



Haematological and Biochemical Indices of Yankasa Rams Fed *Brachiaria Ruziziensis*-*Centrosema Pascuorum* Hay Mixtures with Concentrate

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ABSTRACT

Blood haematological and biochemistry studies of Yankasa rams fed *B. ruziziensis* and *C. pascuorum* hay mixtures supplemented with concentrate was investigated using Yankasa rams. Twelve Yankasa rams with average initial weight 10.25 ± 0.1 kg were assigned to three dietary treatments of *B. ruziziensis* and *C. pascuorum* hay at different mixtures (75BR:25CP, 50BR:50CP and 25BR:75CP, respectively) with concentrate in a Completely Randomized Design for a period of 90 days. Results indicated Packed Cell Volume before feeding was 30.0% in 50BR:50CP, but after feeding, animals on 25BR:75BR diet had the highest PCV value (36.8%). Also, blood haemoglobin concentration in the rams was highest (10.1g/dl) in animals fed 25BR:75CP diet before feeding while after feeding, animal fed 50BR:50CP diets had the best value (10.5g/dl). Blood chemistry after feeding were mostly not significant ($P > 0.05$) from each other, except in glucose levels, alkaline phosphatase (ALP) and urea N which showed significant difference ($P < 0.05$). Animals fed 75BR:25CP diets had better glucose (54.0mmol) and ALP (18.0u/l) values after feeding, while urea-N was higher (28.80mmol) in animals fed 25BR:75CP diets. The urea-N was highest ($P < 0.05$) at 50BR:50CP animals both before and after feeding. Animals fed 25BR:75CP diets gave best values for RBC, WBC, MCHC, creatinine, calcium and Urea-N. Therefore, *B. ruziziensis* and *C. pascuorum* hay be mixed at 25%:75%, supplemented with concentrate for better performance in Yankasa rams.

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INTRODUCTION

Feeds shortage is a critical problem in livestock farming in most areas of the tropics. As grazing lands are gradually brought under cultivation to satisfy the food needs of the increasing human population, livestock are forced to graze on marginal

areas, which combined with the use of low-quality crop residues as feeds has resulted in poor livestock performance. Natural pastures, from such marginal lands are generally high in fibre, low in protein and energy yet they form the main source of animal feed in Tropical Africa (Teshome,



1987). These resources are over utilized to the extent that they fail to meet even the maintenance requirements of indigenous animals especially when the dry season persists for long periods. This has resulted in significant decrease in milk production, loss of body weight, reduced draught power, increased susceptibility to diseases, reduced reproductive performance, retarded growth rate and high mortalities of young animals (Alemayehu, 1997).

The Nigerian livestock population estimates about 13.9 million cattle, 22.1 million sheep and 34.5 million goats (Lawan, 2012, NIAS, 2019). Sources of cheaper alternative forages of high quality for ruminant livestock production have been a subject of research in recent years (Alan *et al.* 2013) especially for small scale livestock producers in tropical areas during the dry season.

The nutritive value of Congo grass (*Brachiaria ruziziensis*) steeply declines with maturity, the crude protein declines to 9-10% after 10 weeks of re-growth, and can be lower than 8% after 15 weeks of re-growth (Milford and Minson, 1968). Increasing livestock production in Nigeria requires an increase in the area and quality of legume-based pasture.

Centrosema pascuorum is a vigorous, trailing, twining and climbing perennial herb with trifoliolate leaves and is fairly drought tolerant (Skerman *et al.*, 1988). Muhammad *et al.* (2002) recommended that *Centrosema spp.* be integrated into the crop-livestock farming system of the low land areas of Northern Nigeria. Ndluvo (2000) reported judicious combinations of these feeds with the more abundant low-quality forages are needed. The digestibility would probably increase as the proportion of forage legume increases because the legumes often have higher digestibility than grasses (Topps, 1995).

Justification for the Study

The low nitrogen (N) content of most matured grasses points to a need to combine them with forage legumes with high nitrogen (N) content. However, it has been reported that the high rates of release of soluble protein and breakdown to small particles from herbaceous legumes is associated with susceptibility of ruminants to bloat (Dewhurst *et al.*, 2009). Feeding of legume species alone to the ruminants is therefore not advisable because of the problem of bloat. Moreover, it has been reported that, the efficiency of N used declined with increasing proportion of legumes in animal diets. So, higher legume proportion in ruminants' diets should be discouraged because it leads to waste of nitrogen and bloat susceptibility (Dewhurst *et al.*, 2009).

This study investigated the effect of mixtures of *B. ruziziensis* and *C. pascuorum* hay supplemented with concentrate on the blood haematology and biochemical profile of growing Yankasa rams.

MATERIAL AND METHODS

Experimental Site

The feeding trial was conducted at the Teaching and Research Farm of the Department of Animal Production, Abubakar Tafawa Balewa University, Bauchi State. Bauchi lies between latitude 10°33'N and longitude 9°31'E. It is bordered by Seven States, Kano and Jigawa to the North, Taraba and Plateau to the South, Gombe and Yobe to the East and Kaduna to the West. It occupies a total area of 49,259.01sq kilometers, representing about 5.3% of the total land mass of Nigeria (Ovimap, 2016).

Management of Experimental Animals

Twelve (12) Yankasa rams of between 9-12 months of age weighing between 10.18-10.28kg were used for the growth performance and nutrient digestibility studies. Each animal was given



prophylactic treatment consisting of Ivermectin (*Ivomec*[®]) at 0.5ml/25kg body weight subcutaneously against ectoparasites (such as fleas, mites and ticks e.t.c) and Oxytetracycline (*Tridox*[®]) antibiotics at 1.0ml/10kg body weight intramuscularly against bacterial infections one week before the start of the experiment. Other routine management practices included; deworming against intestinal parasites using *Albendazole*[®] 10% solution which was administered in drinking water. The experimental animals were housed individually in an open sided and well-ventilated pen. The pens were cleaned, washed and disinfected with Izal solution before the arrival of the animals. The rams were allowed an adaptation period of two (2) weeks during which they were fed with

the experimental diets and were given access to fresh clean water.

Experimental Design and Treatments

Twelve (12) Yankasa rams were randomly allotted into three (3) dietary treatments with four (4) rams per treatment in a Completely Randomized Design (CRD). The experimental diets were: **Treatment 1:** 75% *B. ruziziensis* and 25% *C. pascuorum* hay, **Treatment 2:** 50% *B. ruziziensis* and 50% *C. pascuorum* hay, **Treatment 3:** 25% *Brachiaria ruziziensis* and 75% *Centrosema pascuorum* hay, respectively.

A concentrate diet was formulated using; maize offal, cotton seed cake, cowpea husk, bone meal and common salt as shown in Table 1. The ingredients used in formulation of the concentrate diet were purchased from Muda Lawal market at Bauchi, Bauchi State.

Table 1: Percentage composition of feed ingredients in the concentrate diet fed to Yankasa rams

Feed ingredients	% composition
Maize offal	67.0
Cotton seed cake	21.0
Cowpea husk	10.0
Bone meal	1.50
Common salt	0.50
Total	100
ME (MJ/kgDM)	11.81
Crude protein	13.78

ME= Metabolizable energy

The concentrate feed was offered to the animals at a fixed amount (200g) while experimental diets consisting of forage mixtures were offered at 3% body weight of the animals throughout the experimental period of 90 days.

Experimental Procedure and Data collection

The animals were fed experimental diets (concentrate and basal diet) at 8:00 a.m in the morning and 4:00 p.m in the evening. The concentrate and experimental diets were weighed before feeding to the animals and left-overs weighed the next

morning before feeding to determine the feed intake daily. The experimental animals were provided with fresh and clean drinking water in a graduated plastic container. The amount of water consumed by each animal daily was measured and recorded.

Haematological Indices and Blood Biochemical Profile Measurement

Towards the end of the feeding trial, blood sample were collected from the jugular vein of the animals using disposable syringe with needle at the 10th week of the trial period. About 5ml of blood was



collected from each animal into plastic tubes containing ethylene diamine tetra acetic acid (EDTA) which is an anti-coagulant for haematological analysis. Blood sample were collected in the morning before feeding and 4 hours after feeding. Packed cell volume (PCV) and haemoglobin concentration (Hb) determination was according to the Procedures outlined by Dacie and Lewis (2001), red blood cell (RBC) and differential total white blood cell (WBC) counts were carried out using Neubauer Haemocytometer after appropriate dilution according to the Method of Jelalu (2014). Mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) were calculated according to the Procedures of Jain (1993).

Similarly, at same period as above blood samples of about 5ml were also collected into anti-coagulant free plastic tubes and allowed to coagulate at room temperature. The supernatant sera were

stored in a freezer for subsequent biochemical analysis. Biochemical analysis was done using the Method described by Ogunsanmi *et al.* (2002). Serum total protein was determined by using Buret Method as described by Kohn and Allen (1995) while Albumin was determined using Bromocresol Green (BCG) Method as described by Peters *et al.* (1982). The Globulin concentration was obtained by subtracting Albumin from the Total Protein. Serum urea was determined by urease method and creatine by Folinwu filtrate methods as described by Toro and Ackermann (1975).

RESULTS AND DISCUSSION

Chemical Composition of Experimental Feed Materials Fed To Yankasa Rams

Table 2 showed the percentage composition of feed ingredients in the concentrate diet fed to Yankasa rams in this study while the chemical composition of individual feed ingredients in the diet of Yankasa ram is presented in Table 2.

Table 2: Chemical composition of feed materials of the experimental diets fed to Yankasa rams

Parameters (%)	<i>B. ruziziensis</i>	<i>C. pascuorum</i>	Concentrate diet
Dry matter	90.84	88.13	87.65
Organic matter	81.53	82.59	82.40
Crude protein	7.84	13.01	13.78
Neutral detergent fibre	52.19	44.32	46.55
Acid detergent fibre	49.69	33.53	37.21
Ether extract	0.65	0.63	0.63
Ash	9.31	5.54	5.25
Nitrogen free extract	30.01	36.50	33.79
ME (MJ/kgDM)	11.72	11.80	11.81

ME= Metabolizable energy

Table 3: Chemical composition of experimental diets fed to Yankasa rams

Parameters (%)	Mixture of <i>B. ruziziensis</i> and <i>C. pascuorum</i> hay (%)		
	75:25	50:50	25:75
Dry matter	89.22	88.68	88.52
Crude protein	8.82	10.64	12.14
Ether extract	0.34	0.36	0.39
Ash	2.70	2.62	2.97
Neutral detergent fibre	47.61	48.30	46.80
Acid detergent fibre	38.43	39.22	37.74

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Nitrogen free extract	40.53	38.08	37.70
ME (MJ/kgDM)	11.80	11.82	11.82

ME= Metabolizable energy

Haematological Indices of Yankasa Rams Fed Diets of Mixtures of *B. Ruziziensis* and *C. Pascuorum* Hay Supplemented with Concentrate

The result of haematological parameters of Yankasa rams fed the experimental diets are presented in Table 4. The values obtained for packed cell volume in this study ranged from 26.0-30.0% before feeding and 31.3%-36.8% after feeding treatment diets and were all significantly different ($P<0.05$) both before and after feeding across treatments. The increase in PCV across treatment after feeding could be attributed to increase in feed intake and quality of the diet as *C. pascuorum* increased in the diet (Ross *et al.*, 1978) while the decrease in value before intake could be as a result of reduction in energy content of the mixtures. The results obtained is in contrast with the report of Jain (1993) that an increase in the energy levels of feed leads to an increase in the PCV levels of blood. The results of PCV in this study were within the normal range of 25-45% (Cole, 1986 and Merck Manual, 2016) for rams and was similar to the result of Alasa (2014) when West African Dwarf Rams were fed *Panicum maximum* with Lablab mixture.

Similarly, haemoglobin concentration (Hb) values in this study ranged from 8.5-10.1 g/l before feeding and 8.8-10.5 g/dl after feeding. The increase in haemoglobin as *C. pascuorum* increased in treatment diets before and after feeding indicates higher feed intake and increased quality of the feed. It was observed that haemoglobin value decreased after feeding in 25BR:75CP (8.9 g/dl) from 10.5 g/dl in 50BR:50CP. This indicated a slight negative effect in the diet as *C. pascuorum* exceeded 50% probably due to increased NH_3^+ in the rumen of the rams. Haemoglobin in the

cytoplasm of the red blood cell gives an indication of the oxygen carrying capacity of the animal blood. This was also observed by Alasa (2014) with *P. maximum* 25% and *L. purpureus* 75% mixture down from 50% *P. maximum* 50% *L. purpureus* mixture in a similar experiment when *P. maximum* and *L. purpureus* were fed to WAD rams. The haemoglobin concentration in this study was within the normal range for rams reported by (Coles, 1986, Plumb, 1999, Merck Manual, 2016).

The red blood cells increased both before feeding and after feeding up to 50BR:50CP mixture, after which it declines in 25BR:75CP diet. There was no significance difference ($P>0.05$) in red blood cells across treatment before and after feeding. Shakoore *et al.* (1990) suggested that decrease in RBC count is either an indication of excessive damage to erythrocyte or inhibition of erythrocyte formation. This could suggest that 25%B.R:75%C.P diet had some detrimental effect on red blood cell of the animals. Generally, values obtained in this study was lower than the normal values or range of $9-15 \times 10^6 \text{mm}^3$ for sheep reported by Merck Manual (2016) but compared with RBC counts of 7.58-9.94, 6.4-10.47 and 6.4-14.70 $\times 10^6 \text{mm}^3$ for local, improved and Afec sheep respectively (Jawasreh *et al.*, 2009). Low number of red blood cell can be the result of blood loss (anaemia), inadequate RBC production in bone marrow or actual destruction of the RBC within the body. Similarly, the variation in RBC counts in this study with other studies might be attributed to the age, gender of the animals used and geographical location for the study.

The values obtained in this study for white blood cells ranged from 6.9-13.8 $\times 10^6 \text{mm}^3$ before feeding and 7.6-14.4 $\times 10^6 \text{mm}^3$ after. The white blood cell count



before feeding was significantly higher ($P < 0.05$) in animals on 25%B.R:75%C.P ($13.8 \times 10^6/\text{mm}^3$) compared to values obtained in other treatment diets. White blood cells (WBC) have the chief function of importing immunity into the body (Okunade *et al.*, 2015).

The mean corpuscular haemoglobin (MCH/g) in this study ranged from 13.5-14.4 pg before and 13.7-13.9 pg after feeding and higher than the normal range reported by Merck Manual (2016) for sheep but similar to the values of 9.6-14.4pg for improved Awassi sheep by Jaswareh *et al.*, 2009. Values obtained in this study was not significantly different ($P > 0.05$) between treatments and were within the normal range of 31-34pg for sheep reported by Plumb, 1999. Mean corpuscular haemoglobin concentration (MCHC) is very significant in diagnosis of anaemia and is an index of the capacity of the bone marrow to produce RBC (Njidda, 2013).

Values for lymphocyte before and after feeding were significantly higher ($P < 0.05$) in animals fed 25%B.R:75%C.P diet (70.9% and 57.5%, respectively) compared to values obtained in other treatments. It ranged from 63.7-70.9% before feeding and 25.3%-57.5% after feeding and were mostly within the normal range of 40-70% reported for sheep (Duncan and Prasse, 1986). The lower value below the normal range obtained after feeding in 75BR:25CP and 50BR:25CP could be attributed to the animals being stressed up during blood collection (Girgiri, 2018). Lymphocytes are also WBC that are parts of the immune system. High levels of lymphocytes (lymphocytosis) can occur when animal has achronic infection or lymphocyte leukemia and low level of lymphocytes (lymphopenia) indicates administration or release of steroids, viral infection or stress.

Table 4: Haematological indices of Yankasa rams fed diets containing mixtures of *B. ruziziensis* and *C. pascuorum* hay supplemented with concentrate

Parameters	Period	Mixtures of <i>B. ruziziensis</i> and <i>C. pascuorum</i> hay (%)			SEM
		75:25	50:50	25:75	
Packed cell volume (%)	BF	27.4 ^b	30.0 ^a	26.0 ^a	1.25
	AF	31.3 ^a	35.4 ^a	36.8 ^b	0.68
Haemoglobin (g/dl)	BF	8.50 ^b	9.1 ^a	10.1 ^a	0.20
	AF	8.8 ^b	10.5 ^a	8.9 ^b	0.23
Red Blood cell ($\times 10^6/\text{mm}^3$)	BF	6.1	7.31	7.03	0.16
	AF	6.36	6.83	6.39	0.06
White Blood cell ($\times 10^6/\text{mm}^3$)	BF	7.8 ^b	6.9 ^b	13.8 ^a	0.92
	AF	7.9	7.6	14.4	0.95
Mean corpuscular volume (fl)	BF	44.9 ^c	48.5 ^b	52.4 ^a	0.92
	AF	49.2 ^a	33.3 ^c	40.7 ^b	1.96
Mean corpuscular haemoglobin (fl)	BF	13.9	13.5	14.4	1.80
	AF	13.8	13.7	13.9	0.02
MCHC (pg)	BF	31.0 ^a	31.6 ^a	27.4 ^b	0.56
	AF	28.2 ^c	31.0 ^b	34.2 ^a	0.74
Lymphocyte (%)	BF	63.7 ^b	65.2 ^b	70.9 ^a	0.94
	AF	25.3 ^b	27.9 ^b	57.5 ^a	4.40

^{a, b, c} Means in the same row with different superscript are significantly different ($P < 0.05$), **MCHC** = Mean corpuscular haemoglobin concentration **SEM**= Standard Error of Mean, **BF**= Before feeding, **AF**= After feeding



Blood Serum Chemistry of Yankasa Rams Fed Diets of Mixtures of *B. Ruziziensis* and *C. Pascuorum* Hay Supplemented With Concentrate

The result of blood serum biochemical parameters of Yankasa rams is shown in Table 5. The Creatinine values both before and after feeding were not significantly different ($P>0.05$) across treatments. The creatinine value ranged from 0.79mmol before feeding in rams fed 25%B.R:75%C.P diet to 1.03mmol in rams fed 75%B.R:25%C.P. Also, Creatinine value after feeding ranged from 0.77mmol in 75%B.R:25%C.P to 0.98mmol in animals fed 50%B.R:50%C.P diet. Creatinine values in this study are similar to the values obtained by Alasa (2014), 0.9-2.0 mg/dl reported by Plumb (1999) and compared with the normal range of 1-2.7mg/dl reported by Merck Manual (2016). Elevated creatinine levels may indicate renal dysfunction which is not the case in this study, suggesting the animals used in this study were healthy.

There was no significant difference ($P>0.05$) in glucose value between treatment means before feeding but it varied significantly ($P<0.05$) after feeding. Serum glucose obtained in this study ranged from 49-52mg/dl before feeding and 49-54 mg/dl after feeding. It was observed that there was an increase in blood glucose concentration before feeding and a decrease in glucose concentration after feeding as the level of *B. ruziziensis* decreased in the diet. This could be as a result of *B. ruziziensis* being a grass and contained more energy and carbohydrate than *C. pascuorum*. Blood glucose values in this study were generally within the normal range of 40-100mg/d for sheep reported by Khan (2010) and also 44-81 mg/dl by Plumb (1999). Generally, normal glucose levels in the ram indicate synthesis in the liver from propionate metabolism as the major glucose precursor (Houtert, 1993).

The mean values for albumin was not significantly different ($P>0.05$) across all

the treatments both before and after feeding. Blood albumin in this study ranged from 3.4-4.0 g/dl before and was 3.2-4.3 g/dl after feeding. The values were similar to the values of 2.8-4.34 g/dl obtained by Alasa (2014) in a similar experiment on WAD goats feeding *P. maximum* and *L. purpureus* on similar mixture ratio. It was however slightly higher than the normal range of 2.6-4.0 g/dl for sheep reported by Merck Manual (2016) and 2.7-3.0 g/dl by Plumb, (1999). The highest value (4.3 g/dl) for albumin was obtained in 50BR:50CP treatment animals after feeding, suggesting higher clotting ability of blood, hence prevention of haemorrhage.

The result for alkaline phosphatase showed significant difference ($P<0.05$) before and after feeding in experimental rams. It ranged from 10.2-19 u/l before feeding and 10-18u/l after feeding and were all within the range of 10-25u/l reported for normal sheep. Values for ALP when reasonably low indicate that the diet had enough protein as high value of ALP is an indication of poor-quality protein of the experimental diet (Ologhobo *et al.*, 1993). Therefore, the highest value obtained in 75%B.R:25%C.P showed that 75%B.R:25%C.P had less quality protein in the diet which increased with increase in *C. pascuorum* in the diet of experimental rams.

Blood calcium was highest before and after feeding in animals fed 50%B.R:50%C.P diets and were all not significantly different ($P>0.05$) compared to values obtained in other treatment diets. Calcium values generally increased with increase in *C. pascuorum* in the diet. This could be attributed to the higher levels of calcium obtained in forage legumes compared to grasses. It ranged from 2.8-2.9 mmol/l before feeding to 3.2-3.9 mg/dl after feeding, adequate calcium levels in the blood ensures proper growth in young animals.



Biochemical indices of Yankasa rams in this study indicated that urea N were significant ($P < 0.05$) before and after feeding. The highest level of blood urea was recorded for animals on 25%B.R:75%C.P diet. It was observed that urea nitrogen level (N) increased both before and after feeding as the level of *C. pascuorum* increased across the treatment diets. This could be due to the increase in contribution of crude protein from *C. pascuorum* in the diet. The values obtained in 75%B.R:25%C.P and 50%B.R:50%C.P were within the range of 10-

25mg/dl for sheep by Khanum *et al.* (2010). Values obtained in 25%B.R:75%C.P was however higher than the normal range reported by Khanum *et al.* (2010) but was similar to the values reported by Alasa (2014). When blood urea is higher than normal range, it could be an indication of kidney failure and when lower could indicate decreased hepatic functions. All animals during the experimental period looked healthy suggesting that there was no negative effect on blood urea of Yankasa rams.

Table 5: Blood serum chemistry of Yankasa rams fed diets containing mixtures of *B. ruziziensis* and *C. pascuorum* hay supplemented with concentrate

Parameters	Period	Mixtures of <i>B. ruziziensis</i> and <i>C. pascuorum</i> hay (%)			SEM
		75:25	50:50	25:75	
Creatinine (mg/dl)	BF	1.03	0.89	0.79	0.03
	AF	0.77	0.98	0.89	0.03
Glucose (mg/dl)	BF	49.0	44.0	52.0	0.99
	AF	54.0 ^a	52.0 ^b	49.0 ^c	0.62
Albumin (g/dl)	BF	3.80	3.40	4.00	0.08
	AF	3.20	4.30	3.60	0.14
ALP (u/l)	BF	19.0 ^a	16.0 ^b	10.2 ^c	1.10
	AF	18.0 ^a	12.0 ^b	10.0 ^c	1.03
Calcium (mmol/l)	BF	2.90	3.00	2.80	0.03
	AF	3.20	4.00	3.90	0.11
Urea N (mmol/l)	BF	9.40 ^b	24.90 ^a	28.40 ^a	2.49
	AF	13.80 ^b	23.70 ^a	28.80 ^a	1.88

^{a, b, c} Means in the same row with different superscript are significantly different ($P < 0.05$), **SEM**= Standard error of mean, **NS**= Not significant, **ALP**= Alkaline Phosphatase, **BF**= Before feeding, **AF**= After feeding

CONCLUSION AND RECOMMENDATION

Yankasa rams fed diet containing mixtures of 25%B.R:75%C.P diet had best values for red blood cell, white blood cell, mean corpuscular haemoglobin concentration, lymphocytes, creatine, calcium and urea N. Hence, it is recommended that *B. ruziziensis* and *C. pascuorum* be mixed at 25% *B. ruziziensis* and 75% *C. pascuorum* (1:1) in the diet of Yankasa rams for better performance in Yankasa rams.

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