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Zingiber officinale, a Phytogenic Feed Additive on Haematological and Serum Biochemical Indices of Yankasa Rams

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Authors' contributions

This work was carried out in collaboration between all authors. Author UMI managed the experimental process, wrote the first draft of the manuscript. Author NM designed the study, wrote the protocol, analyse the data, provided PG training for author UMI and finalized the manuscript. Author SAM critiqued the manuscript. Author IAA managed the literature searches and reviewed the manuscript. All authors read and approved the final manuscript.

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ABSTRACT

Aims: The study was carried out to evaluate *Zingiber officinale* supplementation on haematology and serum biochemical properties of Yankasa rams.
Methodology: Four complete experimental diets were formulated with graded levels of ginger at 0, 2.5, 5 and 7.5 g/kg inclusion levels. The four experimental diets were fed to twenty (20) intact male animals. A completely randomized experimental design (CRD) was used in the experiment with number of animals representing replication and graded levels of ginger representing treatments.
Results: Results indicated no significant difference (P>0.05) between treatment means on biochemical and haematological parameters (p>0.05).
Conclusion: It was concluded that ginger supplementation up to 7 g/kg did not have significant effect on the haematological and biochemical indices of Yankasa rams.

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1. INTRODUCTION

Herbs and spices such as ginger have been reported to enhance activities of digestive enzymes and nutrient absorption in livestock animals [1]. Studies on phytogenic additives, antibiotics and organic acids have suggested similar effect on the gut function [1]. These include reduced bacterial colony counts, fewer fermentation products, greater nutrient digestion reflecting an overall improved gut equilibrium [2]. In addition some of the herbs and spices and their derivatives have been reported to promote intestinal mucus production. This effect led to improved production performance following inclusion in animal diet [2]. The biological properties, antimicrobial and antioxidant properties of essential oils of different aromatic plants have been known for long. Due to increased utilization and application of these products, a systematic study on these plant extracts has become very important [3].

The goal of the potential use of these sources is to modify rumen fermentation and increase the efficiency of the symbiosis between ruminant and rumen microorganisms in order to improve animal's performance without negative impact on the environment [4].

Hematological studies are of ecological and physiological importance in helping to understand the relationship between blood characteristics and the environment [5] and in the selection of animals that are genetically resistant to certain diseases and environmental conditions. Hematological parameters are good indicators of physiological status of animals [6] and those parameters that are related to the blood and blood forming organs. As reported by [7], animals with good blood composition are likely to show good performance. The examination of blood gives the opportunity to investigate the presence of several metabolites and other constituents in the body of animals and it plays a vital role in the physiological, nutritional and pathological status of an animal [8]. According to [9], examining blood for its constituents can provide important information for the diagnosis and prognosis of diseases in animals. Blood constituent's changes in relation to the physiological conditions of health. These changes are valuable in assessing response of animals to various physiological situations [3]. According to [10], change in hematological parameters is often used to determine various status of the body and determine stresses due to environmental, nutritional and pathological factors.

However, many supplements are used in animal feeds usually without recourse to health and physiological implications to the animals. Because ingestion of some of these dietary components have measurable effects on blood composition [11,12] and may have long term effects on animal's performance [13]; measuring these implications through haematology and blood chemistry becomes highly relevant [14]. This is because blood conveys different feed components to different parts of the body that could influence normal body function and affects health, growth, maintenance and reproduction of the animals [15]. While other studies deal with moderating the effects of environment, feeds, genetic, and other biological factors on sheep performance, the present study investigated the haematology and blood chemistry of Yankasa rams fed supplemented ginger levels in guinea savannah region of Nigeria.

2. MATERIALS AND METHODS

2.1 Experimental Location

The experiment was conducted at the National Animal Production Research Institute (N.A.P.R.I.), Ahmadu Bello University (ABU), Shika, Zaria, Nigeria. Shika is geographically located at latitude 11°12'N and longitude 7°33'E at an altitude of 640 meters above sea level [16]. It is about 20 km along the Zaria - Sokoto road in Kaduna State, Northern Nigeria. It has three distinct climatic seasons which include the cold dry season (November-February), the hot dry season (March-May) and the wet season (June-October). The total annual rainfall ranges from 617 to 1365 mm with a 50-year average of 1041 mm [17]. Most of the rains fall between July and September. Zaria falls within guinea savanna vegetation zone. The mean maximum temperature varies from 26°C to 35°C depending on the season, while the mean relative humidity during harmattan (winter) period is 21% [17].

2.2 Experimental Animals and Their Management

Twenty apparently healthy intact male Yankasa rams of between 12-18 months of age were used

for the experiment. The animals were procured from the Small Ruminant Research Programme unit of N.A.P.R.I. The animals were quarantined and put on adaptation to their new housing for two weeks. They were orally dewormed with albendazole (2.5% solution) to treat against gastrointestinal parasites. They were also treated with Oxytetracycline long acting broad-spectrum antibiotic based on the manufacturer's recommendation.

2.3 Sanitation and Health Management

Feces and urine were removed daily from the feeding pens to ensure adequate hygiene, less ammonia accumulation, optimal cleanliness of the experimental pens and ensure minimum discomfort and stress of the experimental animals. This practice was maintained for the entire period of the study.

2.4 Experimental Feed Preparation

Four complete experimental diets were formulated (Table 1) with graded levels of ginger at 0, 2.5, 5 and 7.5 g/kg inclusion levels. The four experimental diets were used for the trial. The diets were designated as diets 1, 2, 3 and 4 in the experiments. The formulation was done at the Feeds and Feeding Unit of the N.A.P.R.I. The ginger was sourced from local market and ground with peels before inclusion.

2.5 Experimental Design and Feeding Procedure

A completely randomized experimental design (CRD) was used in the experiment with number of animals representing replication and graded levels of ginger representing treatments. Five animals were allocated to each treatment and their pens were disinfected prior to the commencement of the experiment. The initial weights of the animals were balanced according to treatments. Each group was assigned to one of the experimental diets and fed in for 84 days. The experimental diet was fed according to the body weights (3% body weight) of the animals. The basal diet (*Digitaria* grass) was offered *ad-libitum* three (3) hours after feeding the concentrate. Clean drinking water was also offered *ad-libitum*.

2.6 Data Collection

The data were collected in three phases, as follows.

2.6.1 Phase I

2.6.1.1 Feeding trial

Body weight of each ram was taken at the beginning of the experiment (day 0). Subsequently, the rams were weighed weekly for body weight changes. Feed intake was measured and recorded daily by subtracting the left over from the quantity of feed offered to the animals the previous day. Feed conversion ratio was determined using feed intake and body weight gain. Nutrient intake was obtained from feed intake and chemical composition of the feed material. Feed and nutrient intake as % body weight was obtained from live weight and feed intake of the experimental animals.

Table 1. Gross composition of the experimental diets (%)

Ingredients	Treatments (Ginger level inclusion level (g/kg))			
	1 (0)	2 (2.5)	3 (5.0)	4 (7.5)
Maize	20.46	20.46	20.46	20.46
Cowpea husk	12.55	12.55	12.55	12.55
Cotton Seed Cake (CSC)	10.98	10.98	10.98	10.98
Rice offal	12.65	12.65	12.65	12.65
Cowpea hay	39.86	39.86	39.86	39.86
Salt	0.50	0.50	0.50	0.50
Bone meal	2.50	2.50	2.50	2.50
Premix	0.50	0.50	0.50	0.50
Total	**100.00**	**100.00**	**100.00**	**100.00**
Calculated nutrient contents				
Crude protein (%)	12.00	12.00	12.00	12.00
Energy (kcal/kg)	2600	2600	2600	2600
Crude fibre (%)	23.82	23.82	23.82	23.82
Ginger inclusion level (g/kg)	0	2.5	5.0	7.5

2.6.2 Phase II

2.6.2.1 Haematological assay

At the end of the feeding trial, 10 ml respective blood samples were collected from each of the animals for haematology, serum chemistry and electrolyte evaluation. Each blood sample was collected via the jugular vein into an ethylene-diamine tetra acetic acid (EDTA) coated bottle which serve as anti-coagulant and a plain tube. Each sample was allowed to stand at room temperature and then covered, centrifuged, the serum decanted and deep-frozen for serum biochemistry. Labeled samples (5 ml each) of the whole blood were taken to the ABU Teaching Hospital Zaria, Chemical pathology and haematology laboratory for analysis as follows:

2.6.2.2 Haematological indices

Packed cell volume (PCV) and haemoglobin concentration were determined by the microhematocrit and cyanmethaemaglobin methods respectively as described by [16]. Erythrocyte count was determined by the haematocytometry method as described by [18]. Total white blood cells (WBC) and differential counts were determined. Erythrocyte indices including Mean corpuscular volume (MCV), Mean corpuscular heamoglobin (MCH) and Mean corpuscular haemoglobin concentration (MCHC) were derived from the values obtained from red blood cells (RBC) count, haemoglobin concentration and packed cell volume (PCV) values [19,20] derived as follows;

$$MCV = \frac{PCV}{\text{RBC Count in } 10^6/mm^3} \times \frac{10}{1}$$

$$MCH = \frac{Hb\ (g/dl)}{\text{RBC (in } 10^6/mm^3)} \times \frac{10}{1}$$

$$MCHC = \frac{Hb\ (g/dl)}{PCV\ (\%)} \times \frac{100}{1}$$

2.6.3 Phase III

2.6.3.1 Serum chemistry

The plasma total protein was measured using biuret reaction while albumin was measured by colorimetric estimation using Sigma® diagnostic kit according to the method described by [21]. Globulin was obtained by calculating the difference of total protein and albumin. The serum enzymes, Aspartate aminotransferase (AST), Alanine aminotransferase (ALT) and alkaline phosphatase (ALP) were determined using a photoelectric calorimeter as described by [22]. Blood urea, nitrogen and creatine levels were also evaluated as described by [21].

2.6.3.2 Serum electrolytes

Serum sodium, calcium and potassium were determined using flame spectrophotometry as described by [22].

2.7 Statistical Analysis

The data generated from the experiment were subjected to analysis of variance (ANOVA) at 5% level of significance using completely randomized design using Statview Statistical Package [23]. Least significant difference (LSD) was used to separate the means.

3. RESULTS AND DISCUSSION

3.1 Phytochemical Constituents and Intake

Phytochemical analysis of the ginger indicated a higher oxalate content followed by flavonoid, phenols, alkaloids and saponins in that order. Tannins were the lowest constituents in the sample material (Table 2). Intake of the phytochemicals was higher for animals fed diets containing higher ginger levels and lower for the control diet. Dry matter intake (DMI) and average daily gain (ADG) of the animals decreased slightly with increased intake (Figs. 1 and 2).

Table 2. Quantitative phyto-chemical analysis of the test ingredient (ginger)

Constituents	Composition (mg/100 g ginger)
Tannin	0.26
Saponin	0.80
Oxalates	4.56
Alkaloids	1.20
Flavonoids	2.40
Phenols	1.52

The values for alkaloids, saponins and tannins in ginger are similar to the values respectively reported by [24]. However, the value for flavonoid obtained in the present study is higher than those obtained by [24]. Although animals fed higher ginger supplementation have a reduced DMI, ADG was not significantly affected due to beneficial effects of some phyto-chemicals taken along with other nutrients. Beneficial effect of low

to moderate concentrations of tannins to improve the digestive utilization of feed due to reduction of protein degradation in the rumen and a subsequent increase in amino acid flow to small intestine have been reported [25]. Saponins have also been found to enhance protection of protein from degradation in the rumen and increase bio-availability of protein post-rumination.

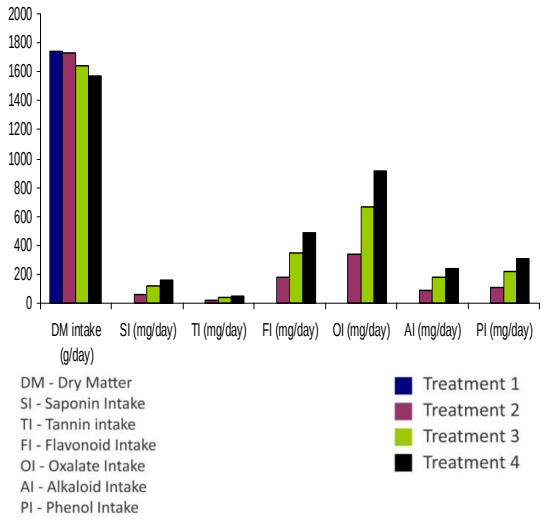

Fig. 1. Showing dry matter intake and phytochemical intake

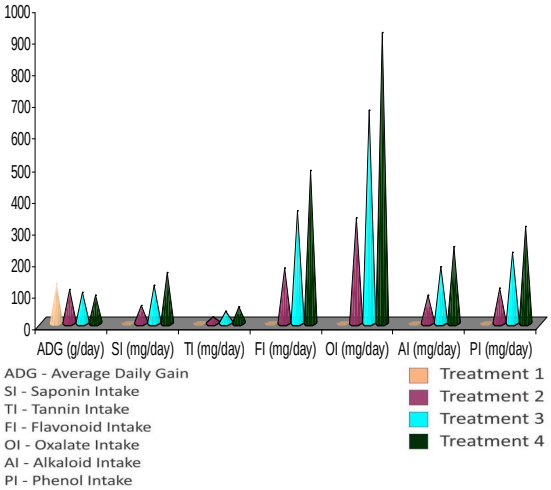

Fig. 2. Average daily gain and phyto-chemicals intake

3.2 Haematological Parameters

MCHC (g/dl) is significantly higher for treatment 3 compared to treatment 4 (P<0.01). There were no significant differences between treatments 2, 3 and 4 in terms of MCHC (g/dl) (P=0.05). All other parameters (PCV, Hb, WBC, RBC, MCV and MCH) showed no significant difference (P=0.05) between all the animals fed ginger (treatments 2, 3 and 4) irrespective of the inclusion levels.

The PCV values of 28.33-32.67% obtained in the present study were within the values 22.00-37.00% reported by [26] for normal healthy sheep. They are also close to 29.9–33.6% reported by [27] for clinically healthy sheep. The haemoglobin values 10.77–11.87% obtained in the present study are comparable to the normal reference values 8-16% as reported by [28]; so also the values of 23–48 fl and 31–38 g/dl for MCV and MCHC respectively. These suggest that even at the highest level of supplementation, ginger inclusion was not detrimental to the health of the animals. In addition, The PCV and Hb values are an indication that the animals were not anemic. For RBC, the range values of 12.03–12.90% obtained in the present study was above the range values of 2.40-4.20% reported by [26]. The values for WBC were above the normal range reported for healthy sheep by [25] suggesting the possible response of the animals to the effect of phyto-chemicals contained in the test ingredient. It has been reported that WBC in animals possesses phagocytic functions and are used as an indicator of stress and immune response to certain conditions [29]. The higher WBC reported in the present study further indicated that the animals seem to possess protective mechanism, providing a rapid and potent defense against infectious agents. This could be attributed to intake of some phyto-chemicals like flavonoids by the animals which functions in immune system by improving the activities of lymphocytes, macrophages, cells and increase phagocytosis as observed by [30].

3.3 Blood Chemistry

The blood chemistry parameters studied (Table 4) were not affected (p>005) by inclusion level of *Z. officinale*.

The values obtained for blood urea are similar to the values reported by [31] for normal healthy sheep. This is an indication that the diets are balanced in nitrogen and are thus safe for ruminant consumption. Na is the most important cation in extracellular fluid, where it is responsible for maintenance of osmotic pressure. The values recorded from the present study (136-145%) were close to the value of 136.6%

Table 3. Haematological parameters of Yankasa rams fed graded levels of *Z. officinale*

Parameter	Treatments (Ginger level inclusion level (g/kg))				SEM
	1 (0)	2 (2.5)	3 (5.0)	4 (7.5)	
Haemoglobin (g/dl)	11.87	11.83	10.77	11.37	0.65
PCV (%)	32.00	32.67	28.33	32.67	1.70
WBC ($\times10^9$/L)	15.10	13.17	11.93	12.10	1.27
RBC ($\times10^{12}$/L)	12.73	12.90	12.8	12.03	2.05
MCV (pl)	25.67	25.87	24.87	29.33	4.50
MCH (pg)	9.53	9.37	9.53	10.17	1.68
MCHC (g/dl)	37.13^{ab}	36.17^{ab}	38.10^{a}	34.80^{b}	0.64

Means within the same row with different subscripts are significantly different (p<0.05).
PCV-packed cell volume, WBC-white blood cell, RBC-red blood cell, MCV-mean corpuscular volume, MCH-mean corpuscular haemoglobin, MCHC-mean corpuscular haemoglobin concentration

Table 4. Blood chemistry of Yankasa rams fed graded levels of *Z. officinale*

Parameter	Treatments (Ginger level inclusion level (g/kg))				SEM
	1 (0)	2 (2.5)	3 (5.0)	4 (7.5)	
Urea (mmol/l)	3.80	3.63	3.63	3.33	0.43
Na (mmol/l)	136.33	147.33	149.00	145.00	3.95
K (mmol/l)	5.20	5.27	5.83	5.03	0.49
Cl (mmol/l)	102.67	110.67	111.51	109.57	3.00
Hco3 (mmol/l)	23.00	24.67	21.00	22.00	0.90
Creatine (mmol/l)	68.67	68.67	86.67	65.00	25.78
Ca (mmol/l)	2.38	2.30	2.29	2.36	0.04
Tp (g/l)	66.67	61.33	68.00	68.33	3.50
Alb (g/l)	32.67	37.33	36.00	36.00	1.78
Ast (iu/l)	114.67	117.00	112.00	117.67	28.24
Alt (iu/l)	18.67	19.67	12.33	25.00	3.77
Alp (iu/l)	708.33	686.00	584.67	545.67	129.72

Means within the same row with different subscripts are significantly different (p<0.05)
Tp-total protein, Alb-albumin, Ast-Aspertate aminotransferase, Alt-Alanin aminotransferase Alp-Alkaline phosphatase, Na-sodium, K-potassium, Ca-calcium, Hco3-bicarbonate, Cl-chlorine

reported by [32]. K is important for regulation of acid-base balance in the body. The values of K obtained from this study were within the range 5.13–5.60 mmol/l reported by [33]. The same observation was also made for Ca as obtained by [34]. The effect of dietary treatments was not significant (p>0.05) on the total blood protein levels of the animals. The creatine and albumin were slightly higher to the normal reference values reported by [35]. The creatine level could be attributed to the growing stage of the animals because it increases with increase in age as observed by [36]. Bicarbonate, AST and Chlorine are within the normal ranges. Though the ALP is slightly higher; high ALP may indicate a liver problem and alkaloid toxicity. However, high levels of ALP can be normal in growing animals due to growth of bones [37]. The ALP being high is an indication of high protein intake. The AST value was not high enough to impede liver function of the animals. ALT also falls within the reference range [37].

4. CONCLUSION

It was concluded that ginger supplementation up to 7.5 g/kg did not have significant effect on haematology and blood chemistry of Yankasa rams. Future research should be conducted with varying inclusion levels on haematology and biochemical indices of the animals.

COMPETING INTERESTS

Authors have declared no competing interests regarding the publication of the manuscript.

REFERENCES

1. Madigan M, Martinko J. Brock biology of microorganisms (11th ed.). Prentice Hall; 2005.
ISBN 0-13-144329-1
2. Odoemelam VU, Idorenyin FE, Ndelekunle EU. Options for sustainable animal

production. Journal of Biology, Agriculture and Health Care. 2013;90:511 520.

3. Hert KJ, Yanez R, Duval SM, Ewan NR, Newbold CJ. Plants extract to manuplate rumen fermentation. Animal Feed Science Technology. 2008;147:8-35.
4. Johnson JK, Moris DD. Alterations in blood proteins in B. P. Smith (Ed), International Animal Medicine (2nd ed.) USA: Mosby Publishers; 1996.
5. Ovuru SS, Ekweozor IKE. Haematological changes associated with crude oil ingestion in experimental rabbits. African Journal of Biotechnology. 2004;3(6):346-348.
6. Khan TA, Zafar F. Haematological study in ruminants. International Journal of Ruminant Science. 2005;4(10):748-751.
7. Isaac LJ, Abah G, Akpan B, Ekaette IU. Haematological properties of different breeds and sexes of rabbits. Proceedings of the 18th Annual Conference of Animal Science Association of Nigeria. 2013;24-27.
8. Aderemi FA. Effect of replacement of wheat bran with cassava root on the haematology and serum biochemistry of ruminants. Tropical Journal of Animal Science. 2004;7:147-153.
9. Olafedehan CO, Obun AM, Yusuf MK, Adewumi OO, Adeniji AA. Effects of residual cyanide in processed cassava peal meals on haematological and biochemical indices of growing West African Dwarf goats. Processings of 35th Annual Conference of Nigerian Society for Animal Production. 2010;212.
10. Afolabi KD. Haematological parameters of the Nigerian local breeds of sheep fed varying dietry levels of palm kernel cake. Proceedings of 35th Annual Conference for Nigerian Society of Animal Producton. 2010;247.
11. Church JP, Judd JT, Young CW, Kebay JL, Kim WW. Relationships among diet constituents and specific serum clinical components of subjects eating self-selected diets. African Journal of Clinical Nutrition. 1984;40:1338-1344.
12. Maxwell MH, Robertson GW, Spences A, McCongrouodala. Composition of haematological values in restricted and ad libitum feed domesticated fowls. RBC characteristics. British Poultry Science. 1990;60:1474-1484.
13. Olabanji RO, Farinu GO, Akinlade JA, Ojebiyi OO. Growth performance and haematological characteristics of weaner rabbits fed different levels of wild sunflower (*Tithonia diversifolia* Hems L A. Gray) leaf blood meal mixture. Proceedings of 32nd Animal Conference of Nigerian Society for Animal Production. Ojanuga AG. 2004; 207-209. Agroecological zone maps of Nigeria. National Special Programme for Food Security, FAO-UNESCO. 2007;124.
14. Aro SO, Ogunwale FF, Falade OA. Blood viscosity of finisher cockerel fed dietary inclusions of fermented cassava tuber wastes. Proc. of the 18th Annual Conf. of Anim. Sci. Assoc. of Nig. 2013;74-77.
15. Oke UK, Herbert U, Ebuzoeme CO, Nwachukwu EN. Effect of genotype on the haematology of Nigerian local chickens in a humid tropical environment. Proceedings of 32nd Annual Conference of Nigerian Society for Animal Production; 2007.
16. Akpa GN, Ifut JO, Mohammed F. Indigenous management of dystocia in Ruminant Livestock of Northern guinea savannah of Nigeria. Journal of Animal Production. 2002;29:264-270.
17. Oni OO, Abubakar BY, Ogundipe SO. Genetic and phenotypic associations of juvenile body weights and egg production traits in 2 strains of Rhode Island chickens. Nigerian Journal of Animal Production. 1991;18:66-69.
18. Bush BM. Interpretation of laboratory results from small animal clinician. United Kingdom: Black Well Scientific Publication. 1991;122.
19. Jain NC. Essentials of veterinary haematology (1st ed.) Philadelphia: Lea and Febiger Publishers. 1993;356.
20. Schalm OW, Jain NC, Carrol EJ. Veterinary haematology. USA: Lea and Febiger Publishers, Philadephia; 1975.
21. Baker FJ. Introduction to medical laboratory technology (6th ed.). London: Butterworth; 2007.
22. Duncan JR, Prasse KW, Mahaffey EA. Veterinary laboratory, medicine (clinical Pathology). Iowa State University Press: Ames. 1994;94-96.
23. SAS. Stat view statistical package, SAS incorporated, New York; 2002.
24. Denman SE, McSweeney CS. Quantitative (real-time) PCR. In methods in gut microbial ecology for ruminants (ed. HPS Makkar and CS McSweeney). Springer, Dordrecht, The Netherlands. 2005;105–108.

25. Frutos P, Raso M, Hervas G, Mantecon AR, Perez V, Giraldez FJ. Is there any detrimental effect when a chestnut hydrolyzable tannins extract is included in the diet of finishing lambs? Anim Res. 2004;56:127-136.
26. Sowande OS. Body measurement of West African dwarf sheep as parameter for estimation of live weight. Tropical Animal Health and Product. 2008;4(6):443–449.
27. Mitruka BM, Rawnsley HM. Clinical biochemical and hematological reference values in normal experimental animals. Masson Publishing, New York; 1977.
28. RAR. Reference values of laboratory animals: Normal haematological values; 2009.
Available:www.ahc.umn.edu/rar/refvalues.html
(Retrieved April 20, 2015)
29. Carsky RJ, Tarawali SA, Becker M, Chikoye D, Tian G, Sangiga N. Mucuna: Herbaceous cover legume with potential multiple use. Resource and crop management. Research monograph. No. 25. International Institute of tropical Agriculture, Ibadan, Nigeria; 1998.
30. Craig WJ. Health-promoting properties of common herbs. American Journal of Clinical Nutritrion. 1999;70:491S–499S.
31. Jokhtan GE, Braimah Y, Muhammed IR, Abdu SB, Mohammed RH. Evaluation of the performance of Yankasa sheep fed acacia sayel del. (Chanchinlo) pods replacement for cotton seed cake. Nigerian Journal of Animal Science. 2010;12:129-136.
32. Bouda J, Jagos P. Biochemical and hematological reference values in ruminants and their significance for health control. Acta Veterinaria Brno. 1983;53(3-4):137-142.
33. Tucker J, Aduku JK. Haematological study in ruminants. International Journal of Ruminant Science. 1999;1984:8(20):548-551.
34. Okine EK, Mathison GW. External and internal markers for appraising site and extent of digestion in ruminants. Journal of Dairy Science. 1991;76:129.
35. Duncan JR, Prasse KW, Mahaffey EA. Veterinary laboratory medicine (Clinical pathology). Ames: IOWA State University Press. 2012;442.
36. Myer DJ, Enrich DJ. Veterinary laboratory medicine interpretation and diagnosis. Philadephia: WB saunders co., USA; 1992.
37. Infovets. ALP reference values; 2015.
Available:infovets.com/books/smrm/D/D080.htm

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