noug seed cake	95.84	88.26	11.4	33.1	62.7	29.5	9.23	7.03
Wheat short	94.35	95.84	4.16	18.8	42.3	6.57	1.19	12.32
Khat leftover meal	88.91	92.64	7.36	12.3	31.9	26.1	10.19	7.18
Treatment rations								
MR-T1	94.00	92.53	7.47	24.5	54.6	18.5	5.31	9.45
MR-T2	94.00	91.72	8.28	21.2	52.0	23.7	7.36	8.48
MR-T3	94.00	90.76	9.24	20.8	46.5	27.7	9.59	7.01
MR-T4	94.00	90.74	9.26	21.6	48.8	28.4	10.10	6.55

DM= dry matter; OM= organic matter; CP= crude protein; NDF = neutral detergent fiber; ADF = acid detergent fiber; ADL = acid detergent lignin; MR-T1= mixed ration treatment one; MR-T2= mixed ration treatment two; MR-T3= mixed ration treatment three; MR-T4= mixed ration treatment four.

Table 2 :Dry matter and nutrient intake of Tigray Highland sheep consumed mixed grass hay and supplemented with concentrate, khat leftover meal or their mixtures

Intake (g/day)	Treatments					
	T1	T2	T3	T4	MSE	SL
Hay DM	495.99^{a}	479.27^{ba}	467.50^{b}	365.81^{c}	12	**
Supplement DM	303.70^{a}	303.70^{a}	303.70^{a}	303.70^{a}	0	ns
Total DM	799.77^{a}	783.04^{ba}	771.28^{b}	650.09^{c}	12	**
Ash	68.97^{b}	69.87^{ba}	71.68^{a}	60.44^{b}	1	**
OM	730.77^{a}	713.15^{ba}	699.57^{b}	589.63^{c}	11	**
CP	117.97^{a}	108.77^{b}	103.96^{c}	95.7^{d}	1	**
NDF	497.18^{a}	478.12^{b}	453.55^{c}	379.58^{d}	8	**
ADF	257.08^{b}	266.1^{a}	273.48^{a}	226.53^{c}	5	**
ADL	42.8^{d}	48.14^{c}	54.28^{a}	49.31^{b}	0.6	**
ME(MJ/Kg)	69.18^{a}	64.88^{ba}	59.45^{c}	48.17^{d}	1	**

^{abcd}means within a row with different superscript letters are significantly different at ** p<0.001; ns= non-significant; DM= dry matter; OM= organic matter; CP= crude protein; NDF = neutral detergent fiber; ADF = acid detergent fiber; ADL = acid detergent lignin; MSE= mean standard error; SL= level of significance; T1= 100% concentrate mix + mixed grass hay; T2 = mixed grass hay + 85% concentrate mix + 15% Khat leftover meal; T3 = mixed grass hay+ 70% concentrate mix + 30% Khat leftover meal; T4 = mixed grass hay + 55% concentrate mix + 45% Khat leftover meal.

Table 3 - Body weight change and feed conversion efficiency of Tigary Highland sheep consumed mixed grass hay and supplemented with concentrate mixture, Khat leftover meal or their mixture

Parameters / Treatments	T1	T2	T3	T4	MSE	SL
Initial body weight (kg)	17.66^{a}	17.92^{a}	17.83^{a}	17.83^{a}	0.33	ns
Final body weight (kg)	22.25^{a}	22.33^{a}	22.16^{a}	20.9^{b}	0.51	**
Total body weight change (kg)	4.60^{a}	4.42^{a}	4.33^{a}	3.083^{b}	0.67	**
Average daily gain (g/day/head)	56.6^{a}	54.5^{a}	53.5^{a}	38.06^{b}	4.5	**
FCE (ADG/TDMI)	0.07^{a}	0.069^{a}	0.069^{a}	0.058^{b}	0.06	*

^{ab}means within a row with different superscript letters are significantly different at **p<0.001; *p<0.05; ns= non-significant; ADG= Average daily weight gain; FCE= Feed conversion efficiency; TDMI= total dry matter intake MSE= mean standard error; SL= level of significance; T1= 100% concentrate mix + mixed grass hay; T2= mixed grass hay + 85% concentrate mix + 15% Khat leftover meal; T3= mixed grass hay + 70% concentrate mix + 30% Khat leftover meal; T4= mixed grass hay + 55% concentrate mix + 45% Khat leftover meal.

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Table 4 - Dry matter and nutrient apparent digestibility coefficient (%) of Tigray Highland sheep fed rations containing different levels of Khat leftover meal.

Digestibility \ Treatments	T1	T2	T3	T4	MSE	SL
DM	62.59[a]	59.04[ba]	55.74[c]	50.89[c]	2.5	**
OM	69.65[a]	69.57[a]	66.44[b]	64.34[b]	1.8	**
CP	87.22[a]	84.98[b]	83.28[c]	81.95[c]	0.92	**
NDF	63.72[b]	69.59[a]	50.62[c]	44.88[d]	2.5	**
ADF	39.15[b]	47.67[a]	39.15[b]	27.44[c]	3.5	**

[Abcd]means within a row with different superscript letters are significantly different at **p<0.001; DM= dry matter; OM= organic matter; CP= crude protein; NDF= neutral detergent fiber; ADF= acid detergent fiber; MSE= mean standard error; SL= level of significance.

DISCUSSIONS

The dry matter (DM), organic matter (OM), neutral detergent fiber (NDF), acid detergent fiber (ADF) and ash content of mixed grass hay used in the present experiment was almost similar with other authors (Alemu et al., 2014; Gebru and Tesfay, 2017). The DM, CP and ash contents of the commercial feeds: noug seed cake, sesame cake and wheat short were comparable with previous studies (Gebru and Tesfay, 2017; Mekasha et al., 2008).

The DM, OM and ADF contents of khat leftover meal seen in this study was comparable with other research work (Getinet and Yoseph, 2014). However; the CP content was higher than the value reported by the same authors (Getinet and Yoseph, 2014). Khat leftover meal has moderate crude protein content (12.3%) indicating its potential use as protein supplementation for the roughage-based ruminant animals. The medium CP value of khat meal is believed to be sufficient for rumen function (McDonald et al., 2002). The lower fiber value of khat meal as compared to the basal feed (hay) and commercial concentrate can enhance diet digestibility (McDonald et al., 2002). The NDF, ADL and ash content were less than the value reported by Getinet and Yoseph (2014) for khat meal and Gebru and Tesfay (2017) for *Acacia saligna* pod meal. This variation could be raised from maturity and species of the khat plant, soil fertility where the khat plant grown and the season of the leaf harvest. The energy value of khat meal (7.18 ME MJ/kg DM) was slightly higher than sesame cake (6.21 ME MJ/kg DM) but lower than maize grain (13.52 ME MJ/kg DM).

All the dietary treatments had almost similar CP value which was made intentionally to create iso-nitrogenous rations. The fiber content (NDF) showed reducing across treatments (T1 to T4) with increasing inclusion level of khat meal. This is attributed to the lower fiber content of khat meal when compared to commercial feeds. Similarly, the increased ash amount across treatment is attributed to the higher ash content of khat meal. On the other hand, the reducing trend of energy value (9.45 to 6.55 ME MJ/kg DM) along treatments is apparently related to the lower energy content of khat meal as compared to commercial feeds.

Basal mixed hay DM intake in this study was higher than the value reported by Getinet and Yoseph (2014). All experimental sheep consumed the dietary supplement without any refusal across the experiment. The higher feed and nutrient intake in sheep groups in T1 (0% khat meal) and T2 (15% khat meal) could be attributed to the lower ADF and higher energy contents. Similarly, by increasing the total amount of concentrate offered, cows had higher DM and energy intake, which resulted in increased milk production and reduced negative energy balance and body condition score loss (Lawrence et al., 2015). Jung and Allen (1995) also insisted that cell-wall concentration and digestibility limit the intake potential and energy availability of forage crops in ruminant production. The indifference in DM intake between T1 and T2 might be explained by their similar apparent digestibility. The study showed decrease in dry matter intake with increasing level of khat leftover meal across the treatments. This could be explained by the higher ADF and ADL content of khat meal. Likewise, decreased DM intake was reported by Getinet and Yoseph (2014) in goats fed on 100% khat leftover meal. Conversely, increased feed intake and digestibility was seen in Somali goats when sorghum stover was replaced by khat leftover meal up to 60% on DM base (Tamir and Ismail, 2006).

Sheep placed in dietary T1, T2 and T3 had heavier total body weight change and average daily weight gain than sheep in dietary T4 and the difference might be most likely due to the reflection dense nutrient content (energy concentration) and higher feed intake. Mahgoub et al. (2000) reported that sheep fed high energy diet had heavier body weight than sheep fed medium and low energy diets. In support of this finding, Wallie et al. (2012) obtained that mixtures of different proportions of dried khat leftover meal and concentrate supplementation resulted in differences in daily body weight gain in goat. The same authors added that feeding leftover khat to goats consuming low to moderate quality forage-based diets can increase growth performance. Supplementation of dried khat leftover meal up to 45 % of the diet DM has no deleterious effects on the performance and health of sheep. The daily body weight gain (53 to 56 g/day) was comparable to 63.89 g/day gain of Tigray Highland ram fed on mixed grass hay *ad libitum* and supplemented with 200 g of air dried *Acacia saligna* leaves and 200 g wheat bran (Gebru and Tesfay, 2017) and value reported by Getinet and Yoseph (2014).

The apparent digestibility coefficient of dry matter and nutrients showed decrements trend with increase of khat leftover meal level across dietary treatments. Nutrient apparent digestibility decline was observed when moving from dietary T1 and T2 to dietary T3 and T4 fed groups. This might be related to the higher cell wall fraction contents of dietary T3 and T4 as well as negative association effect of lower nutrient intake. McDonald et al. (2002) noted that the fiber fraction of a food has the greatest influence on its digestibility. However, there was no significant difference among

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dietary T1 and T2 in DM, OM and CP apparent digestibility coefficient and this was mainly due to positive association effect and similarity in their total DM and nutrient intake. It has long been recognized that in ruminants there is a positive relationship between the digestibility of foods and their intake (McDonald et al.,, 2002).

CONCLUSIONS

The study showed that khat leftover meal has moderate crude protein content indicating its potential use as protein supplementation for the roughage-based ruminant animals. No significant change was observed in feed intake and growth performance with increasing levels of khat meal substitution till 30% to concentrate mixture. This implies that khat leftover meal can be utilized as alternative option in the animal feeding system. Hence, it can be concluded that khat leftover meal can be included up to 30% in the animal ration without negatively affecting performance and health of ruminant animals.

DECLARATIONS

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Competing interest

The authors declare that they have no competing interests.

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REFERENCES

Alemu B, Animut G and Tolera A (2014). Effect of Millettia ferruginea (Birbra) foliage supplementation on feed intake, digestibility, body weight change and carcass characterstics of Washera sheep fed natural pasture mixed grass hay basal diet. SpringerPlus, 3(1):50. DOI: 10.1186/2193-1801-3-50

AOAC (2005). Official Methods of Analysis. 18th Ed, Association of Official Analytical Chemists, Washington DC, Method 935.14 and 992.24.

CSA (Central Statistical Agency) (2015). Report on area and production of major crops. Volume I, Statistical bulletin 278, Addis Ababa, Ethiopia

Desalegn G (2017). Comparative assessment of biomass and soil carbon stock in multistrata agroforestry system and chat: the case of Raya Azebo, Tigray, Ethiopia. An MSc. Thesis Presented to School of Graduate Studies of Hawasa University. Dire-Dawa, Ethiopia.

Gebru G and Tesfay Y (2017). Utilization of wheat bran and dried Acacia saligna (*Labill*) HL Wendl leaves by highland rams. African Journal of Agricultural Research, 12(15): 1286 - 1292. DOI: 10.5897/AJAR2015.10376

Getinet Y and Yoseph M (2014). Effect of feeding concentrate, dried khat (*catha edulis*) leftover or their mixtures on feed intake, digestibility and body weight change of Hararge highland goats fed basal diet of natural grass hay. The Journal of Animal and Plant Sciences, 24(1): 35 - 42.

Jung H and Allen M (1995). Characteristics of plant cell walls affecting intake and digestibility of forages by ruminants. Journal of Animal Science, 73(9): 2774 - 2790. DOI: 10.2527/1995. 7392774x

Lawrence D, O'donovan M, Boland T, Lewis E and Kennedy E (2015). The effect of concentrate feeding amount and feeding strategy on milk production, dry matter intake and energy partitioning of autumn-calving Holstein-Friesian cows. Journal of Dairy Science, 98(1): 338 - 348. DOI: 10.3168/jds.2014-7905

Lemessa D (2001). Khat (*Catha edulis*): botany, distribution, cultivation, usage and economics in Ethiopia. Addis Ababa: UN-Emergencies Unit for Ethiopia.

Mahgoub O, Lu C and Early R (2000). Effects of dietary energy density on feed intake, body weight gain and carcass chemical composition of Omani growing sheep. Small Ruminant Research, 37: 35 - 42. DOI: 10.1016/S0921-4488(99)00132-7

McDonald P, Edwards RA, Greenhalgh JFD and Morgan CA (2002). Animal Nutrition, 6th ed., Prentice Hall, London. Pp 464 - 479.

Mekasha Y, Tegegne A and Rodriguez-Martinez H (2008). Feed Intake and Sperm Morphology in Ogaden Bucks Supplemented with Either Agro-industrial By-products or Khat (*Catha edulis*) Leftover. Reproduction in Domestic Animals, 43(4): 437 - 444. DOI: 10.1111/j.1439-0531.2007.00931.x

Moges A (2015). Effects of different nitrogen rate and plant population density on yield and yield related traits. A MSc. Thesis Presented to School of Graduate Studies of Haramaya University. Dire-Dawa, Ethiopia.

NRC (National Research Council) (1984). Nutrient Requirements for Sheep: 6th Revised Edition, National Academy Press, Washington DC, USA.

SAS (Statistical Analysis System) (2008). SAS/STAT. 9.2 user's guide. Cary, NC. SAS Institute Inc. United States of America.

Tamir B and Ismail M (2006). Effect of supplementing different levels of leftover of khat (*Catha edulis*) to sorghum stover on nutrient intake and digestibility by goats. Tropical Science, 46(4): 213 - 215. DOI: 10.1002/ts.180

Van Soest PJ (1994). Nutritional ecology of the ruminant, Cornell University Press.

Wallie M, Mekasha Y, Urge M, Abebe G and Goetsch A (2012). Effects of form of leftover khat (*Catha edulis*) on feed intake, digestion and growth performance of Hararge Highland goats. Small Ruminant Research, 102(1): 1 - 6. DOI: 10.1016/j.smallrumres.2011.07.014

Woldu Z, Belew D and Benti T (2015). The coffee-khat interface in Eastern Ethiopia: a controversial land use and livelihood change scenario. Journal of Agriculture Science and Technology, 5(3): 149 - 169. DOI: 10.17265/2161-6264/2015.03B.001

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PREVALENCE OF PARASITIC HONEYBEE DISEASES, PESTS AND PREDATORS IN NORTH GONDAR ZONE

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ABSTRACT: A cross-sectional study was conducted on parasitic honeybee diseases, pests and predators Lay armachiho and Tach armachiho districts of North Gondar zone from October 2017 to May 2018. The objectives of this study were to assess the different parasitic honeybee diseases, pests and predators. The study mainly involves with the collection of adult honeybee abdominal suspension of wet mount for nosema and amoeba diseases and direct observation of varroa mite and bee louse from adult honey bee and brood cells. Questioner survey was used to know different honeybee pests, predators and agrochemicals in the study area. The SPSS version 20 was used for chi-square test and p-value < 0.05 was taken as statistical significant. During the study period a total of 384 honey bee sample in traditional, modern and transitional hives were examined. The overall prevalence 24.47% (94/384) of hives were found positive for *Nosema apis*, 17.2% (66/384) foramoeba (*malphighamoeba mellificae*), 30.5% (117/384) for *varroa destructor* and 37.5% (144/384) for bee louse (*braula cocae*). Bee louse was the predominant external parasitic disease in adult honeybees followed by *varroa destructor*. There was statistically significant variation between *Nosema Apis* and bee louse observed among the two selected districts and hive types (x^2=23.5, p-value=0.0001) for bee louse and (x^2=5.3, p-value=0.071) for *Nosema Apis*. About the 100 respondents are 46%, 42%, 39%, 35%, 28%, small hive beetle 26% and spider 24% were complaining on the impact of wax moth, chemical spray, skunk, birds, small hive beetle and spiders respectively. They have also responded that pests play a major role on the production loss, damaging of honey bee colony and absconding of the colony. In conclusion, the highest prevalence parasitic honeybee diseases and pests were recorded in the study areas that signify the occurrence of the parasitic burden has to be carried out and immediate intervention was implemented.

Keywords: Honeybees, Parasites, Pests, Traditional And Modern

INTRODUCTION

The essential and valuable contributions of honeybees depend upon the healthy population of honeybees (FAO, 2012). The health of honeybees has been one of the most important topics in apiculture research in recent years (Genersch, 2010). This is mainly associated with the recent emergence of high honeybee colony losses in many parts of the world and the vulnerability of honeybees to parasitic mites, fungi, viruses and bacteria (Bradbear, 2009). These pathogens and parasites can have harmful effects on honeybee health and the services they offer, which in turn can lead to severe economic losses (Genersch, 2010).

Moreover, modern agriculture increasingly depends on the use of chemical substances to control weeds, fungi and arthropod pests to ensure high yields. Honey bees may frequently become exposed to environmental chemicals as a consequence of their foraging activities (Vanengelsdorp and Meixner, 2010). It has been reported that several biological and environmental factors acting alone or in combination have the potential to cause premature colony mortality. In United States the average honeybee loss per beekeeping operation was 25.4% (Spleen et al., 2013). Similarly, 16 % honeybee colony reduction has been reported in Europe (Hendrikx et al. 2010; Potts et al., 2010).

The ectoparasitic mite *Varroa destructor* as well as the bee pathogenic viruses have been identified as a marker of dramatic colony losses termed colony collapse disorder in the USA, the *Microsporidium nosema* species and bacterial diseases are causing economic losses to beekeepers worldwide (Genersch, 2008). Even though, the majority of pathogens and parasites affecting honeybees have an almost worldwide distribution (Ellis and Munn, 2005).

The most commonly known honeybee diseases reported to exist in Ethiopia are *Nosema Apis* and *Melpighamoeba mellificae* (Amssalu Bezabeh and Desalegn Begna, 2005).To this effect, honeybee diseases are report to be among the major constraints in beekeeping (Workneh Abebe and Puskur, 2011). The identification and severity of each economically important honeybee disease have not been well documented in amhara region, despite little information is available. To fully exploit the opportunities in beekeeping sector, addressing the constraint and detecting the occurrence and distributions of honeybee's health problems is key step to prevent their harmful effects. Therefore, the objectives of this study were: A) to determine the occurrence and prevalence of parasitic honeybee diseases and pests; B) To determine the

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effect of honeybee disease on honey bee colonies and their products; C) To assess the different risk factors associated with honeybee parasitic disease, pest and predators of the honeybees.

MATERIALS AND METHODS

Description of Study area

The study was conducted in the selected district of north Gondar zone, north east of Ethiopia, (lay armachiho and tach armachiho) from October 2017 to May 2018. These areas are located 763, 810 kms from Addis Ababa respectively. The climatic condition of the study areas are highland and kola and its annual temperature range from 10°c-40°c respectively. The study areas are located at 13°N latitude and 37° 10, E longitude. The elevation of in lay armachiho 1730 and tach armachiho 950-1100 m.a.s.l and its annual rain fall in tach armachiho and lay armachiho is 300-750mm with slight rain in April and May and heavy rain in July and august.

Study design

A cross-sectional study was conducted on honeybee colonies to assess the prevalence of common parasitic honeybee disease, pest and predator by using microscopic examination for protozoa parasite and macroscopic examination for Varroa mite and bee louse from adult worker honey bee. Semi structured questionnaire was administered from each apiarist to assess honeybee parasitic disease, pest and predator in the study area.

Study population

The study districts have traditional, modern and transitional types of hive in the north Gondar zone and most of bee hives are managed under traditional system. The study was conducted in all types of honeybee hives to estimate the prevalence of parasitic honeybee diseases, predators and pest. The honeybee colonies were selected randomly and collection of these adult bees were at the entrance of honeybee hives for Varroa mites. The colonies were randomly selected from the study areas 10-20 bees were taken from the top frames and hive entrances of each of these colonies and placed in separate transparent sample bottle. The collected bees were immediately immersed in to 10% formalin or 70% ethanol during sample transportation. Sample size determination and sampling method

The required sample size for this study was estimated by assuming expected prevalence of 50%, and study on parasitic honey bee disease and pest in the study area. Thus the sample size collected according to Thrusfield (2005) using 95% confidence interval at 0.05% absolute precision and calculated by the following formula.

$$n = \frac{(Za)^2 \times Pexp \times (1-Pexp)}{d^2}$$

Where, n = required sample size, Pexp= Expected prevalence (50 %), d= Desired absolute Precision (5 %), Za=confidence level, 95%=1.96

By using 50% expected prevalence with 95% confidence interval at 5% absolute precision (Thrusfield, 2005). The number of hives required estimating the prevalence of honeybee disease and pest was calculated to be 384. A total of 384 bee colonies were selected by randomly and collected honey bees from sampling sites. The sample was collected during at night time to reduce disturbance of bees in the environment. After wearing of protective cloth and beekeeper glove samples were collected from the selected hive in a jar by using bee brush and preserve70% ethyl alcohol or 10% formalin and labeled immediately.

Study methodology

Microscopically examination for nosema and amoeba

For quick qualitative examination of nosema spores and amoeba cysts, the abdomen from at least 10 sample bees were remove, place in pistle and crush using mortar and pistle by adding distle water (1 ml/ abdomen) and homogenize. A wet mount was prepared from the resulting suspension and the existence or non-existence of nosema spore was examined by 100x and used oil immersion for magnification of the field.

Macroscopic examination procedure for varroa mite and bee louse

Varroa mite is dislodging by shaking the sample bees in liquid such as water, 70% alcohol, detergent solution and 10% formalin. Hand shaking bees in alcohol for 1 minute dislodge about 90% of the mite. The mite was directly observed by the naked eye from the transparent sample bottle or beaker, the parasite settle at bottom of beaker and the sample adult bees were float above liquid.

In bee louse the sample were examined using shaking method, bees sample preserved were taken and detergent solution (10 ml of detergent is used to 1000ml of detergent-water solution) was poured into each of jar containing bees up to half of the jar get full. Then shaking for one minute until the lice dislodged from adult honeybees. Straining the solution through a ladle (8-12 mesh) to remove the bees and then sieving the solution through tea strainer to collect bee lice. The tea strainer was examined for presence of bee lice either by necked eye or by using hand lens and count bee lice (Cramp, 2008).

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Questioner survey and regular visits

The questioner was conducted to acquire information on observing clinical sign of honey bee disease, pest, management (feeding, watering, cleaning), types of hives, absconding, and related problem. These questioner surveys were collected from apiarists and extension workers of the districts using structured interview, observations and personal interviews.

Data management and analysis

All data was entered in to Ms- Excel spread sheets after the completion of data collection work from the study areas. Then, the analysis was done by using SPSS version 20, while the result was summarized by using descriptive statistics (means, standard errors and percentages).

RESULTS

Laboratory finding

A total of 384 honey bee colonies were examined and 24.47% (94/384) were positive for *Nosema APIs*, 17.2 % (66/384) for amoeba disease (*Malphighamoeba mellificae*), 30.5% (117/384) for varroa mite (*varroa destructor* or *varroa jacobsoni*) and 37.5% (144/384) for bee louse (*braula coeca*). Bee louse was the highest external parasitic disease in adult honey bees and followed by varroa mite in the current study. Both protozoan parasitic honey bee disease (*Nosema APIs* and *Malphighamoeba mellificae*) were found in the abdominal contents of the adult honey bee colonies. The prevalence of all bee hives examined for parasitic honey bee disease and pests contributed from two selected districts in north Gondar zone (tach armachiho and lay armachiho).

Risk factors

The assessment of the risk factors by using questionnaire and regular visits in two districts showed that hive type in the apiarists used and the study districts were the major predisposing factor. But these risk factors were not found affecting the prevalence of parasitic honey bee diseases statistically except bee louse.

Districts

During the study period the prevalence of nosema, amoeba, varroa mite and bee louse of parasitic honey bee disease in tach armachiho and lay armachiho were presented in (Table 1). The prevalence of Nosema apis in these two selected districts are 22.8% and 26.1% respectively, however there was no statistically significance difference (P>0.05) between the study area.

Hive type

Among 201 traditional bee hives, 123 modern bee hive and 60 transitional bee hives examined 27.9%, 18.7% and 16.7% were positive for nosema apis respectively. Even though the higher prevalence of *Nosema Apis* was observed in traditional bee hives, it was not significantly difference (P>0.05) (Figure 1).

Prevalence of *Malpighamoeba mellificae* was 15.2% and 19% in tach armachiho and lay armachiho respectively (Table 1). The prevalence of Malpighamoeba mellificae almost similar in two districts and there was no statistical significance difference (P>0.05) between two districts. Of 201 traditional hives, 123 modern and 60 transitional hives examined 16.9%, 11.4% and 13.3%were positive for *Malpighamoeba mellificae* respectively as seen from (Figure 2). Even if higher prevalence of *Malpighamoeba mellificae* was observed in traditional bee hives the association between hive type and the occurrence of Malpighamoeba mellificae was not significance difference (P>0.05). During the study period, the prevalence of varroa mite and bee louse in two districts was recorded in the result. The result revealed that the prevalence of bee louse was 35% and 40.2% respectively (Table 1). Relatively highest prevalence of bee louse was observed in tach armachiho than lay armachiho. However, the association between the districts and causative agent were statistical significance (P<0.05). The prevalence of varroa destructor in these two districts was 28.8% and 32% respectively (Table 1). However, the association was not statistical significance (P>0.05). Origin and hive type were the two most important risk factors for the occurrence of honey bee mite. From 201 traditional hive, 123 modern hives and 60 transitional hive examined 26.4%, 17.8% and 18.3% varroa destructor positive colonies and 37.3%, 32.5% and 33.3% bee louse positive colony were found respectively (Table 2) and the higher prevalence of bee louse and varroa mite was observed in traditional hive and the association result in the case of bee louse.

Prevalence of honeybee pests, predators and pesticides

Based on this, the existence of pests was the major challenge to the honeybees and beekeepers. After identifying the major pest facing the beekeeping activities, the prevalence of major bee enemies in two selected districts of north Gondar were listed according to farmer perception (Table 3). From the lists of pests and predators in the study area wax moth and honey badger were found significantly affecting the honey bee hives but the other pests and predators was not significant.

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Table 1 - Prevalence of Nosema APIs, M. Mellificae, varroa mite and bee louse in two districts of north Gondar

Districts / Honey bee parasite	Tach armachiho		Lay armachiho		Total	
	Negative	Positive	Negative	Positive	Negative	Positive
Nosema [a]	136(73.9%)	42(22.8%)	154(77%)	50(26%)	290(75.5%)	94(24.47%)
Amoeba [b]	156(85%)	28(15.2%)	162(81%)	38(19%)	318(83%)	66(17.2%)
Varroa mite [c]	131(71.2%)	53(28.8%)	136(68%)	64(32%)	267(69.5%)	117(30.5%)
Bee louse [d]	110(60%)	74(40.2%)	130(70.6%)	70(35%)	240(62.5%)	144(37.5%)

[a]X^2=0.494, *df* =1, p-value=0.482; [b]x^2=0.401, *df* =1, p-value=0.527; [c]x^2 =0.462, df = 1, p-value=0.467 and [d]x^2=23.5, df=1, p-value=0.000

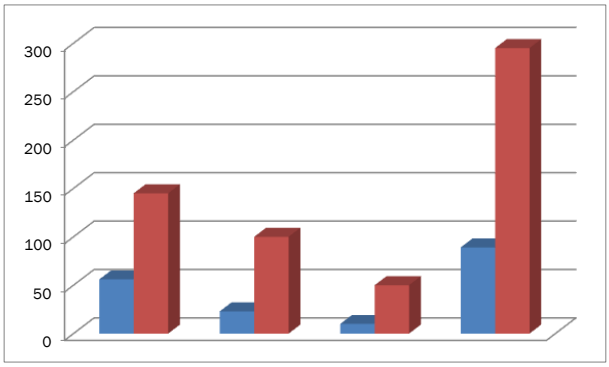

Figure 1 - Nosema Apis is showed more prevalent in traditional hives than modern; hiveX^2=5.3, df =2, and p-value=0.071

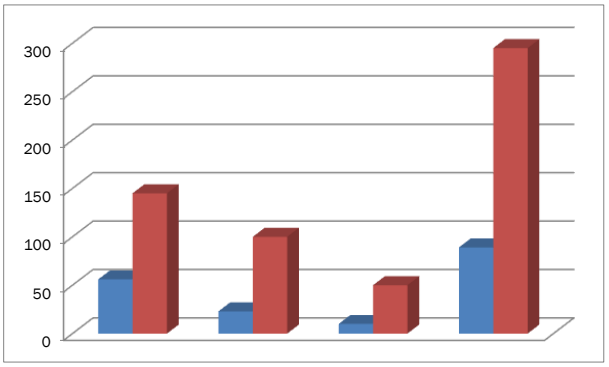

Figure 2 - Prevalence of amoeba in traditional hives is higher than modern hives; X^2=1.96, df=2, and p-value=0.374

Table 2 - Prevalence of varroa mite (varroa destructor) and bee louse (braula coeca) in three types of honeybee hives from two selected districts of north Gondar zone.

Causative agent / Hive type		Traditional	Modern	Transitional	Total
Varroa destructor [a]	Positive	53(26.4%)	22(17.8%)	11(18.3%)	96(25%)
	Negative	148(73.6%)	101(82%)	49(81.6%)	355(92.4%)
Bee louse [b]	Positive	75(37.3%)	40(32.5%)	20(33%)	135(35.2%)
	Negative	126(62.7%)	83(67.5%)	40(66.7%)	249(64.8%)

[a]x^2=11.98, df=2 and p-value=0.003; [b]x^2=0.21, df =2 and p-value=0.98

Table 3 - Farmer perception on the Prevalence of honeybee pests, predators and use of chemicals

Major pests and predators	Total sample (n=100)	Positive	Chi-square(X^2)	df	p-value
Wax moth	100	46(46%)	7.8	1	0.005
Spider	100	24%	0.16	1	0.68
Small beetles	100	26%	1.46	1	0.22
Ant	100	28%	0.12	1	0.81
Birds	100	35%	1.99	1	0.15
Lizard	100	16%	0.04	1	0.84
Chemical spray	100	42%	0.65	1	0.41
Honey badger	100	19%	5.26	1	0.02
Qunchichr	100	23%	0.17	1	0.68
Skunk	100	39%	0.04	1	0.82

DISCUSSIONS

The prevalence of nosema apis reported in the present study was relatively lower than previous reports in Ethiopia was reported from different regions with varying prevalence ranges such as 58% in Oromia, 60% Benishangul-Gumuz and 47% in Amhara regions by Aster Yohannes et al. (2010) and other countries such as Nigeria (64.29%) by Akinwande et al. (2013), Kenya 83.3% by Fazier et al. (2010) and Turkey 65.25% by Aydin et al. (2001) and Diagnosis made on honey bees

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in field and laboratory at Addis Ababa reported a prevalence rate of 73% of amoeba prevalence. The diseases was also reported with high prevalence rate in different regional state of Ethiopia such as; Oromia region with prevalence rate (88%), Amhara region (95%) and 60 % in Benishangul- Gumuz by Aster Yohannes et al. (2010).

The prevalence of parasitic honey bee disease in the selected districts is relatively higher in traditional bee hive than modern and transitional hive type. The result of this study indicated that either origin or hive type not significance influence in prevalence of parasitic honey bee disease in the study area except bee louse which was strong statistical significance in the origin (P=0.00) with 35.5% prevalence. The difference prevalence of the study area of the disease may be colony management of the beekeepers during swarming, extraction of honey and stress condition. Tach armachiho districts have relatively less prevalence in Nosema Apis (23%) and 28.8% of varroa destructor as compared to lay armachiho districts, which have high prevalence. This is due to dry climatic condition which crates less favorable condition. In lay armachiho districts the honeybee beekeepers were contamination of colonies with honey extracting equipment was observed that aggravates the spread of protozoan parasitic disease. In addition to this less awareness about beekeepers for honey bee colony and less practice of modern hive type was the cause of increasing parasitic infection rate in the study area. The observed overall prevalence of Varroa mite and bee louse in the study area was lower than in the reported in some African countries such as Tanzania and Uganda 84.6% by Fazier et al. (2010), Nigeria 78.6% by Akinwande et al. (2013), and 100% in Jordan by Alattal et al. (2006) and the overall prevalence of bee lice observed in the current study was much greater than the previous reports in Wukro woreda 5.5% by Adedy Gidey et al. (2012), the current finding was also much lower than the report in and around holeta 42% by Gizachew et al. (2013), Jordan 64.3% by Al-Ghzawi et al. (2009), South Africa 92% by Strauss, et al. (2013). The variation of the prevalence due to ecological and climatic difference, types of hive and improper hive management, seasonal difference and the high density of honey bee colonies in one apiary are the major challenges of honey bee. In Ethiopia, as one of the subtropical countries the land is suitable to honeybee and different types of pest and predators that are interacting with the honeybee (Keralem et al. 2009). Based on the respondents complain the wax moth, honey badger, birds ant, spider, small hive beetles and others has higher prevalence rates (16-46%) and they were the most harmful pests and predators. They are also decreasing honey production, causes of swarming, causes of absconding and damaging of different types of hive. Some pests simply use the hive for living and shelter their own nest but some pests caused harmful by feeding on pollen, honey and beeswax (Tessega, 2009) in Burie district north west Ethiopia.

According to the survey, most honey bee hives were damaged by pests and parasitic disease but the severity of pests and parasitic disease varied from hive to hive and apiary to apiary. The traditional hive where found heavily attacked by pests because of the materials and difficult to control the pests and predators due to limited accessibility to the hives, different types of size and shapes. The modern hives were less damaged by pests and parasitic honeybee disease due to the beekeeper can easily inspect the hives and control the pests (Kajobe et al., 2009) reported from Uganda. The occurrence of honey bee pests were 46% for wax moth, 28% for ant, 35% for birds, 19% for honey badger and 26% for small hive beetles. From the total respondents 42% had observed agrochemical poisoning and attack their colonies using herbicide for cereals and crops. This reported by Tessega (2009) that performed many beekeepers lost their honey bee by every year because the farmer used agrochemicals for crop and cereals.

Most honeybee keepers used traditional protection method to control different types of honeybee pest and predators in the study area such as clean the hive, removal of the old comb, place fresh ash, hot water for ants and application of dirty engine oil. These control method for honeybee pests and predators were practice in different regions of Ethiopia reported by Gidey et al. (2011) in northern parts of Ethiopia.

CONCLUSION AND RECOMMENDATIONS

Alarmingly increasing honeybee colony losses have been reported from researches over the last few years. From recent survey of honeybee losses in Ethiopia it became evident that pests and parasitic diseases could be identified as the most important cause of these colony losses. Several colony losses honeybee diseases which are thought to be involved in such honey bee colony losses. Therefore, in Ethiopia diverse pathogens are involved in the presumed inexplicable colony losses. Although the decline in managed honeybee equally seems to be problems in North Gondar despite great differences in beekeeping practices, the factors responsible for colony losses differ from region to region and from site to site. It should be prepared that it does not find a globally valid solution to honeybee decline but that will have a panel of possible factors, all of them asking for a specific solution to address the problem. If we are to explain unusual colony losses and if we are to find the cause for these losses, then we need to move from the mere detection of bee parasitic diseases in individuals and colonies to molecular bee disease focusing on host and vector interaction of parasitic disease with equal emphasis on the disease or vector and host. To understand the parasitic disease of honeybees this in turn will enable us to develop adequate control measures. In order to reduce a disease to the best advantage, it is clear that its cause must be known as well as the means by which the infection is transmitted and the environmental condition which are favorable for the breaking out of the disease.

Based on the above conclusion the following recommendation forwarded:

- Further study should be under taken on parasitic honeybee disease with more diagnostic tests to generate more reliable data.

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- The study survey should be carried out during different season of the year to know adequate data on the distribution of different parasitic honeybee disease in different seasons.
- Special attention should be given for bee louse and varroa mite infestation in honeybee
- Any apiarists and farmers should be intensively encountered to change the traditional bee hive into modern type and give advice to manage properly by cleaning their apiary and bee hives.
- Government, researchers and beekeepers should be focused on the effects of agrochemical application on honeybees and to minimize their effects as well as on the development of non-chemical methods of herbicide, insecticides and pesticides control measures.

DECLARATION

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Authors' contributions

MB conceived the study, coordinated the overall activity, and carried out the statistical analysis, drafted the manuscript. TD, AK, TM, BB participated in drafting and reviewing the manuscript. MY conceived the study, coordinated the overall activity, and reviewed the manuscript. MC participated in drafting and reviewing the manuscript. TN participated in the design of the study, and reviewed the manuscript. All authors read and approved the final manuscript.

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Availability of data and materials

Data will be made available up on request of the primary author

Consent to publish

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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REFERENCES

Adedy G, Shiferaw M and Abebe (2012): Prevalence of Bee Lice Braula coeca (Diptera: Braulidae) and Other Perceived Constraints to Honey Bee Production in Wukro Woreda, Tigray Region, Ethiopia. Global Veterinarian, 8 (6): Pp 631-635.

Akinwande L, Badejo A and Ogbogu S (2013): Challenges associated with the honey bee (Apis mellifera adansonii) colonies establishment in south western Nigeria. African

Akinwande L, Badejo A and Ogbogu S (2012): Incidence of the Korea haplotype of Varroa destructor in southwest Nigeria. Journal of Apicultural Research, 15: Pp 369– 370.

Alattal R, Claus P and Zebitz (2006): Infestation levels of Varroa destructor in local honey bees of Jordan.15: Pp 321-326

Al-Ghzawi A, Zaitoun T and Shannag K (2009): Incidence and Geographical Distribution of Honeybee (Apis mellifera) Pests in Jordan. Ann. soc. entomol. Fr., 45 (3): Pp 305-308.

Amsalu B and Desalegn B (2005): Distribution of honeybee diseases Nosema APIs and Melpighamoeba mellificae in Ethiopia. 4th Proceedings of Ethiopian Beekeepers Association, Addis Ababa, Ethiopia, Pp 19-26

Amsalu B and Desalegn B (2001): "Survey of honeybee pest and Pathogen in South and Southwest parts of Ethiopia". Pp. 86-93,

Amsalu B, Alemayeh G, Gemechis L and Kibebew W (2010): "Diagnostic survey of honeybee diseases and pests in Ethiopia.

Amssalu B and Desalegn B (2006): Occurrence of small hive beetle (Aethina tumida) in honeybee (Apis mellifera) Ethiopian Veterinary Journal. 10: Pp. 101-110.

Amssalu B and Desalegn B (2008): Study on the ecological distribution of small hive beetles in maize-coffee growing areas.

Amssalu B (2012): Prevalence and Effects of Nosemosis on Central highland honeybees (Apis mellifera bandasii).

Aster Y, Amssalu B, Betre Y, Desalegn B, Yosef S, Yosef K and Nohe K (2010): Ecological distribution of honeybee Chalk brood disease (Ascosphaera Apis) in Ethiopia. Ethiopian Journal of Animal Production 9(1): Pp 177-191

Aydin L, Gulegen E, Cakmaki I, Girisgin G, and Harrington W (2006): Relation between Nosema and Chalk brood disease and its implication for an apiary management model. Bull Vet Inst Pulawy, 50: 471–475

Bradbear J (2009): Bees and their roles in forest livelihoods: A guide to the services provided by bees and the sustainable harvesting, processing and marketing of their products. FAO Non-Wood Forest Products 19, FAO, Rome.

Coffey M (2007): Parasites of the honeybee. The Department of Agriculture, Fisheries and Food, Crops Research Centre, Oak Park, Carlow, Ireland

Conte Yand Navajos M (2008): Climate change: impact on honey bee populations and diseases. Rev. sci. tech. Off. int. Epiz., 27 (2): Pp 499-510.

Cramp D (2008): A practical manual of bee keeping. United Kingdom. Oxford. Available at http://files.uniteddiversity.com/beekeeping/A practical manual of beekeeping.pdf

Desalegn B (2014): Occurrences and distributions of varroa mite (Varroa destructor) in Tigray regional state, Ethiopia. Journal of Fisheries and Livestock production, 2(3):1-4.

Desalegn B. (2007): Assessment of the effect of ant (Dorylus fulvus) on honeybee colony (A. mellifera) and their products in West & South-West Shoa Zones, Ethiopia. Ethiopian Journal of Animal Production; 7(1): Pp 12-26.

Ellis D (2004): The ecology and control of small hive beetles (Aethina tumida Murray) Rhodes University, Graham's town, South Africa, 385 pp.

Ellis D (2008): Bee louse, bee fly, Braulid, Braula coeca Nitzsch (Diptera: Braulidae). In J L Capinera, Encyclopaedia of Entomology. Springer; Berlin, Germany, Pp 417-419

Ellis D and Munn A (2005): The worldwide health status of honey bees. Bee World, 86: Pp 88-101.

Ellis D, Graham R, and Mortensen A (2013): Standard methods for Wax moth research. In: V Dietemann; J D Ellis; P Neumann (Eds).The COLOSS BEE BOOK.Volume II: standard methods for Apis mellifera pest and pathogen Research: Journal of Apicultural Research, 52(1). Pp 1-17.

Ellis D.and Macedo A (2001): Using the Sugar Roll Technique to Detect Varroa Mites in Honey Bee Colonies. Lincoln, NE: University of Nebraska. 4 Pp

Etsay K and Ayalew K (2001): "Survey on honeybee diseases and pests in Tigray". Bureau of Agriculture and National Resources (BOANR), Mekelle, Ethiopia.

Fakhimzadeh K (2001): Detection of major mite pests of Apis mellifera and development of non-chemical control of Varroasis. Department of Applied Biology, 46 Pp

FAO (2006): Honey bee diseases and pests: A practical guide. Food and Agriculture Organization of the United Nations (FAO agricultural and food engineering technical report 4), Rome, Italy, 33 pp.

FAO (2012): Environment and Natural Resource Management: Adaptation to Climate Change in Semi- Arid Environments Experience and Lessons from Mozambique. FAO, Rome, Italy. 71P.

Fischer D and Moriarty T (2011): Pesticide risk assessment for pollinators: Summary of a SETAC Pellston Workshop. Pensacola FL (USA): Society of Environmental Toxicology and Chemistry (SETAC) and Global Pesticide and Agrochemical Market to 2018.

Fraizer M, State P and Caron D (2010): honey bee diseases and their maladies. The mid atlantic apiculture research and extension: Pp 61-75

Fries I and Compazine S (2001): Implications of horizontal and vertical pathogen transmission for honey bee epidemiology. Apidologie, 32: 199–214.

Genersch E (2010): Honey bee pathology: current threats to honey bees and beekeeping. Applid.Microbiology and Biotechnology, 87: 87-97.

Gezahegne T (2001): Beekeeping, Mega Printer Enterprise, and Addis Ababa, Ethiopia

Gizachew G, Sefinew A, Amssalu B and Malede B (2013): "Prevalence and Associated Risk Factors of Bee Lice in Holeta and its Surroundings, Ethiopia. Journal of Veterinary Science Technology: 4, 130 Pp

Haylegebriel T (2014): Honey Bee Diseases, Pest and Their Economic Importance in Ethiopia. International Journal of Innovation and Scientific Research, 10(2): Pp 527-535.

Hendrikx P, Debin M, and Chauzat P (2010): Bee mortality and bee surveillance in Europe. Pp 1-278.

Hood M (2004). The small hive beetle, Aethina tumida: a review, Bee World, 85(3): 51–59. Janine K, Martin D, Jose-Anne L, Coralie M and Noa Simon-D (2012). Assessment of pesticides risk for bees: methods for PNEC measurements. 11th International Symposium of the ICP-BR Bee Protection Group, Wageningen (The Netherlands), November 2-4, 2011

Johnson M, Ellis D, Mullin A and Frazier H (2010). Pesticides and honey bee toxicity – U.S.A., Apidologie, 41: 312–331. Journal of Food, Agriculture, Nutrition and development, 13(2): Pp 1-18.

Kajobe R, Agea G, Kugonza R, Alioni V, Otim S, Rureba T and Marris G (2009). National beekeeping calendar, honeybee pest, and disease control methods for improved production of honey & other hive products in Uganda. A research report submitted to Natural Agricultural Research Organization (NARO), Entebbe Uganda.

Citation: Nega T, Yayeh M, Mitiku T, Birhan M, Bogale B, Chanie M, and Kinubeh A (2019). Prevalence of parasitic honeybee diseases, pests and predators in North Gondar zone. *Online J. Anim. Feed Res*., 9(5): 198-205. www.ojafr.ir

Kerealem E, Tilahun G and Preston R (2009). Constraints and prospects for apiculture research and development in Amhara region, Ethiopia. Livestock Resource and Rural Development, 21(10): Pp 1-14.

Keshlaf M (2017). The past and present status of beekeeping in Libya. Journal of Apicultural Research, pp. 190-195.

Maarec (2004). Beekeeping basics. delawar, Maryland ,new jersey, Pennsylvania, west Virginia, and USDA cooperating, Pp 46-47.

Marterre B (2008). Certified naturally grown apiary certification standards. Eastern apicultural society master beekeeper, North Carolina state beekeeper association master beekeeper, past president Forsyth county beekeepers association, past vice president and membership secretary.Pp.46-47.

Mustafa M and Williams R (2002): "New record of the small hive beetle in Egypt and notes on its distribution and control," Bee World. 83, Pp. 99–108.

OIE (2008): Nosemosis of honeybees. In: Manual for diagnostic tests and vaccines for terrestrial animals. Paris, Pp. 390-392.

Razmarajii N and Karimi H (2010): A survey of Nosema of honeybees (Apis mellifera) in East Azarbaijan province of Iran. Journal of Animal and Veterinary advances, 9(5):879-882.

Shimanuki H and knox A (2000): diagnosis of honey bee disease. U.S. department of agriculture, agriculture hand book. Pp. 16-35.

SNV (2009): Integrated Value Chain Analyses for Honey and Beeswax Production in Ethiopia and Prospects for Exports. The Netherlands Development Organization.

Sokh R, Molaska M and Siuda M (2007): The influence of the invasion of Nosema Apis on the number of pollen seeds in bee's intestine. Polish Journal of Natural Science, 22:150-156.

Strauss U, Human H, Gauthier L, Crewe M, Dietemann V and Pirk W (2013): Seasonal prevalence of pathogens and parasites in the savannah honeybee (Apis mellifera scutellata). Journal of invertebrate Pathology, 114(1): Pp 45-52.

Tessega B (2009): Honeybee Production and Marketing Systems, Constraints and Opportunities in Burie District of Amhara Region, Ethiopia. Pp116.

Tumlinson Y, Evans D and Suresh R (2010): A scientific note on Varroa destructor found in East Africa; threat or opportunity? Apidologie, 41: Pp 463–465.

Workneh A and Puskur R (2011): Beekeeping Sub Sector Challenges and Constraints in Atsbi Wemberta District of Eastern Zone, Tigray Region, Ethiopia. Journal of Agriculture Extension and Rural development, 3(1): Pp 8-12.

Citation: Nega T, Yayeh M, Mitiku T, Birhan M, Bogale B, Chanie M, and Kinubeh A (2019). Prevalence of parasitic honeybee diseases, pests and predators in North Gondar zone. *Online J. Anim. Feed Res*., 9(5): 198-205. www.ojafr.ir

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THE EFFECTS OF DIFFERENT LITTER MATERIAL ON BROILER PERFORMANCE AND FEET HEALTH

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Supporting Information

RESEARCH ARTICLE

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ABSTRACT: The study was implemented at the National University of Lesotho with the aim to find a potential litter material for use in broiler production. The study followed a completely randomized design with four litter treatments replicated three times. A well ventilated house divided into 12 pans was used where each pan or a replicate contain 15 birds with a total of (n=180) birds. Feeding and watering were done on *ad libitum* basis while the normal routine for broiler production was followed. Litter treatments were made up of control represented by wood shaving and it was compared to dried pine leaves, decomposed kraal manure and sand. All litter materials were applied at the depth of 10 cm. Data was collected on the following parameters broiler production, feet health and chemical and physical properties. Litter material treatment had no significant influence on feed intake, body weight, body weight gain, and feed conversion ratio and mortality rate. Regarding broiler feet health litter treatment had significant influence on foot pad dermatitis while hock burns and broiler temperature were not statistically different amongst litter treatments. Litter evaluation results revealed that different litter sources were differing significantly in terms ammonia emissions, water holding capacity. pH and bulk density while litter temperature did not differ significantly between litter treatments. It was concluded that both dried pine leave and decomposed kraal manure are potential sources that could replace wood shavings in broiler production. Dried pine leaves ideal for improved production while decomposed kraal manure deemed fit for ensure good feet health. It is therefore recommended that farmers in Lesotho can use both decomposed kraal manure and dried pine leaves to replace wood shavings.

Keywords: Feet health, Kraal manure, Lesotho, Litter material, Wood shaving.

INTRODUCTION

Litter quality is of great importance to the welfare of broiler chicken, as they generally spend their entire life in contact with litter (Lonkar, et al., 2018 and Kryeziu et al., 2018). Litter serves several functions that include thermal insulation, moisture absorption, protective barrier from the ground, and it also allows for natural scratching behavior. The quality of the in-house environment is highly dependent upon litter quality (Rizt et al., 2017). An effective bedding material must be readily available, absorbent, lightweight, inexpensive and non-toxic (Rizt et al., 2009; Waziri, Kaltungo, 2017 and Munir et al., 2019). The litter environment is ideal for bacterial proliferation and ammonia production. The two factors that influence litter conditions most are manure and moisture. The ideal litter material should have a moisture content of 20-25%, a pH of 8-10, and ammonia content should not exceed 25 ppm (Dunlop et al., 2016 and Gençoğlan and Gençoğlan, 2017). Excess moisture in the litter increases the incidence of breast blisters, skin bums, scabby areas, bruising, condemnations and downgrades (Hossain et al., 2018). Wet litter is the primary cause of ammonia emissions, one of the most serious performance and environmental factors affecting broiler production today (Rizt et al., 2017). Controlling litter moisture is the most important step in avoiding ammonia problems.

Pine shavings and saw dust have been the most preferred litter source for broilers in Lesotho but because of the high demands, their prices and availability is now a big problem for farmers. In turn farmers are forced to use alternative bedding materials such as dry grass, undecomposed kraal manure and shredded paper characterized by low moisture holding capacity and this results in fungal growth which causes many diseases, increased mortality and welfare concerns in the form of foot pad dermatitis (FPD). Contact dermatitis affects skin surfaces that have prolonged contact with wet litter or other wet flooring surfaces. The condition is manifested as blackened skin progressing to erosions and fibrosis on the lower surface of the footpad, at the back of the hocks, and sometimes in the breast area (World Organization for Animal Health, 2018). If severe, the foot and hock lesions may contribute to lameness and lead to secondary infections (Shepherd et al., 2017). Animal welfare audits in Europe often use foot, hock, and breast burn-lesions as an indicator of housing conditions and the general welfare of the birds (Haslam et al., 2007). Therefore, the aim of this study is to evaluate alternative bedding materials that are inexpensive but had the potential to give comparable results to those of wood shavings under Lesotho condition.

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MATERIALS AND METHODS

Ethical approval

The scientific and ethics committee of the Faculty of Agriculture, National University of Lesotho approved the study protocol.

Study area

The study was carried out at the National University of Lesotho, Faculty of Agriculture farm in Roma some 34 kilometers southeast of Maseru, the capital of Lesotho. The Roma valley is broad and is surrounded by a barrier of rugged mountains which provide magnificent scenery. The university enjoys a temperate climate with four distinct seasons.

Experimental Design

Completely randomized design was used whereby four different sources of litter materials were used as treatments and replicated three times. Wood shavings was considered as the control, while, decomposed kraal manure, sand and dry pine tree leaves were considered as treatments 1, 2 and 3 respectively. Bedding material was laid at the depth of 10 cm. On daily basis, patches of bedding that were too wet as a result of spills from the drinkers were removed from the pans and at end of every week bedding was raked to reduce the caking effect and also to allow air circulation.

Birds and Management

One hundred and eighty (n=180) Ross 308 broiler chickens were reared under deep litter system. Before the arrival of chicks, the house was cleaned thoroughly and disinfected to kill the micro-organisms that might harm the chicks. All the birds were brooded together using a standard bedding material which is wood shavings for the first two weeks and then at the start of growing period, they were randomly distributed into four bedding material treatments, each treatment replicated 3 times with 15 birds per replicate. Birds were provided with continuous light for 24 hours. The three phase feeding was used and the birds were each phase lasted for two weeks. Feeds and water were given *ad libitum*.

Data collection

Data was collected on daily basis on health related parameters and on weekly basis for production performance and physical and chemical assessment of different litter sources.

Production

Feed intake was determined as the difference between total feed supplied and leftovers on weekly basis. Feed conversion ratio was calculated as, feed intake (kg)/lives weight (kg). The body weight was measured using a measuring spring balance scale. Growth rate was measured as the final weight minus initial weight divided by number of days.

Feed intake = total feed supplied- total feed left

Feed conversion ratio (FCR) = feed intake (kg)/live weight gained (kg)

Growth rate= final weight-initial weight/ number of days

Average body weight=total body weight/number of birds

Feet health parameters

Data was collected on the following parameters: mortality rate, signs of illness due to litter material emissions, body temperatures, hock burns and footpad dermatitis. Body temperature was measured using the digital thermometer at the end of each week. The hock burn were scored in six birds per replicate at six weeks of age using a 3 point scoring system (0= no burns; 1 = mild burns and 2= severe burns). The hock burns were assigned to one of three score. The mean score of hock was calculated as the cumulative total of the lesion scores divided by the total number of birds examined. (Thomas et al., 2004) Footpad lesions were assessed by eye and scored according to the protocol of Berg (1998) at the end of every week as follows;

- Class 0 = lesions absent or minor
- Class 1 = medium or mild lesions
- Class 2= severe lesions.

Litter material assessment

A number of laboratory tests were done to assess the quality of litter materials and they included dry matter content, pH, Bulk density, water holding capacity, water releasing capacity and ammonia emitted by litter material. Litter samples were collected at the beginning and at the end of every feeding phase from five locations within each pen and thoroughly mixed. Subsamples were submitted to the Animal Science Laboratory for chemical analysis. Chemical analysis was performed according to Brake et al. (1992) and AOAC International (1995) guidelines. Bulk density was expressed as the weight of 1 litre of as-is litter material. Litter moisture was measured after drying for 24h at 105°C. The pH was recorded using an electronic meter after 30grams of macerated litter were added to 250 milliliter of deionised water, agitated for 5 minutes, and suspended for 30 minutes.

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Water-holding capacity was determined as follows: Litter samples were dried until constant weight and 50grams of litter was placed in a 500 milliliter beaker, the beaker was filled with water and left to stand for 30 minutes, excess water was then drained for 3 minutes and the sample was weighed again; the percentage of water absorbed was calculated on a DM basis.

To determine the water-releasing capacity, each litter sample was placed in a 3-cm-deep pan; the pan was filled with water and allowed to stand for 30 minutes; after draining the excess water for 3 minutes, the litter sample was weighed, the pan was then weighed 5 and 24 hours after draining, moisture loss at each time point was expressed as a percentage of the initial wet weight of the sample.

Determination of NH3 emissions was based on the micro-diffusion method as follows: 100 g of fresh litter was weighed, placed in a 500-millilitre cylindrical flask, and leveled. A 50-millitre beaker containing 10 millilitre of 2% (m/v) boric acid was placed on top of the litter; the flask was closed and incubated for 20 h at 30°C; the boric acid solution was then titrated against sulphuric acid 0.1 *N* with metal orange and bromocresol green; volatilized NH3 (in milligrams per 100 grams of litter) was calculated by multiplying the amount of sulphuric acid used (A) by its normality and the molecular weight of ammonia: NH3 = A × 0.1 × 17.

Data analysis

Data was analyzed using IBM SPSS version 20 (2011) package and data was subjected to Analysis of Variance (ANOVA) to determine the differences among different litter materials.

Statistical model used is as follows; Yij=μ + Bj + Єij

Where;

Yij =ith observation of jth bedding material

μ=Overall mean (mean effect)

Bj=Effect due to bedding material

Єij=Experimental or random error

RESULTS AND DISCUSSIONS

Production parameters

The litter material treatments had no significant (P ≥0.05) influence in all broiler production parameters such as-feed intake, feed conversion ratio, body weight, growth rate and mortality. However, with the comparison to the control, broilers that were reared on dry pine leaves had closest performance in terms of feed intake, feed conversion ratio, body weight, and growth rate. In terms of mortality decomposed kraal manure gave similar results to control.

The results of the current study are in agreement with the findings of Hafeez et al. (2009); Davis et al. (2010); Mendes et al. (2011); Villagra et al. (2011); van Harn et al. (2012); Bjedov et al. (2013); Taherparvar et al. (2016) and Shepherd et al. (2017) who reported that different litter material sources had no effect on feed intake, weight gain, efficiency of feed conversion and growth rate. On the other hand these results are in contrast with the results of Anonymous (1992); Asaniyan et al. (2007); Toghyani et al. (2010), and Lonkar et al. (2018) who reported that the litter material had a significant (P≤0.05) influence on body weight, body weight gain and feed conversion ratio. Based on feed intake and growth rate dried pine leaves were found to be the closest potential litter to wood shavings in terms of production performance.

Table 1 - The effects of litter material on broiler production

Parameters	Litter treatments				Significance	
	Control	T1 (DPL)	T2 (DKM)	T3 (SAND)	P[1]	CV[2]
Feed intake (g/week)	580	581	637	633	0.975	27.8
FCR[3]	1.6	1.7	1.84	1.77	0.946	15.9
Live weight (grams)	2380	2340	2420	2470	0.997	40.9
Mortality	0.000	0.333	0.000	0.333	0.588	27.3
Growth rate (grams)	87	89	81	80	0.401	51.9

[1] Probability level at 0.05 percent; [2] Coefficient of Variation; [3] Feed Conversion Ratio (gram feed/gram weight gain), Control = Wood shavings; T1 = Dried Pine Leaves; T2 = Decomposed kraal manure; T3 = Sand

Feet health

The influence of litter material on broiler feet health results are shown in Table 2. Litter material treatments had a significant influence (P≤0.05) on footpad dermatitis while hock burn and broiler body temperature were not statistically (P≥0.05) different amongst the litter treatments. The highest incidences of footpad dermatitis were found in sand, followed by dry pine tree leaves while decomposed kraal manure had the lowest incidences. Hock burns results on the other hand were highest in pine tree leaves and sand while decomposed kraal manure had no incidences of hockburns. The findings of the current study are in agreement with the findings of Mendes et al. (2013) who compared wood shaving and saw dust litter materials and observed significant differences between two sources whereby wood shavings had

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higher incidences than saw dust. Garcia et al. (2012) also reported that birds raised on litter made of 100% wood shaving had no incidences of footpad dermatitis. Hock burn results are in line with the results of Varol Avcilar et al. (2018) that compared wood shavings and rice hulls and reported that were no significant difference in hock burns between two litter sources.

On the other hand, Bilgili et al. (2009), observed highest incidences of footpad dermatitis in wood shavings and dry pine tree leaves, while mortar sand was ranked as the bedding material with the lowest severity. Lacy et al. (2002) discovered that a problem with dry pine tree leaves is breast blisters and greater incidence of hock burns during the first two weeks. Bilgili et al. (2000) and Chuppava et al. (2018) reported contrasting results and indicated that broilers raised on sand performed as well as or better than those raised on pine shavings and it had the lowest incidences and severity of hock burns and other skin lesions. Body temperatures of birds reared in wood shavings, which was the control, were increasing gradually across the weeks while those reared in sand showed the least body temperatures of all litter materials. These findings are in agreement with the report of Gernat (2009) stating that sand had the lowest temperatures resulting in low body temperatures of the birds but that did not have a negative impact on productivity. Decomposed kraal manure in terms of feet health gave similar performance to wood shavings and could be a potential litter that can be used to improve broiler feet health.

Table 2 - The effects of litter material on birds feet health

Parameters	Litter treatments				Significance	
	Control	T1 (DPL)	T2 (DKM)	T3 (SAND)	P[1]	CV[2]
Incidence of FPD[3] (%)	0.00[a]	16.25[b]	9.85[a]	64.25[c]	0.01	13.73
Body temperature (^{0}C)	40.75	40.93	40.76	40.46	0.46	0.33
Hock burns	2.50	8.25	0.00	4.00	0.12	15.5

[a,b,c] Means within a row with different superscripts differ significantly (P≤ 0.05), [1] Probability level at 0.05 percent; [2] Coefficient of Variation; [3] Foot Pad Dermatitis. Control = Wood shavings; T1 = Dried Pine Leaves; T2 = Decomposed kraal manure; T3 = Sand

Physical and chemical evaluation

The comparison of physical and chemical properties of different litter materials results are shown in table 3. According to these results there were significant differences among different litter treatments in physical and chemical properties with the exception of litter temperature. According to mean comparison test there was no significant difference between control, dry pine leaves and sand in terms of ammonia emission. In terms of water holding capacity, there was no significant difference between the control and dry pine leaves. With regard to litter pH, there was no significant difference between control, dry pine leaves and decomposed kraal manure. Lastly mean comparison test result shows that there was no significant difference between wood shavings and decomposed kraal manure in terms of bulk density.

These results are in agreement with the findings of Brake et al. (1993) and Kuczynski and Slobodzian (2002) who noticed that there were no a significant difference (P≥0.005) in physical and chemical properties between different litter sources while comparing wood shavings with wheat straw, rice straw and sand. Farhadi (2014) used both wood shavings and saw dust as control in the study that seeks to find a potential litter material for broiler and compared them to wheat straw, sugarcane bagasse, sugarcane peat; rice hulls and the researcher found no significant difference between control and the tested litter materials in terms of moisture content, pH, water holding and water releasing capacity and bulk density. Ammonia emission results pointed out the use of kraal manure as litter material should be handled with extra care especially if the substrate is not fully decomposed. Ammonia emissions in this study were higher than the recommended threshold value of 25 ppm for broiler house. The higher values could be associated with poor level of litter decomposition prior to its use as litter material. The higher bulk density also could have contributed significantly because lower bulk density of a material shows high porosity and moisture absorbing capacity, air circulating and moisture releasing capacity than high bulk density (Ataputta and Wickramasinghe, 2007). Litter pH was similar between dried pine leaves and decomposed kraal but they were significantly higher than the control. It was reported that the low pH level of litter material has an advantage because in acidic pH of litter, the conversion of uric acid to ammonia will be reduced (Moore et al., 1996). The lower litter temperature observed under sand could be a contributing factor for higher feed intake for birds under this treatment in an effort to maintain high metabolic activities to keep warm.

Table 3 - Physical and chemical properties of different litter materials

Parameters	Litter treatments				Significance	
	Control	T1 (DPL)	T2 (DKM)	T3 (SAND)	P[1]	CV[2]
Ammonia emissions (mg/L)	6.93[a]	19.50[a]	27.29[b]	17.25[a]	0.001	42.00
Water holding (%)	114.95[a]	105.81[a]	92.20[b]	56.94[c]	0.001	25.00
Litter temperature	17.35	17.96	17.84	16.90	0.681	7.36
pH	6.18[a]	6.73[b]	6.75[b]	7.36[c]	0.001	10.00
Bulk density (g/cm^3)	300.14[a]	205.06[b]	370.50[a]	1788.75[c]	0.001	101.00

[a,b,c] Means within a row with different superscripts differ significantly (P≤ 0.05), [1] Probability level at 0.05 percent; [2] Coefficient of Variation; [3] Foot Pad Dermatitis. Control = Wood shavings; T1 = Dried Pine Leaves; T2 = Decomposed kraal manure; T3 = Sand

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CONCLUSION AND REECOMMENDATION

In accordance with the findings of the current study, separated into production, feet health and chemical and physical assessment, it can be concluded that both dried pine leaves and decomposed kraal manure can be used as broiler litter material. Kraal manure had been found to be a good source that can support proper feed health with the lowest incidence of feet pad dermatitis and hock burns. Dried pine tree leaves proved to be a good source that can maintain production in the similar manner to wood shavings. The use of kraal manure needs full decomposition in order to reduce the incidences of ammonia emissions. The two litter sources are therefore recommended for farmers who want to use them because they are readily available in Lesotho.

DECLARATIONS

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Author's contribution

N.Kuleile participated in the design of the study. Metsing and M.Phororo collected the litter materials, constructed the pens and tested the litters on broilers. I.Metsing, M.Phororo, T.Jobo, and C.Tjala performed the experiments and analyzed the data. N.Kuleile critically revised the manuscript for important intellectual contents and wrote the manuscript. All authors read and approved the final manuscript.

Competing interests

The authors declare that they have no competing interests.

REFERENCES

Anonymous (1990). General note on stock management (litter management). Poultry Management Guide, pp. 4-5.

Asaniyan K, Agbede O and Laseinde O (2007).Impact assessment of different litter depths on the performance of broiler chickens raised on sand and wood shaving litters. WJZ, 2(2): 67-72. Google Scholar

Brake D, Fuller J, Boyle R, Link E, Peebles D and Latour A (1993). Evaluations of whole chopped kenaf and kenaf core used as a broiler litter material. Poultry Science. 72(11): 2079–2083. Google Scholar

Bjedov S, Žikić D, Perić L, Đukić Stojčić M and Milošević N (2013). Effect of different litter treatments on production performance of broiler chickens. Biotechnology in Animal Husbandry 29(4): 625-630. Google Scholar I http://doi.org/10.2298/BAH1304625BM

Bilgili S, Hess J, Blake J, Eckman M (2000). Turning trash into treasure: Sand as bedding material for rearing broilers. Highlights of Agricultural Research. 47 (1). http://www.ag.auburnedu/aaes/information/highlights/spring00/sand.html I Google Scholar

Bilgili F, Montenegro I, Hess B and Eckman K (2009). Live performance, carcass quality, and deboning yields of broilers reared on sand as a litter source. Journal of Applied Poultry Research, 8(3): 352–361. Google Scholar

Chuppava B, Visscher C and Kamphues J (2018). Effect of different flooring designs on the performance and foot pad health in broilers and turkeys. Animals Basel, 8(5):70. http://doi.org/10.3390/ani8050070

Davis D, Purswell L, Columbus P and Kiess S (2010). Evaluation of chopped switch grass as a litter material. International Journal of Poultry Science, 9(1): 39-42. Google Scholar

Dunlop W, McAuley J, Blackall J and Stuetz M (2016). Water activity of poultry litter: Relationship to moisture content during a grow-out. Journal of Environmental Management, 172: 210-201. Google Scholar I http://dx.doi.org/10.1016/j.jenvman.2016.02.036

Toghyani M, Gheisari A, Modaresi M, Tabeidian SA, Toghyani M (2010). Effect of different litter material on performance and behavior of broiler chickens. Applied Animal Behaviour Science, 122(1), 48-52. Google Scholar

García RG, Almeida Paz ICL, Caldara FR, Nääs IA, Pereira DF and Ferreira VMOS (2012). Selecting the most adequate bedding material for broiler production. Brazil Journal of Poultry Science, 14(2): 121-127. Google Scholar

Gernat A (2009). Use of sand as litter for broilers. https://www.wattagnet.com/articles/3911-use-of-sand-as-asbroiler.litter

Gençoğlan S and Gençoğlan C (2017). The effect of the litter materials on broiler chickens welfare and performance. Turkish Journal of Agriculture, Food Science and technology. 5(12): 1660-1667. https://doi.org/10.24925/turjaf.v5i12.1660-1667.176

Hafeez A, Suhail M, Durrani R, Jan D, Ahmad I and Rehman A (2009). Effect of different types of locally available litter materials on the performance of broiler chicks. Sarhad Journal Agriculture. 25(4): 581-586

Haslam S, Knowles G,Brown N, Wilkins L and Kestin C (2007). Factors affecting the prevalence of foot pad dermatitis, hock burn and breast burn in broiler chicken. British Poultry Science, 48 (03):264-275. http://dx.doi.org/10.1080/00071660701371341

Hossain A, Zulkifili I, Islam S and Awad A (2018). Effect of wood shaving litter density on the growth, leg disorders and manurial value in broiler. Bangladesh Journal of Animal Science. 47(1): 21-27. Google Scholar

Kuczynski T and Slobodzian O (2002). Physical properties of different types of litter and their effect on animal health and welfare in turkey housing. Annuals of Animal Science, 1: 31-33. Google Scholar I Link

Kryeziu J, Mestani N, Berisha H and Kamberi A (2018). The European performance indicators of broiler chickens as influenced by stocking density and sex. Agronomy Research 16(2): 483-491. https://doi.org/10.15159/AR.18.040

Lacy P (2002). Litter quality and broiler performance. The University of Georgia College of Agricultural & Environmental Sciences. Cooperative Extension Service. http://www.ces.uga.edu/pubcd/L426-w.html

Lonkar D,Ranade S, Kulkarni R, Pathak B, Yenge D and Daware G (2018). Effect of organic acid treated corn cob bedding material on broiler performance, hock burn incidence and litter quality. International Journal of Science, Environment and Technology, 7(2): 397–409. ISSN: 2278-3687. Google Scholar

Mendes S, Paixão J, Restelatto R, Reffatti R, Possenti C, Moura DJ, Morello Z and Carvalho R (2011). Effects of initial body weight and litter material on broiler production. Brazilian Journal of Poultry Science 13(3):165-170. Google Scholar I https://doi.org/10.1590/S1516-635X2011000300001

Munir MT, Belloncle C, Irle M and Federighi M (2019). Wood-based litter in poultry production: a review. Worlds Poultry Science Journal, https://doi.org/10.1017/S0043933918000909

Musilova A, Lichovnikova M and Przywarova A (2014). The effect of the type of litter on the occurrence of footpad dermatitis in broiler chickens. Mendel Net. pp 167-171. Google Scholar

Ritz CW, Fairchild BD and Lacy MP (2017). Litter Quality and Broiler Performance. University of Georgia, College of Agricultural and Environmental Sciences. Extension Bulletin. No 1267. Google Scholar

Ritz CW, Fairchild BD, Lacy MP (2009). Litter quality and broiler performance. University of Georgia. College of Agricultural and Environmental Sciences. Link

Shepherd EM, Fairchild BD, and Ritz CW (2017). Alternative bedding materials and litter depth impact litter moisture and footpad dermatitis. Journal of Applied Poultry Research. 26: 518–528. Google Scholar I http://dx.doi.org/10.3382/japr/pfx024

Taherparvar G, Seidavi A, Asadpour L, Rita Payan-Carreira R,Laudadio V and Tufarelli V (2016). Effect of litter treatment on growth performance, intestinal development, and selected cecum microbiota in broiler chickens. Revista Brasileira de Zootecnia, 45(5): 257-264. Google Scholar

Van Harn J, Aarnink J, Mosquera J, van Riel W and Ogink M (2012). Effect of bedding material on dust and ammonia emission from broiler houses. American Society of Agricultural and Biological Engineers, 55(1):219-226. http://dx.doi.org/10.13031/2013.41249

Varol Avcilar Ö, Kocakaya A, Onbasilar E and Pirpanahi M (2018). Influence of sepiolite additions to different litter materials on performance and some welfare parameters of broilers and litter characteristics. Poultry Science Journal, 97(9):3085-3091. http://dx.doi.org/10.3382/ps/pey185

Villagra A, Olivas I, Benitez V and Lainez M (2011): Evaluation of sludge from paper recycling as bedding material for broilers. Poultry Science, 90(5): 953–957. Google Scholar I https://doi.org/10.3382/ps.2010-00935

Waziri I and Kaltungo Y (2017). Poultry litter selection, management and utilization inthe tropics. In Poultry Science web book. Chapter 8: pp 192-208. Google Scholar I http://dx.doi.org/10.5772/65036

World Organisation for Animal Health (2018). Animal welfare and broiler chicken production, chapter 7.10. In; Terrestrial Animal Health code-10/08/2018. Pp. 2.

Citation: Kuleile N, Metsing I, Tjala C, Jobo T and Phororo M (2019). The effects of different litter material on broiler performance and feet health. *Online J. Anim. Feed Res.*, 9(5): 206-211. DOI: https://dx.doi.org/10.36380/scil.2019.ojafr29

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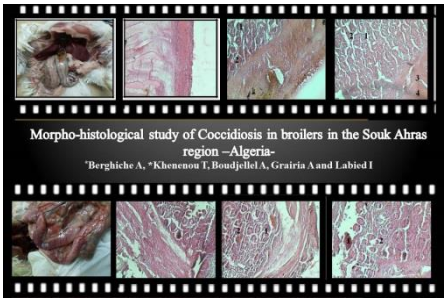

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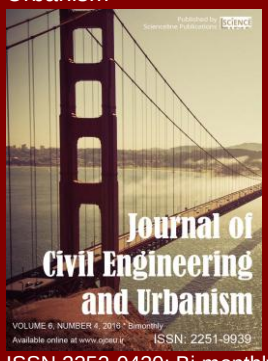

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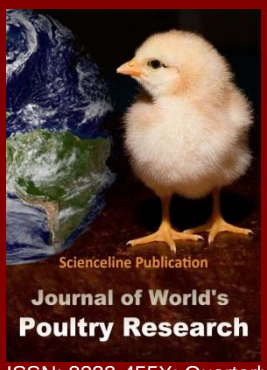

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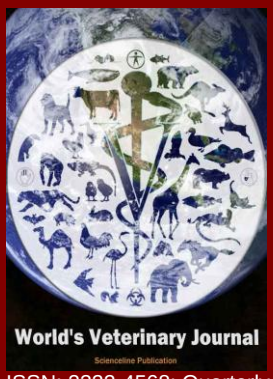

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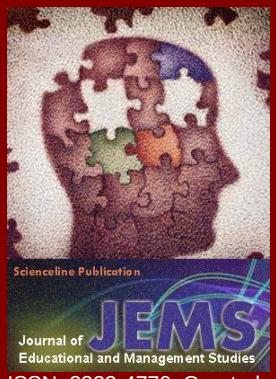

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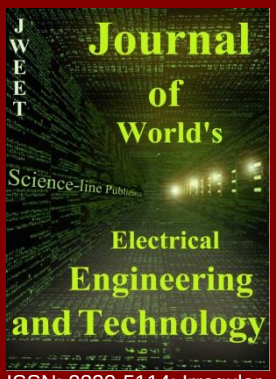

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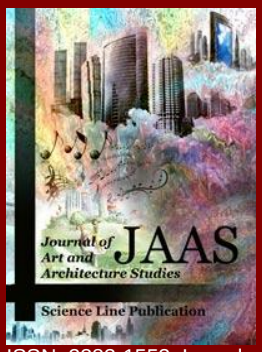

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