

Growth performance and organ histological changes of WAD goats fed varying levels of *Moringa oleifera* leaf meal-based diets

¹Sanusi, G.O., ²Oderinwale, O. A , ²Tovies, D. P. ²Somoye, O.O. ²Okole, T. A. ³Olalere, J.O. ⁴Oluwatosin, B O.

¹Department of Animal Health and Production Technology, School of Agricultural Technology and Engineering, Edo State College of Agriculture and Natural Resources, Iguoriakhi, Edo State, Nigeria.

²Department of Animal Production and Health, Federal University of Agriculture, Abeokuta, Nigeria.

³Livestock Science and Sustainable Environment Programme, World Bank Africa Centre of Excellence in Agricultural Development and Sustainable Environment, Federal University of Agriculture, Abeokuta.

⁴Institute of Food Security, Environmental Resources and Agricultural Research, Federal University of Agriculture, Abeokuta, Ogun State.



Corresponding author: onesantayo@gmail.com, +2348032655408; +2348032655408

Abstract

Plant protein is an important component of ruminants' diets. *Moringa oleifera* leaf meal is a good and cheap source of protein which could improve the nutrition and performance of goat. An 84-day feeding trial was carried out to determine the growth performance and histology of internal organs of West African Dwarf (WAD) bucks fed air-dried *Moringa oleifera* leaf meal (MOLM) -based diets. Twenty 6-8 month old WAD bucks with an average weight of 7.66 kg were assigned to treatments containing 0%, 5%, 10% and 15% of MOLM. Daily feed intake, weight gain, histological examinations of the kidney, liver and small intestines were measured in a Completely Randomised Design. Dry matter intake ranged 382 – 391 g/day. The average feed intake per day ranged between 0.44 and 0.64 kg. Goats fed MOLM at 10% and 15% recorded the highest final body weight of 9.32 kg and 9.10 kg and body weight gain of 1.76 kg and 1.76kg, respectively. The results on the histological examination of the kidneys, livers, and small intestines of the bucks fed 0% showed no visible lesion at Haematoxylin and Eosin Bar=100µm. However, the photomicrographs of bucks in 5%MOLM showed lymphocytic infiltration of the lamina propria of the small intestines, mild diffuse centrilobular areas of necrosis of the hepatocytes and tubular degeneration in the kidneys. There was tubular epithelial regeneration of the kidneys and all cells were normochromic. *Moringa oleifera* leaf meal 15 % posed little or no threat to WAD bucks.

Keywords: Haematoxylin, lesion, necrosis, Photomicrograph, weight gain.

Running title: performance of WAD goats fed varying levels of *Moringa oleifera* leaf meal-based diets

Performance de croissance et modifications histologiques des organes de chèvres WAD nourries avec des régimes à base de farine de feuilles de *Moringa oleifera* à différents niveaux

Résumé



Les protéines végétales sont un composant important de l'alimentation des ruminants. La farine de feuilles de *Moringa oleifera* est une source de protéines de bonne qualité et économique qui pourrait améliorer la nutrition et les performances des chèvres. Un essai d'alimentation de 84 jours a été réalisé pour déterminer la performance de croissance et l'histologie des organes internes de boucs nains d'Afrique de l'Ouest (WAD) nourris avec des régimes à base de farine de feuilles de *Moringa oleifera* séchées à l'air (MOLM). Vingt boucs WAD âgés de 6 à 8 mois et pesant en moyenne 7,66 kg ont été répartis selon des traitements contenant 0%, 5%, 10% et 15% de MOLM. La consommation alimentaire quotidienne, le gain de poids et les examens histologiques des reins, du foie et de l'intestin grêle ont été mesurés selon un dispositif complètement aléatoire. L'ingestion de matière sèche a varié de 382 à 391

g/jour. La consommation alimentaire moyenne par jour a varié entre 0,44 et 0,64 kg. Les chèvres nourries avec 10 % et 15 % de MOLM ont enregistré le poids corporel final le plus élevé, respectivement 9,32 kg et 9,10 kg, et un gain de poids corporel de 1,76 kg dans les deux cas. Les résultats de l'examen histologique des reins, du foie et de l'intestin grêle des boucs du groupe 0% n'ont montré aucune lésion visible à l'hématoxyline et à l'éosine (Barre = 100 µm). Cependant, les photomicrographies des boucs du groupe 5% MOLM ont montré une infiltration lymphocytaire de la lamina propria de l'intestin grêle, des zones nécrotiques centrilobulaires légèrement diffuses dans les hépatocytes et une dégénérescence tubulaire dans les reins. Une régénération de l'épithélium tubulaire des reins a été observée et toutes les cellules étaient normochromiques. La farine de feuilles de Moringa oleifera à 15 % a présenté peu ou pas de menace pour les boucs WAD.

Mots-clés : Hématoxyline, lésion, nécrose, photomicrographie, gain de poids.

Introduction

Moringa oleifera Lam. (Moringaceae) is native to the southern part of the Himalayas in northwest India and the most widely cultivated species of the genus (Osewa *et al.*, 2013). It has become naturalized in many tropical countries of Africa. The common English names are: Moringa, drumstick tree, horseradish tree and benzoil tree. There has been an increasing interest in the use of Moringa as a protein source for livestock feeding (Sarwatt *et al.*, 2004; Asaolu *et al.*, 2010). It is one of the vegetables which has gained acceptance in Nigeria. It is widely grown or cultivated in different parts of Nigeria where it is locally called Zogeli (among the Hausa speaking people) and Èwè igbale in Yoruba and 'Okweoyibo' in Igbo. It is also known as "Miracle tree", (Ijeoma *et al.*, 2012). Fresh Moringa foliage has been included into the diet of different animals. Positive effects on feeding behaviour in goats and growth rate in sheep (BenSalem and Makkar, 2009) have been reported. Studies have shown that Moringa oleifera plays an important role in animal's voluntary intake and performance by providing nutritional, medicinal and therapeutic value as its numerous functions (Kakengi *et al.*, 2007, Al-kharusi *et al.*, 2009; Sanusi, 2017). The crude protein content (circa 25%) in the air dried leaves is high (Reyes-Sánchez *et al.*, 2006). About twenty potentially harmful compounds commonly found in several fodder plants are present in Moringa

such as soluble calcium oxalates (Noonan and Savage, 1999; Radek and Savage, 2008), mimosine, cyanogenic glycosides, moringine, condensed tannins and alkaloids. The use of forage trees and shrubs have been limited and *ad libitum* feeding of these forages is rarely used in livestock feeding. However, the presence of various anti-nutritional compounds and their deleterious effects in animals have also been discussed (Ghosh *et al.*, 2008). These molecules can cause digestive problems, side effects and the formation of kidney stones not only in animals but also in human beings (Finkielstein and Goldfarb, 2006). Although air-drying has been acclaimed to have reduced some antinutritional factors in Moringa oleifera leaf. It will not be conclusive to adjudge this method as the panacea without scaling its effects on the tissues of the leftover phytochemicals. Hence, the aim of this study was to determine the growth performance and organ histology of WAD bucks on certain internal organs of the body fed air-dried Moringa oleifera leaf based diets.

Study area

The study was carried out at the Kalahari Unit of the Institute of Food Security, Environmental Resources and Agricultural Research, Federal University of Agriculture, Abeokuta, Ogun State, which is located in the tropical rainforest zone in Nigeria within 7°13'47.4''N, 3°23'43.4''E. Seasonal distribution of rain is approximately 163 mm (14.6%) in the early dry season and 110 mm (9.97%) in the late dry

season (Jan-March) (October – December) (OORBDA, 2012).

Harvesting and processing of *Moringa oleifera* leaves

Moringa oleifera leaves of Nigerian ecotype were obtained in Odeda Local Government, Ogun state. The harvested *Moringa* leaves were air-dried by spreading on a tarpaulin and cemented floor in a roofed and well ventilated room. The leaves were frequently turned until they were crispy to touch while retaining their greenish colouration. The leaves were then hand-milled to obtain a product herein referred to as *Moringa* leaf meal (MOLM) which was stored in sacs until needed.

Management and feeding of experimental animals

Twenty West African Dwarf bucks of about 6-8 months of age weighing 7.66 ± 0.22 kg were randomly assigned to four treatments of five replicates each that received experimental diets containing 0 (control), 5, 10 and 15% of air-dried *Moringa oleifera* leaf meal on weight basis. The experiment lasted for a duration of 98 days (14 days of adjustment period, 84 days for the feeding trial). Prior to the start of the experiment, goats were dewormed. The animals were managed under intensive system. Five goats were served with 5 kg of the experimental diets at 08:00 hours every morning and water was given *ad libitum*.

Table 1: Composition (% DM) of *Moringa oleifera* leaf meal-based concentrates fed to West African Dwarf bucks

Ingredients	0 %MOLM	5 %MOLM	10 %MOLM	15 %MOLM
MOLM (kg)	0.00	5.00	10.00	15.00
Concentrate (kg)	100.00	95.00	90.00	85.00
Total (kg)	100.00	100.00	100.00	100.00

The concentrate mixture fed to West African Dwarf bucks, expressed on a dry matter (DM) basis, comprised groundnut cake (29 kg), corn bran (65 kg), molasses (2 kg), bone meal (2 kg), salt (1 kg), and premix (1 kg). These ingredient proportions remained constant across all dietary treatments.

Specimen collection and histology

After the feeding trial period, one buck per treatment was slaughtered and parts of the organs such as liver, kidney, heart, small intestine were dehydrated in graded levels of alcohol (about 70% - 80%) in ascending order to remove the water content. After the dehydration, the tissues were cleared in xylene impregnated with paraffin wax and sectioned at 5 microns thickness using rotary microtone. The sections were floated on a paraffin water bath maintained at a temperature of 2-3 °C below melting point of the paraffin wax 56°C

after which the sections were dried on a slide dryer maintained at a temperature of 3°C higher than the melting 58-59 °C point of the paraffin wax used. After proper drying, the sections were stained with haematoxylin and eosin (H and E) and mounted using histomount. Photography of the desired sections was made for further observation (Isaac *et al.*, 2023).

Statistical analysis

All investigated parameters were based on completely randomized design using the procedure of Statistical Analysis Software 9.1 (SAS, 2003) and treatment means were compared using Duncan Multiple Range Test (Duncan, 1955) of the same software. The statistical model focused primarily on inclusion level effect as the main treatment. The following model was used: $Y_{ij} = \mu + M_j + e_{ij}$; where Y_{ij} is the dependent, continuous variable; μ is the overall mean; M_j is the fixed effect of

the j th inclusion of air-dried *Moringa oleifera* leaves ($j = 0\%, 5\%, 10\%, 15\%$) and e_{ij} is the residual error.

Results

The proximate composition of experimental diets fed to West African Dwarf goats was

shown in Table 2.0. Dry matter content was in the range of 83.4% and 88.3% while the highest crude protein was 8.54% in 0% MOLM. The values nitrogen free extract, acid detergent fibre and hemicellulose in 15% MOLM were 46.0, 21.2 and 15.2%, respectively.

Table 2: Proximate composition of experimental diets (% dm) fed to West African Dwarf bucks

Parameter	0%MOLM	5%MOLM	10%MOLM	15%MOLM
Dry Matter	88.35	83.47	84.66	83.35
Crude protein	8.54	8.17	7.25	7.93
Crude fibre	6.44	9.01	7.57	6.80
Ether extract	14.23	8.66	2.51	6.67
Ash	15.51	24.07	18.66	16.44
Nitrogen free extract	43.63	33.56	48.67	46.00
Neutral detergent fibre	49.17	37.31	22.00	36.55
Acid detergent fibre	32.38	11.47	11.59	21.27
Hemicellulose	16.79	25.84	10.41	15.28

Adapted from Oluwatosin et al.,(2017).

Nutrient intake of West African Dwarf Goat fed *Moringa oleifera* leaf meal-based diets is shown in Table 3.0. The lowest dry matter intake of 366.8 g/day was observed in bucks fed 10 % MOLM. The highest crude protein, ether extract, neutral detergent fibre and acid detergent fibre intakes were 32.6 g, 61.5 g, 140 g and 297 g, respectively in bucks fed 0 % MOLM.

Table 3: Nutrient intake of West African Dwarf goat fed Moringa oleifera leaf meal -based diets

Parameters	0% MOLM	5% MOLM	10% MOLM	15% MOLM	SEM
Dry Matter	382.90	382.59	366.88	391.39	15.73
Crude Protein	32.69	31.25	26.59	31.03	1.80
Ether Extract	61.57 ^a	39.69 ^b	10.88 ^c	34.43 ^b	6.87
Ash	59.51 ^c	92.09 ^a	68.46 ^b	71.16 ^b	2.61
Neutral Detergent Fibre	140.41 ^a	52.43 ^c	50.69 ^c	109.76 ^b	14.69
Acid Detergent Fibre	297.29 ^a	231.19 ^b	46.39 ^c	193.41 ^b	35.10

^{a,b,c} means within a row with different superscript are different ($p < 0.05$) ns means not significantly different ($p > 0.05$). Adapted from Oluwatosin et al., (2017).

Table 4 shows Performance of West African dwarf (WAD) bucks fed Concentrate diets at various levels of *Moringa oleifera* leaf meal. Varying levels of MOLM inclusion in the experimental diets significantly affected ($p < 0.05$) total feed intake which was in the range of 36.74 to 53.46 kg. Animals fed 15% MOLM had the highest feed intake of 53.46kg while 5% MOLM had the lowest feed

intake of 36.74kg. Bucks fed 0, 5 10 and 15% MOLM had initial body weight of 7.18, 8.08, 8.04 and 7.34 kg, respectively while total weight gain ($p > 0.05$) were 1.58, 0.92, 1.28 and 1.76 kg respectively. The total weight gain was with bucks fed 15% MOLM which had the highest weight gain of 1.76kg. There was no significant difference ($p > 0.05$) in weight gain among the treatments.

Table 4: Performance of West African Dwarf (WAD) bucks fed Concentrate diets at various levels of *Moringa oleifera* leaf meal

Parameters	0%MOLM	5%MOLM	10%MOLM	15%MOLM	SEM
Total feed intake (kg)	38.70 ^b	36.74 ^b	43.84 ^b	53.46 ^a	1.82
Daily feed intake (kg/day)	0.46 ^b	0.44 ^b	0.52 ^b	0.64 ^a	0.02
Initial weight /goat (kg)	7.18	8.08	8.04	7.34	0.22
Final weight/ goat (kg)	8.76	9.00	9.32	9.10	0.18
Total weight gain/goat (kg)	1.58	0.92	1.28	1.76	0.15
Daily weight gain/ goat (kg/day)	0.02	0.01	0.02	0.02	0.00
Daily metabolic weight gain/goat (kg W ^{0.75} /day)	8.93	9.59	7.59	9.69	0.79

^{a,b,c} means on the same row having different subscripts are significantly different (P<0.05)

The histological examination of the kidney fed control 0% *Moringa oleifera* showed mild diffuse degeneration while 5% MOLM inclusion in diet showed tubular degeneration and necrosis but also showed tubular epithelial regeneration. The kidney of the animals in 10% MOLM and 15% MOLM showed tubular epithelial regeneration. The section of the hepatocyte of 0% MOLM showed no visible lesion while hepatocyte with 5% and 10%

MOLM in diet showed mild diffuse centrilobular areas of necrosis respectively. The hepatocyte of animals in fed 15% *Moringa oleifera* inclusion in diet showed focal areas of necrosis with mononuclear cells infiltration. The small intestine of the 0% *Moringa oleifera* showed no visible lesion while 5%, 10%, and 15% MOLM all showed lymphocytic infiltration of the lamina propria

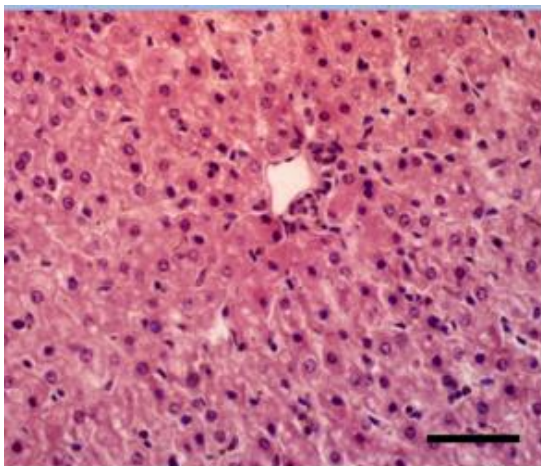


Fig. 1: Photomicrograph of liver section of 0% MOLM showing no visible lesion H andE Bar=100μm

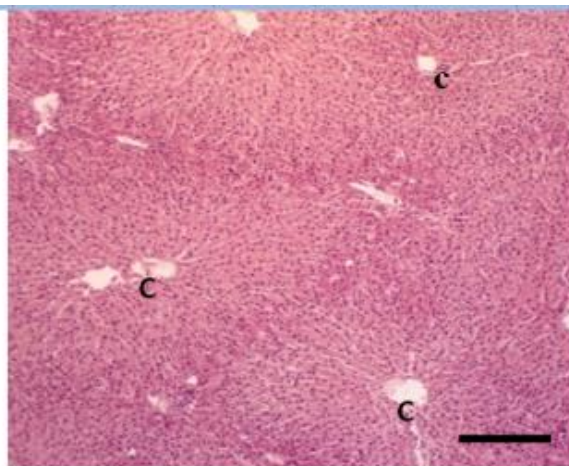


Fig. 2: Photomicrograph of liver section of 5% MOLM showing mild diffuse centrilobular areas of necrosis (C) H andE Bar=200μm

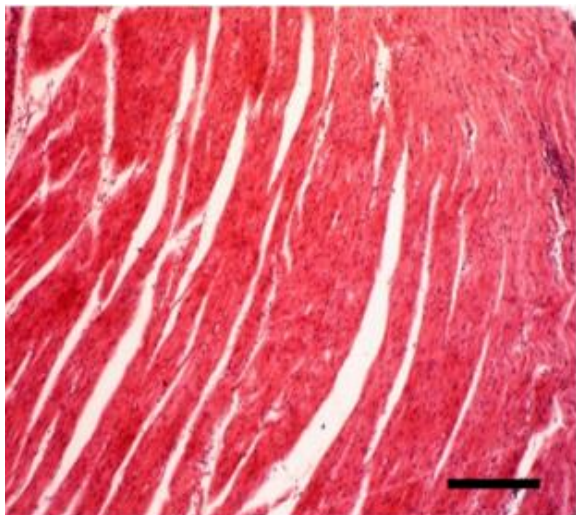


Fig. 5: Photomicrograph of kidney section of 0% MOLM showing no visible lesion H andE Bar=100 μ m

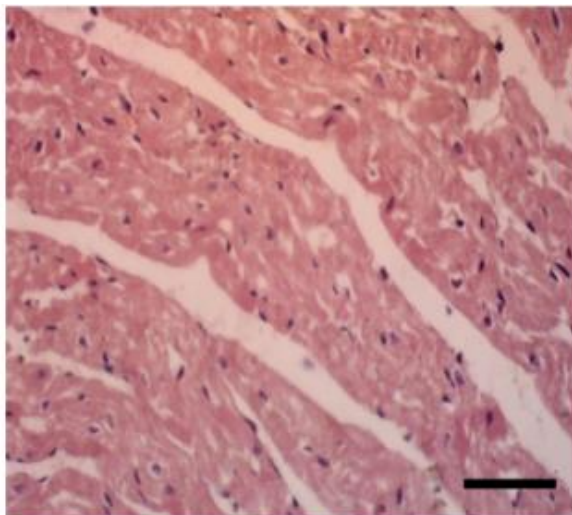


Fig. 6: Photomicrograph of kidney section of 5% MOLM showing no visible lesion H andE Bar=100 μ m

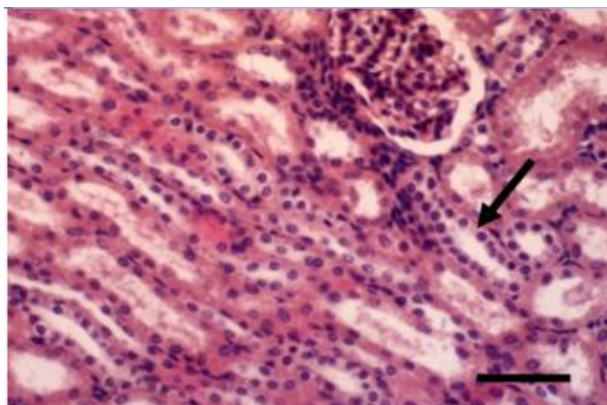


Fig. 7: Photomicrograph of kidney section of 10% MOLM showing tubular epithelial regeneration (arrow)H andE Bar=100 μ m

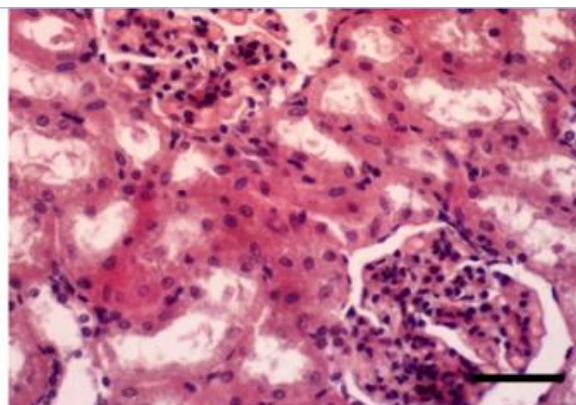


Fig. 8: Photomicrograph of kidney section of 15% MOLM showing tubular epithelial degeneration and necrosis with mild regeneration of epithelial cells H andE Bar=100 μ m

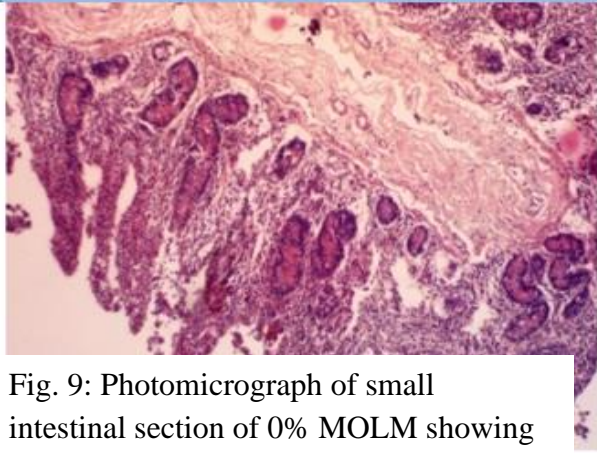


Fig. 9: Photomicrograph of small intestinal section of 0% MOLM showing no visible lesion. H and E Bar=100µm

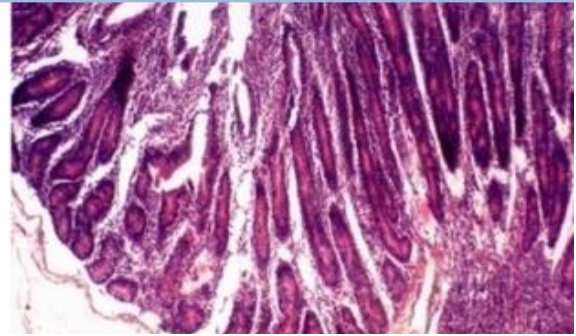


Fig. 10: Photomicrograph of small intestinal section of 5% MOLM showing lymphocytic infiltration of the lamina propia H and E Bar=200µm

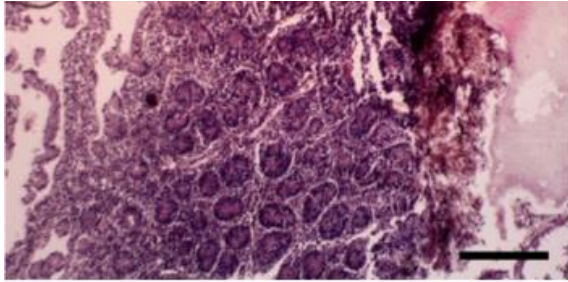


Fig. 11: Photomicrograph of small intestinal section of 10% MOLM showing lymphocytic infiltration of the lamina propia H and E Bar=200µm

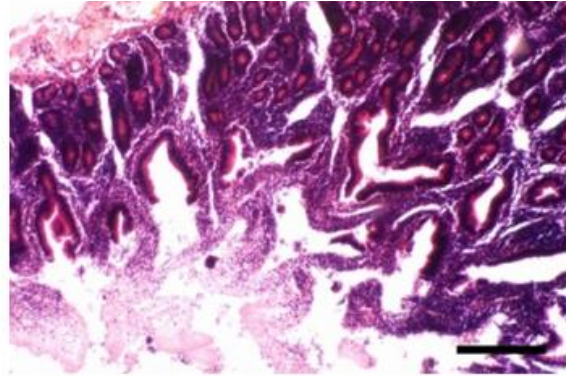


Fig. 12: Photomicrograph of small intestinal section of 15% MOLM showing lymphocytic infiltration of the lamina propia H and E Bar=200µm

Discussion

Inclusion of *Moringa oleifera* leaf meal with other ingredients resulted in a rich blend of good-quality feed. There was little variability between the experimental diets with regards to their chemical composition. The crude protein (CP) content of the MOLM-based diets (5%, 10% and 15 %) compared favourably with that of the control diet (0% MOLM) which was in a range of 7.25 g/Kg to 8.54 g/Kg. This was within the normal requirement of goats (Sanusi, 2017). The CP of the control diet was slightly higher than those of other diets and tended to decrease with increasing levels of MOLM in the diet and stepped up in 15%. The crude protein of *Moringa oleifera* has been reported by Sanusi (2017) to be of better quality for ruminants because of its high content of by-pass protein. Apori (1988) suggested a range of 7% to 12% CP which he believed to be adequate for West African dwarf goats. Protein forms the structure of most body organs and tissues, if less protein is present in the ration, then less quantity of tissues will be deposited as flesh in the body. A low protein diet was found to reduce the capacity for protein synthesis in tissues such as liver and skeletal muscles (Oduguwa, 2000). Neutral detergent fibre and acid detergent fibre fractions of the diets were within the same range. The fat content ranged between 2.51 and 14.2. These rations were low in fat content. Liver damage usually results from a diet high in fat by means of oxidative stress, inflammation, fibrogenesis, and liver necrosis (Nurhayati *et al.*, 2024). It has been suggested that fat content influences the acceptability and palatability of the feed.

Feed intake was described by Masafu (2006) as a measure of diet appreciation, selection and consumption by an animal. It was reported that the supplementation of other feedstuffs with moringa leaves improved dry matter intake (Gebregiorgis *et al.*, 2012). The dry matter intake was similar ($p > 0.05$) among treatments with 15%, 5%, 0%, 10% having 391g, 382g, 382g and 366g/buck/day respectively. These values compared with the range of 291 – 313

g/animal/day which was reported by Ukanwoko *et al.* (2009) for West African Dwarf goats fed cassava peels-cassava leaf meal-based diets. These values fell within dry matter intake recommended levels for small ruminants (NRC, 1985). This superiority stems mainly from the balanced and complete nutrient profile of the ingredients. Asaolu *et al.* (2010) reported DMI values ranging from 54.6 to 59.6 g/kg-0.75 for West African Dwarf goats on groundnut hay basal diets, and offered Moringa and bamboo foliages as supplements. The observed high DMI of the experimental diets by the goats indicate the adaptability of the goats to the test ingredient. This trend of feed intake observed by the bucks could also be as a result of the acceptability and palatability of the feed, because the higher the palatability of a feed, the more the animal consumes the feed (Asaolu *et al.*, 2012). Masafu (2006) also described feed intake as a measure of diet appreciation, selection and consumption by an animal. The mode of feed presentation can stimulate the appetite and feed intake thus the high value of intake for the diets. This can be deduced that more of a diet with high crude protein would be consumed compared with that of low crude protein content (Taiwo and Anosa, 1995). Crude protein intake was highest for 5% and lowest for 0% MOLM -between the treatments ($p < 0.05$). Similar observations had been earlier reported by Arigbede (2007). Animals fed at 5% MOLM had the highest crude protein intake which could be due to variation in the level of inclusion. (Becker 1995) reported that the crude protein content of *Moringa oleifera* is of better quality for ruminants because of its high content of by-pass protein (47% versus 30% and 41% for *Gliricidia sepium* and *Leucaena leucocephala* respectively). Higher proportions of by-pass protein have been reported to result in faster weight gains in livestock (McNeill *et al.*, 1998).

There were no statistically significant differences ($p \geq 0.05$) in the weight gain by the bucks throughout the course of experiment. This indicates that the MOLM did not affect the

metabolic processes, hormones for growth (body weight) It shows that the experimental diets contained a profile of important minerals and protein, vitamins, carotene and various phenolics which are responsible for growth (Awodele *et al.*, 2012). Growth can be expressed as the positive change in body weight per unit of time. The increase in body weight of farm animals is a reflection of the growth of tissues. Growth is strongly influenced by breed, milk production, the environment under which the animals are maintained, including the availability of adequate feed supply in terms of both quantity and quality (Sanusi, 2017).

The control diet showed no visible lesion, damage, degeneration or marked regeneration in all organs except for the kidney which showed mild degeneration which might not be as a result of the test ingredient but may be due to health challenges in the animals. Zafra *et al.*, (2010) suggested that worm burden, especially from *Fasciola hepatica* can cause hepatic damage and local cellular and humoral immune responses in goats. The histopathological examination of all the kidneys of the experimental animals fed 5 % *Moringa oleifera* inclusions suffered from tubular degeneration of the kidney, but the degeneration diminished as *Moringa oleifera* inclusion increased leading to tubular epithelial regeneration. This might be as a result of the cytokinin-type hormone present in the *Moringa oleifera* as reported by Foidl *et al.* (2001) which accelerates cell growth. Necrosis especially at the centrilobular areas accompanied with cellular infiltration by the mononuclear cells of the hepatocytes which were focal at 15% of the *Moringa oleifera* was similar to the ones observed by Odetola *et al.* (2012) when rabbits were fed with graded levels of whole kenaf. This differ from the report of Eugenio-Pérez *et al.*(2016) who stated that several hypotheses have been proposed to explain the mechanisms of liver injury caused by hepatotoxins such as quercetin, kaempferol, myricetin, gallic acid, ellagic acid present in moringa leaves (Thangaiah *et al.*, 2024). Sudden damage to

liver cells disrupts their transport function and the permeability of their membranes, causing the release of marker enzymes (Nurhayati *et al.*, 2024)

There was no sign of damage in the heart. The heart is the main organ responsible for pumping of blood and an important index of physiological, pathological and nutritional status in the animal (Ewuola *et al.*, 2004) to all parts of the body. It was observed that there were cases of lymphocytic infiltration of the lamina propria of the intestine across the treatments. This suggests that *Moringa oleifera* has immunogenic effect which can help to fight pathogens or diseases in the gastro intestinal tract. This will help the animal to have a healthy gastro intestinal tract, aids digestion, nutrient uptake and absorption. The level of mildness of the necrosis in the liver which is not severe could be as a result of the medicinal effect of *Moringa* and the nutrient density.

Conclusion and application

Air-dried Moringa oleifera should be included in concentrates to enhance better nutrient utilization and performance of West African dwarf bucks. The results obtained in the present study confirms a positive effect of using MOLM in feeds for goats. The response in terms of feed intake and growth indices show that the treatment diets can serve as sustainable feeds that will overcome dry season weight losses and poor performance in terms of health wise of the bucks. The results revealed that the MOLM up to 15% level did not affect growth, feed consumption, general health status of the animals. Histopathological results revealed that the incidence of lesions in some organs should be investigated further. In conclusion, *Moringa oleifera* leaf meal can be included at the tested levels of inclusion in diet.

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Declaration of interest statement:

The authors here-by declare no conflict of interest

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