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EFFECT OF WHOLE COTTONSEED AND COTTONSEED CAKE ON HEAMATOLOGY AND SERUM CHEMISTRY OF RED SOKOTO BUCKS

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ABSTRACT

A study was conducted to evaluate the effect of feeding cotton products (whole cottonseed (WCS) and cottonseed cake (CSC)) and level (10, 20 and 30%) on haematology and serum biochemistry of Red Sokoto bucks in a 2 x 3 factorial arrangement in a completely randomized design. A total of twenty-four healthy Red Sokoto bucks were randomly allocated to the treatments. The diets were fed to the bucks over a period of 98 days after a 14-day adjustment period. Blood samples were collected from all the bucks fortnightly through the jugular vein. Results showed no significant ($P>0.05$) differences in most haematological parameters and serum chemistry of bucks fed diets containing either WCS or CSC. However, PCV, RBC ($P<0.01$) and albumin ($P<0.05$) were significantly higher in bucks fed CSC diet than those fed WCS diet. Total proteins ($P<0.05$) and globulin ($P<0.01$) levels increased significantly with increasing inclusion of the cotton products. Lymphocyte count increased slightly from 63.83 (10%) to 64.25 (20%) and then dropped sharply to 54.94 on 30% level of inclusion. Neutrophils were similar for bucks fed 10 and 20% inclusion of the cotton products but lower than 30%. There was significant decrease in PCV ($P<0.0001$), Hb ($P<0.01$) and RBC ($P<0.0001$) with increase in duration of feeding while white blood cells, eosinophils and monocytes were not affected by duration of feeding. There was significantly ($P<0.05$) higher PCV, haemoglobin (Hb), WBC, lymphocyte, neutrophils, band, total protein and globulins at 30% inclusion of the cottonseeds than 10 and 20% inclusion levels. There was significant interaction between cotton products and their inclusion level on lymphocyte, neutrophils and band. It is concluded that up to 30% cottonseed cake and 10% whole cottonseed can be fed to bucks without adverse effects.

Keywords: Bucks, cottonseed, heamatology, serum chemistry, red Sokoto goats.

INTRODUCTION

The goat is an economically important specie in rural areas. Goats are able to tolerate high rainfall areas and other harsh conditions associated with the semi-arid climate. Goats can efficiently ingest low quality feedstuff especially fibrous materials; they are favorite animal in rural area of most sub-tropical countries (Green and Baker, 1996; Adams *et al*., 1997). These low quality feedstuffs are low in protein. Protein is an expensive but essential nutrient for animal growth and performance (Dabiri and Thonney, 2004). Different protein sources have varying effects on ruminants' blood metabolites, and serum biochemistry (Jorgensen *et al*., 1984).

Gossypol is a phenolic yellow pigment produced by the pigment glands found in cotton roots, branches, leaves and seeds (Cheeke, 1998; Rogers *et al*., 2002) that confers resistance of the plant to pathogens (Abdurakhimov *et al*., 2009). Heat treatment of cottonseed promotes binding of gossypol to proteins, probably in the region of the radical ε-amine of lysine (Calhoun *et al*., 1995). Gossypol naturally exists in either a free form or bound to proteins, with the former being

most common in cottonseeds. Bound gossypol is considered to be physiologically inactive (Randel *et al*., 1992) and goats and sheep fed with cottonseed treated in this way have shown no clinical signs of poisoning and no changes to their sperm quality (Guedes *et al*., 2010; Nunes *et al*., 2010).

Ruminants with a well-developed microbial rumen population are able to detoxify gossypol by binding it to amino acids in the rumen (Santos *et al*., 2002). However, the detoxifying mechanism can be overwhelmed when excessive gossypol content is combined to a low protein diet in the rumen (Risco *et al*., 1998). Ruminants from 8 to 24 weeks old can tolerate up to 200 mg/kg of dietary free gossypol and older animals can tolerate up to 600 mg/kg without adverse effects (Risco *et al*., 1998). The effect of gossypol is cumulative and may appear suddenly as result of prolonged periods of ingestion (Cheeke, 1998). Signs of toxicosis include decreased growth rate, anorexia, laboured breathing, and dyspnea [Randel *et al*., 1992].

The life of all flesh is the blood and its usefulness for assessing the health status, chemical evaluation for survey, physiological pathological conditions and diagnostic and prognostic evaluation of various types of diseases in animals cannot be over emphasize (Tambuwal *et al*., 2002; Alade *et al*., 2005). It also helps in distinguishing normal state from state of stress, which could be nutritional, environmental or physical (Aderemi, 2004). Haematological values are widely used to determine systematic relationship and physiological adaptation including the assessment of general health condition of animals (Kamal *et al*., 2007). The changes in these parameters have been studied in cattle (Ghergariu *et al*., 1984) sheep (Kausslish and Arora, 1977) and Red Sokoto goat (Tambuwal *et al*., 2002).

Dietary gossypol from cottonseed products has been shown to cause haematological changes in ruminants (Lindsey *et al* 1980). Increase in erythrocyte fragility is one haematological change that occurs in animals consuming diets with gossypol. Gossypol toxicosis causes erythrocyte membrane fragility (Gray *et al*., 1993; Mena *et al*., 2004). These haematological changes affect animal health and performance. The effect of gossypol on erythrocyte fragility can be used to measure the safe feeding levels of cottonseed products to animals. Velasquez-Pereira *et al.* (1998) proposed that erythrocyte fragility can be used as a sensitive indicator of systemic gossypol status. Mena *et al*. (2001) argued that the detoxification in the rumen is reduced with increased ruminal passage rate of foods with gossypol. According to Braga *et al*. (2012) residual gossypol present in extruded-expelled cottonseed meal can affect the leukocyte numeration, particularly lymphocyte counts in sheep, which may compromise immune-competence. Gossypol causes cytotoxic effects and apoptosis of lymphocytes (Yurtcu *et al*., 2003; Xu, *et al*., 2009). This study was designed to evaluate the effect of whole and processed cottonseed on haematology and serum biochemistry of Red Sokoto bucks.

MATERIALS AND METHODS

Location of Experimental Site

The study was carried out at the Teaching and Research Farm, Department of Animal Science, Faculty of Agriculture, Ahmadu Bello University, Zaria, Kaduna state, Nigeria located on latitude $11^0$33'N and longitude $7^0$42'E. Annual rainfall in this area ranges from 1102 to 1904mm and lasts from late April or early May to mid-October. The site is situated within the Northern-Guinea Savannah zone of Nigeria at an altitude of 610 mm above sea level (Ovimaps, 2014).

Experimental Animals and their Management

Twenty-four (24) Red Sokoto goat bucks of about 6-8 months of age were sourced from Zaria metropolis. They were dewormed using albendazole tablets (Centre-Alben®- Aether Centre Biology Co., Ltd. Beijing) at the rate of 5 mg/kg body weight, covered with TLA-Tetracycline long acting-TLA (oxytetracycline dehydrate) 20% weight/volume injectable solution administered at 0.1 mg/10 kg live weight against possible bacterial infection. The bucks were kept separately in individual pens. The animals were quarantined for a period of 3 weeks and a further 2 weeks for adjustment period before commencement of the study.

Experimental Design and diets

A study was conducted to evaluate the effect of feeding cotton products (whole cottonseed and cottonseed cake) and level (10, 20 and 30%) on blood parameters of Red Sokoto bucks in a 2 X 3 factorial arrangement in a completely randomized design. The experimental diets containing 14% CP (Table 1) were fed to the bucks at 3% per kg body weight every morning with clean fresh water provided *ad libitum.*

At the end of the adjustment period, the bucks were randomly allocated to six sub dietary groups of four animals per group. Each group constituted a treatment and each buck within a group was a replicate in a completely randomized design with the treatments consisting of either whole cotton seed (WCS) included at 10, 20 and 30% or cottonseed cake (CSC) included at 10, 20, and 30% levels.

Table 1: Ingredient composition of experimental diets fed to Red Sokoto bucks

	Inclusion levels of whole cottonseed and cottonseed cake (%)					
	WCS			**CSC**		
Ingredients	**10**	**20**	**30**	**10**	**20**	**30**
Maize offal	20	15	10	23	21	19
WCS	10	20	30	-	-	-
CSC	-	-	-	10	20	30
BDG	26	21	16	23	15	7
Rice offal	40	40	40	40	40	40
Molasses	2	2	2	2	2	2
Bone meal	1.5	1.5	1.5	1.5	1.5	1.5
Salt	0.5	0.5	0.5	0.5	0.5	0.5
Total	100	100	100	100	100	100

WCS= Whole Cottonseed, CSC=Cottonseed cake, BDG= Brewers dried grain

Blood Sample Collection and Analysis

Blood samples (5ml) were withdrawn from the jugular vein of the bucks with hypodermic needles and syringes and placed into two sets of labelled sample bottles fortnightly for 98 days. One set of the sample bottles was treated with Ethylene Diamine Tetra Acetate (EDTA) as an anti-coagulant while the other set without EDTA were used to withdraw blood from the jugular vein. Blood samples in the set of bottles without the anti-coagulant were kept in the refrigerator at about 4°C for about 3 hours to coagulate. The samples were later spun using a centrifuge at 3,000 rpm for 10 minutes and the serum separated, collected and stored frozen at -10°C until required for analysis. The sera were used to assay the serum metabolites: total protein, albumin and globulin, concentrations. Plasma albumin concentrations were determined by spectrophotometric procedure based on the dye-binding reaction between albumin and bromocresol green in suitably buffered solution. Total plasma globulins were determined by calculating the difference between total proteins and albumins. Blood samples in the set of bottles containing EDTA were analyzed for haematological parameters: packed cell volume, haemoglobin, red blood cell, white blood cell, lymphocyte count, neutrophils, eosinophils, monocytes, band cells and total protein.

Statistical Analysis

All data generated were subjected to two way Analysis of Variance (ANOVA) using the General Linear Model (GLM) Procedure of Statistical Analysis System (SAS, 2008) and where statistical significance was observed, the mean values were separated using Tukey.

The following Linear Model was used for the statistical analysis of the haematological and semen parameters:

$$Y_{ijk} = \mu + A_i + B_j + (A \times B)_{ij} + e_{ijk}$$

Where:

Y_{ijk} = Observations.

μ = Overall mean

A_i = Effect of cotton products (whole cottonseed and cottonseed cake)

B_j = Effect of inclusion levels

C_k= Effect of bi-weekly collection periods

$(A \times B)$ = Interaction between cotton products and inclusion levels.

e_{ijk} = Experimental error

RESULTS

Table 2 shows the effect of whole cottonseed and cotton seed cake on haematology and serum biochemistry of bucks. There were no significant ($P>0.05$) differences in most haematological parameters and serum chemistry of bucks fed diets containing either whole cottonseed or cottonseed cake. However, PCV and RBC were significantly ($P<0.01$) higher in bucks fed CSC diet than those fed WCS diet. Similarly, albumin levels were significantly ($P<0.05$) higher in bucks fed CSC than those fed WCS.

The effect of inclusion level of cottonseed products on haematology and serum biochemistry of bucks is shown in Table 3. Apart from WBC, eosinophils, monocytes and albumin that were similar, level of inclusion had significant (P<0.05) effect on every other haematological parameter. Bucks fed 30% inclusion level of cottonseed had significant (P<0.05) higher PCV, RBC, Hb, neutrophils, band values than those on 10 and 20% inclusion levels. Lymphocyte count increased slightly from 63.83 (10%) to 64.25 (20%) and then it dropped sharply to 54.94 with 30% level of inclusion. Neutrophils were similar for bucks fed 10 and 20% inclusion of the cotton products. However, there was significant (P<0.01) increase in the blood neutrophils (39.64%) when the cotton products inclusion level was increased from 20 to 30%. Band cells differed significantly (P<0.0001) across levels following similar pattern as neutrophils. Total proteins (P<0.05) and globulin (P<0.01) levels increased significantly with increasing inclusion of the cotton products.

Table 2: Effect of whole cottonseed and cottonseed cake on haematology and serum biochemistry of Red Sokoto bucks

Parameters	WCS	CSC	SEM	P-value
PCV (%)	28.33^{b}	31.93^{a}	1.44	0.0029
Hb (g/dl)	9.77	10.56	0.64	0.1379
RBC ($x10^{6}$/l)	4.70^{b}	5.36^{a}	0.24	0.0011
WBC ($x10^{9}$)	6.43	6.17	0.66	0.6292
Lymphocyte (%)	58.89	63.13	2.82	0.0693
Neutrophils (%)	36.57	32.78	2.65	0.0836
Eosinophils (%)	0.41	0.20	0.86	0.1252
Monocytes (%)	3.11	2.96	1.98	0.8208
Band (%)	1.00	0.81	0.31	0.46
TP (g/dl)	6.74	6.97	0.19	0.1367
Albumin (g/dl)	3.51^{b}	3.74^{a}	0.13	0.0331
Globulin (g/dl)	3.22	3.23	0.17	0.9790

SEM: Standard error of means; LOS: Level of significance Means within rows with different superscripts are significantly different (P<0.05; P<0.01). WCS: Whole cottonseed CSC: Cottonseed cake. PCV: Packed cell volume; Hb: Haemoglobin; RBC: Red blood cells; WBC: White blood cells; Band: Band neutrophils; TP: Total proteins.

Table 3: Effect of inclusion levels of WCS and CSC on haematological and serum biochemical parameters of Red Sokoto bucks

	Inclusion level of cottonseed in diets (%)				
Parameters	**10**	**20**	**30**	**SEM**	**P-value**
PCV (%)	29.47^{b}	28.72^{b}	32.19^{a}	1.44	0.0442
Hb (g/dl)	9.76^{b}	9.47^{b}	11.26^{a}	0.64	0.0136
RBC ($x10^{6}$/l)	4.85^{b}	4.76^{b}	5.49^{a}	0.24	0.0057
WBC ($x10^{9}$)	6.70	6.23	5.96	0.66	0.5283
Lymphocyte (%)	63.83^{a}	64.25^{a}	54.94^{b}	2.82	0.0016
Neutrophils (%)	32.47^{b}	31.92^{b}	39.64^{a}	2.65	0.0071
Eosinophils (%)	0.19	0.57	0.17	0.86	0.0591
Monocytes (%)	2.73	2.81	3.58	1.98	0.5056
Band (%)	0.69^{b}	0.33^{b}	1.69^{a}	0.31	<.0001
TP (g/dl)	6.59^{ab}	6.92^{a}	7.05^{a}	0.19	0.0443
Albumin (g/dl)	3.69	3.56	3.62	0.13	0.6062
Globulin (g/dl)	2.90^{b}	3.36^{a}	3.43^{a}	0.17	0.0052

Means within rows with different superscripts are significantly different (P<0.05; P<0.01; P<0.001). SEM: Standard error of means; LOS: Level of significance; NS: Not significant. PCV: Packed cell volume; Hb: Haemoglobin; RBC: Red blood cells; WBC: White blood cells; Band: Band neutrophils; TP: Total proteins; WCS: Whole cottonseed; CSC: Cottonseed cake.

There was significant decrease in PCV (P<0.0001), Hb (P<0.01) and RBC (P<0.0001) with increase in duration of feeding (Table 4). White blood cells, eosinophils and monocytes were not affected by duration of feeding. An increase in lymphocyte count was observed from weeks 2 to 6 after which there was a decrease in week 8. At week 10, the lymphocyte count increased again and dropped finally in week 12, hence significant differences. Band neutrophils generally increased significantly (P<0.01) from week 2 to 8 but declined at week 10 and increased again towards the 12^{th} week.

The interaction between cotton products and level of inclusion on haematology and serum chemistry of the bucks is presented in Table 5. The result shows significant interaction (P<0.05) on lymphocytes, neutrophils and band cells. The PCV, Hb and RBC values were generally higher in bucks fed CSC at different levels than those fed WCS. The values however, increased with increase in the levels of the WCS as well as CSC in the diets, although this increase was not significant. Within treatment means, bucks fed CSC had significantly (P<0.05) higher lymphocyte counts (67.44, 62.33, 59.61) than those fed WCS (60.22, 66.17, 50.28), although both groups had decreasing lymphocyte counts as the inclusion levels increased. The CSC group

had significantly lower (P<0.05) neutrophils but higher total protein, albumin and globulins which did not differ significantly (P>0.05).

Table 4: Effect of duration of feeding on haematological and serum biochemical parameters of Red Sokoto bucks fed different levels of WCS and CSC

Parameters	Duration of feeding (Weeks)							
	2	4	6	8	10	12	SEM	P-value
PCV %	33.28^{a}	34.28^{a}	31.67^{ab}	29.06^{b}	25.56^{cd}	26.94^{bc}	1.44	0.0001
Hb (g/dl)	12.18^{a}	11.41^{ab}	10.41^{bc}	9.57^{cd}	8.48^{d}	8.94^{d}	0.64	0.0005
RBC (x10^{6}/l)	5.62^{ab}	5.78^{a}	5.21^{b}	4.71^{c}	4.31^{c}	4.55^{c}	0.24	0.0001
WBC	7.26	5.46	5.53	6.64	5.78	7.13	1.59	0.1891
Lymphocyte (%)	58.67^{cd}	62.56^{bc}	65.28^{ab}	57.06^{de}	70.83^{a}	51.67^{e}	2.82	0.0002
Neutrophils (%)	39.17^{ab}	34.28^{bc}	30.11^{c}	37.78a^{b}	24.56^{d}	42.17^{a}	2.65	0.0001
Eosinophils (%)	0.00	0.33	0.56	0.50	0.44	0.62	0.35	0.7854
Monocytes (%)	1.98	2.17	3.51	2.89	3.44	3.54	0.81	0.2159
Band (%)	0.39^{c}	0.56^{c}	0.78b^{c}	1.94^{a}	0.72^{c}	1.06^{ab}	0.31	0.0101
TP (g/dl)	6.57^{bc}	6.90^{ab}	6.76^{bc}	6.47^{c}	7.17^{a}	7.24^{a}	0.19	0.0229
Albumin (g/dl)	3.66^{bc}	4.01^{a}	3.86^{ab}	3.39^{d}	3.30^{d}	3.52^{cd}	0.13	0.0009
Globulin (g/dl)	2.91^{b}	2.89^{b}	2.89^{b}	3.09^{b}	3.87^{a}	3.72^{a}	0.17	0.0001

Means within rows with different superscripts are significantly different (P<0.05; P<0.01; P<0.001). SEM: Standard error of means; LOS: Level of significance; NS: Not significant; PCV: Packed cell volume; Hb: Haemoglobin; RBC: Red blood cells; WBC: White blood cells; Band: Band neutrophils; TP: Total proteins.

Table 5: Interaction between WCS and CSC and inclusion levels on haematological and serum biochemical parameters of Red Sokoto bucks

Cottonseed product	Whole cottonseed			Cottonseed cake				
Level of inclusion	10	20	30	10	20	30	SEM	P-value
PCV (%)	28.56	26.78	29.67	30.39	30.67	34.72	3.98	0.5267
Hb (g/dl)	9.48	8.86	10.98	10.03	10.08	11.56	1.51	0.8386
RBC (x10^{6})	4.57	4.46	5.09	5.13	5.07	5.88	0.79	0.8776
WBC (x10^{9})	7.09	6.34	5.86	6.31	6.13	6.08	0.66	0.7466
Lymphocyte (%)	60.22^{a}	66.17^{a}	50.28^{c}	67.44^{a}	62.33^{a}	59.61^{ab}	3.63	0.0485
Neutrophils (%)	34.39^{b}	30.28^{b}	45.06^{a}	30.56^{b}	33.56^{b}	34.22^{b}	3.75	0.0338
Eosinophils (%)	0.28	0.74	0.22	0.11	0.39	0.11	0.86	0.3257
Monocytes (%)	3.67	2.72	2.94	1.78	2.89	4.22	1.98	0.1442
Band (%)	1.28^{a}	0.22^{b}	1.50^{a}	0.11^{b}	0.44^{b}	1.89^{a}	0.31	0.0255
TP (g/dl)	6.54	6.76	6.91	6.63	7.08	7.19	0.23	0.8037
Albumin(g/dl)	3.67	3.40	3.46	3.71	3.72	3.77	0.20	0.4716
Globulin(g/dl)	2.88	3.35	3.45	2.92	3.36	3.42	0.31	0.2648

Means in the same row within cottonseed type with different superscripts are significantly different (P<0.05)
SEM: Standard error of means; LOS: Level of significance. PCV: Packed cell volume; Hb: Haemoglobin; RBC: Red blood cells; WBC: White blood cells; Band: Band neutrophils; TP: Total proteins

PCV and Hb content decreased as duration of feeding increased for all cottonseed inclusion levels (Fig. 1). PCV was slightly higher in bucks fed 30% inclusion level than in those fed 10 and 20%. However, after the sixth week of feeding there was general decrease in PCV levels across inclusion levels. Hb level (Fig. 1) and RBC count (Fig. 2) levels were also slightly higher in bucks fed 30% inclusion than those fed 10 and 20% cottonseed. However, there was a general decrease in both Hb levels and RBC count with increase in duration of feeding. WBC count (Fig. 2) increased with duration of feeding being higher in bucks on 10% inclusion than 20 and 30% inclusion levels. Lymphocytes count did not appear to be adversely affected by duration of feeding (Fig. 3) but was higher on 20% inclusion of cottonseed while there were great changes in neutrophils on the other hand, which increased after an initial decrease between weeks 2 and 4, but decreased drastically from week 8 to 10 and thereafter increased sharply, being higher on 30% cottonseed than 10 and 20% levels. Monocytes counts (Fig. 4) increased with duration of feeding. There was a sharp increase in eosinophils in bucks receiving 20 and 30% of the cotton products from week 2 to week 6 after which it began to decrease until the 10th week when the levels rose again. However, bucks that received 10% inclusion of cottonseed had a slower but steady increase of eosinophils from the beginning up until the end of the study.

Blood total proteins (Fig. 5) increased for all levels of cottonseed inclusion as feeding duration increased while albumin decreased with duration of feeding. Albumin level decreased slightly but progressively in bucks fed 20 and 30% inclusion levels while in those fed 10% inclusion, albumin increased until the 6th week after which it decreased by the 8th week and increased thereafter. Globulins (Fig. 6) generally decreased slightly and remained constant until between the 6th and 8th week when it began to increase again. In bucks fed 30% inclusion of cottonseed, globulin levels increased between the 2nd and 4th week but decreased on 10% inclusion level, remaining constant between weeks 4 and 6 then declined between weeks 6 and 8, and increased thereafter. However, in bucks fed 20% inclusion, globulin levels were constant during the first 8 weeks but increased thereafter.

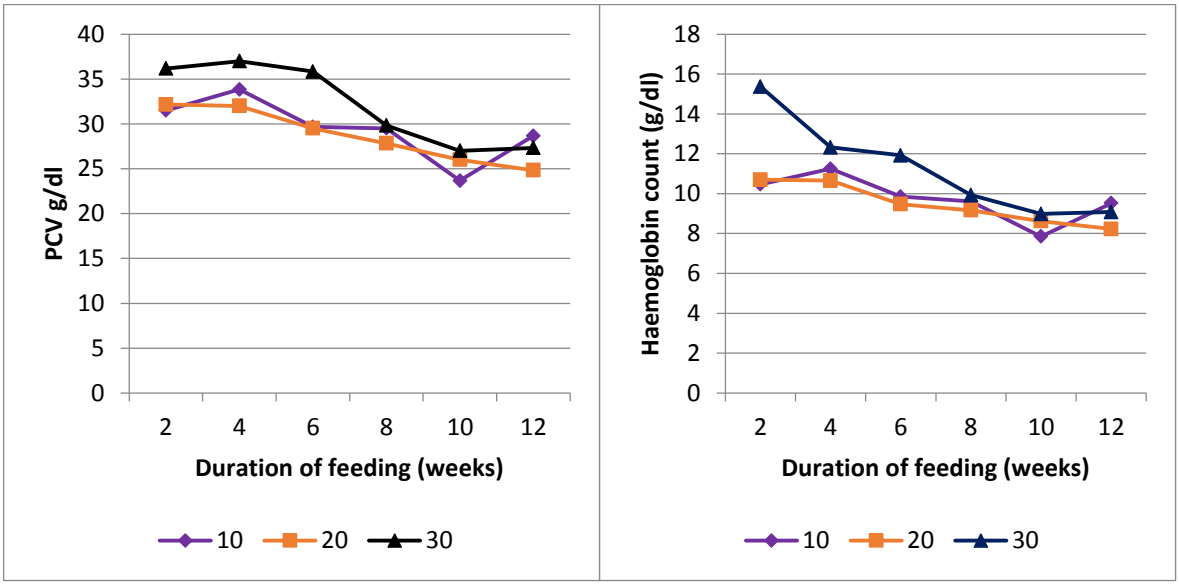


Figure 1: Effect of feeding duration of cotton seed on PCV and Haemoglobin content of Red Sokoto Bucks fed varying levels of cotton seed

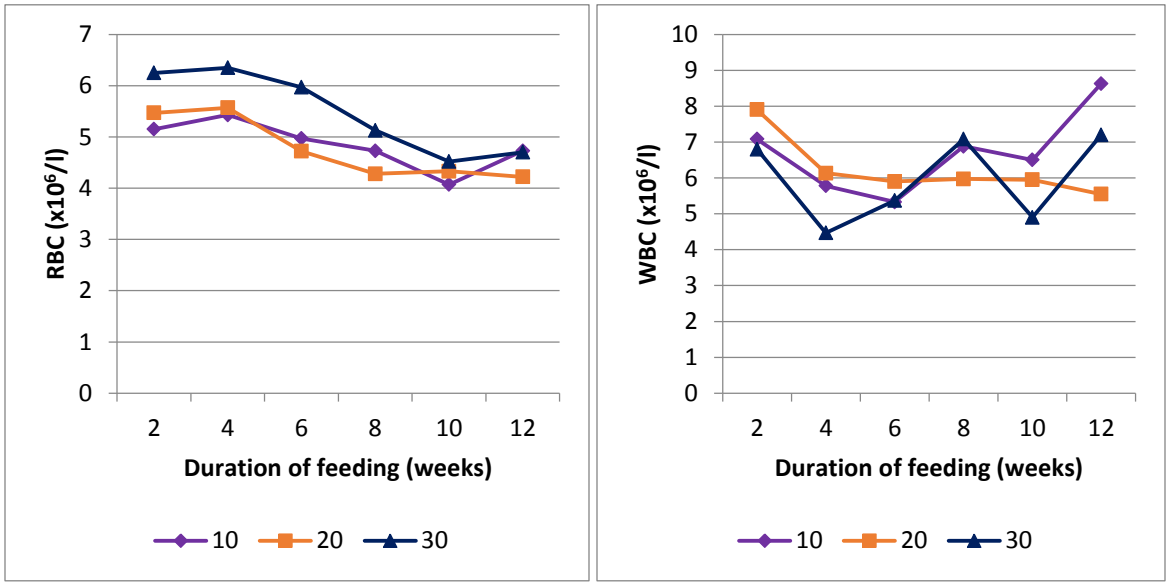


Figure 2: Effect of Feeding duration on RBC and White blood cells of Red Sokoto Bucks fed varying levels of cotton seed.

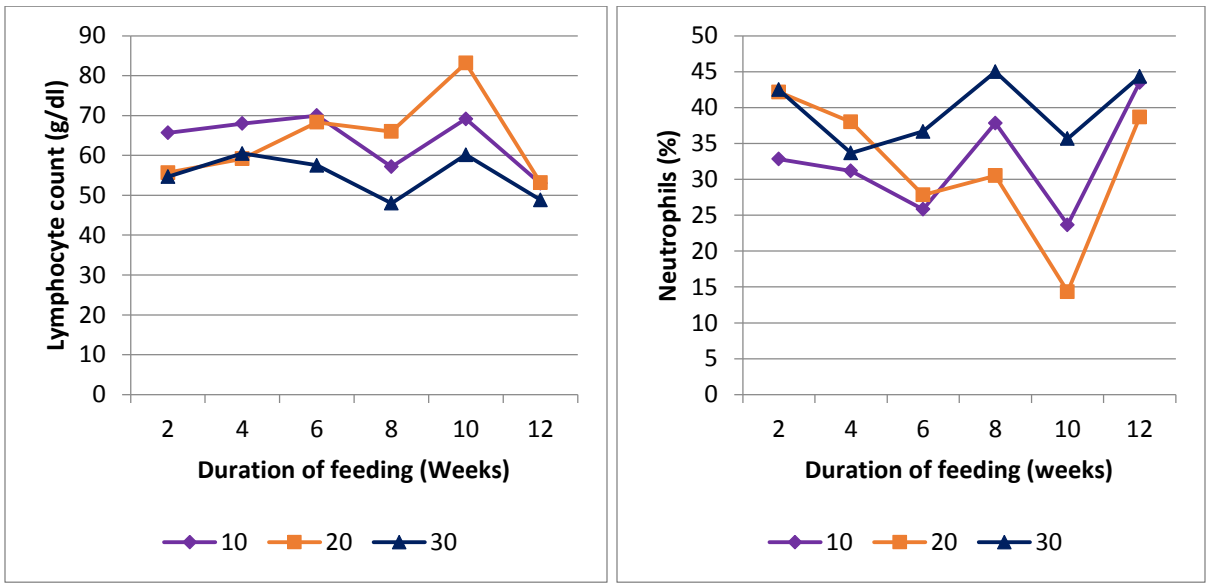


Figure 3: Effect of period on Lymphocyte and Neutrophils of Red Sokoto Bucks fed varying levels of cotton seed.

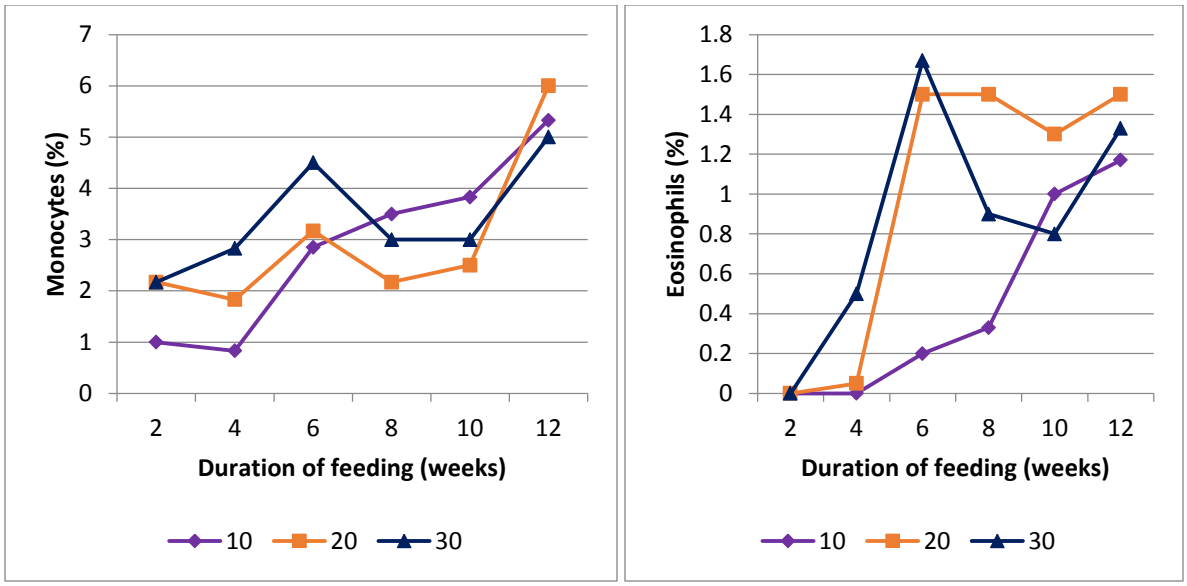


Figure 4: Effect of duration of feeding on Monocytes and Eosinophils of Red Sokoto Bucks fed varying levels of cotton seed.

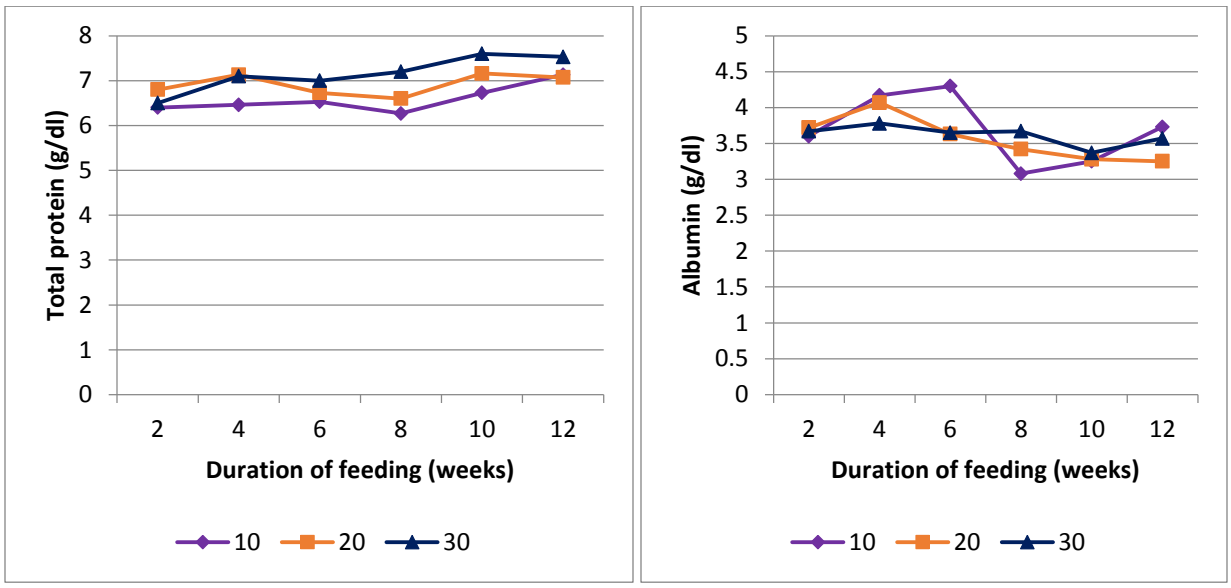


Figure 5: Effect of Duration of feeding on blood Total protein and Albumin content of Red Sokoto Bucks fed varying levels of cotton seed.

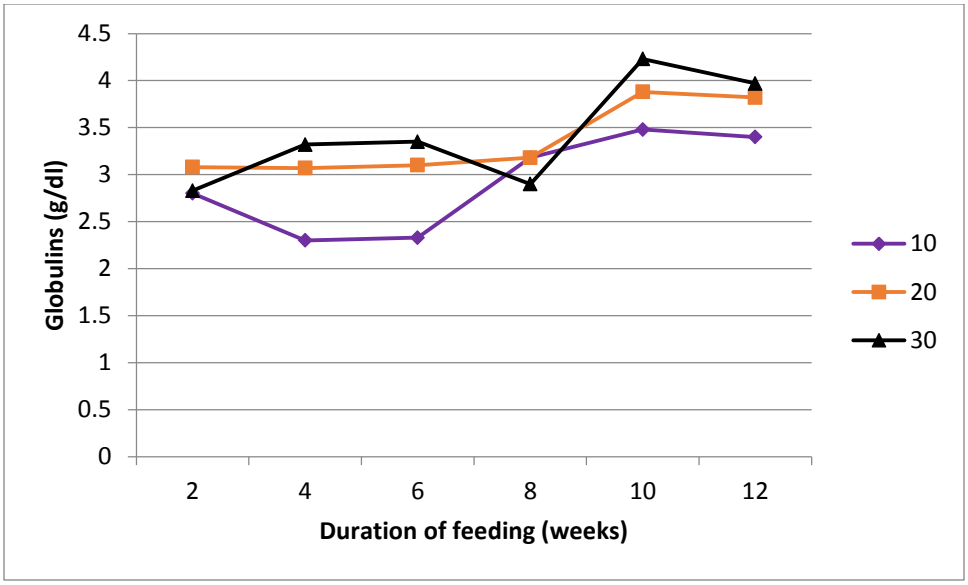


Figure 6: Effect of duration of feeding on globulins values of Red Sokoto Bucks fed varying levels of cotton seed.

DISCUSSION

The effect of whole cottonseed and cottonseed cake on haematological and serum biochemical parameters of Red Sokoto bucks studied shows that packed cell volume (PCV), red blood cell (RBC) and serum albumin (Alb) were significantly ($P<0.01$) higher in bucks fed CSC than those fed WCS. Haemoglobin (Hb) was not significantly ($P>0.05$) affected but increasingly higher among the CSC fed bucks than their WCS counterparts. This agrees with Barje *et al*. (2008) who reported slightly lower PCV and RBC values in calves fed whole cottonseed at higher levels (50 and 75%). The higher haematocrit and albumun values recorded in bucks fed CSC can also be attributed to reduced anti-nutritional factors as Akinmutimi, (2004) reported that level of anti-nutritional element or factors present in the feed influences hematological and biochemical values. The significant ($P<0.05$) increase in PCV, Hb, RBC, neutophils, band cells, total proteins and globulins with increase in the levels of WCS and CSC in the diets indicates some level of response to stress. Considerable evidence points to oxidative stress, formation of reactive oxygen species (Kovacic, 2003) causing the animals to have increased PCV, haemoglobin concentration in order to balance their oxygen carrying capacity. The decrease in lymphocyte counts with increase in the level of cotton products agrees with the report of Braga et al. (2012) who tested the effects of gossypol on lymphocyte count of sheep and reported a decrease in lymphocyte counts as the gossypol level increased. Gossypol present in whole cottonseed as well as residual gossypol in extruded-expelled cottonseed meal can affect the leukocyte numeration, particularly lymphocyte counts, by being cytotoxic and apoptotic which may compromise immune-competence (Yurtcu *et al*., 2003; Xu, *et al*., 2009). The insignificant difference in serum albumin levels could have been due to the fact that serum albumin concentration were not affected by the levels of WCS and CSC in the diet. This agrees with Solaiman *et al*. (2009) who reported no significant difference ($P>0.05$) in serum albumin, but there were significantly higher total protein and globulins among the treatments as a result of increased protein intake.

The effect of duration of feeding on haematological and serum biochemical parameters of Red Sokoto bucks indicates that PCV, Hb as well as WBC decreased significantly ($P<0.05$)with increase in feeding period this is probably related to changes in weather conditions such as ambient temperature, relative humidity and solar radiation. The effect of gossypol is cumulative and may appear suddenly as a result of prolonged periods of ingestion (Cheeke, 1998). This is in agreement with the observations of Koubkova *et al*. (2002) who reported changes in values of these parameters as a result of seasonal changes and variations in environmental temperatures. Jabbar *et al*. (2012) observed that high erythrocyte count could be responsible for increased PCV value in heifers. Hb concentration is especially indicative of adaptation to environmental conditions. Thus, blood haemoglobin concentration was depressed and it fell to 8.48 g/dl on the penultimate collection. It is possible, that further feeding of the diets could have depressed the haemoglobin levels even more and this is a probable indication of accumulative effect of feeding gossypol.

Periodic decrease in PCV values in this study may be attributed to decrease in environmental temperature (Palterson *et al*., 1960; Isidahomen *et al.,* 2010). The reduced PCV values observed within the period of study, may indicate a decrease in the number of circulating RBC in the goats. This is in line with the report of Kopp and Hetesa (2000) that high PCV haematocrit values indicate either an increase in the number of circulating RBC or reduction in circulating plasma volume. The values of WBC and RBC counts however, were within the ranges reported by Sandra (2007) who feed goats with 15% and 30% WCS and observed a decline in the WBC and RBC when WCS was increased from 15% to 30%. The WBC values are lower than those recorded by Njidda *et al*. (2013) who carried out a study to determine haematological and biochemical parameters of goats fed on natural grazing rangeland in the semi-arid environment of northern Nigeria. The lower values obtained in this study suggest underdeveloped immune system of the goats. This difference might also be due to management practices as the bucks in this present study were subjected to intensive system of management rather than the extensive system of rearing amenable to goats. It was reported that, like other ruminants goats have more lymphocytes than neutrophils in circulation (Olusanya *et al*., 1976). This could account for the significant ($P<0.05$) difference in lymphocyte, neutrophils and band cell recorded as time went on. The observed increases in total proteins and globulins with time agrees with that of Nikokyris *et al*. (1991) who reported increases in plasma protein and urea over time. These higher concentrations of plasma protein may be due to increased quantity of ammonia absorbed from the rumen and circulating in the plasma because of increased feed consumption. Risco *et al*. (1993) stated that the changes observed in haematological and serum composition of cows fed diets with gossypol have not been enough to be used as a diagnosis of toxicity.

The generally low lymphocyte values observed in this study are within the range (50.28 to 67.44%) reported by Opara *et al.* (2010) for goat kids but higher than those reported by Tariq Javed *et al*. (2010) ranging from 43.89% in young goats to 45.86% for adult goats. Lymphocytes are the key elements in the production of immunity, therefore at low levels, the animals are prone to bacterial infections, aplastic anaemia, and some forms of leukemia while high values can be observed in viral infections, and in some forms of leukemia (Ganong, 2005). The decline in neutrophils with increase in either WCS or CSC could be an indication of gossypol toxicity and consequent apoptosis of lymphocytes. Gossypol content of WCS varies among varieties and high levels have been found to cause fragility of the RBC (Calhoun *et al*., 1990a). According to Greenhalg *et al*. (1972), concentration of blood hemoglobin <80g/L (8g/dl) in cattle indicates anemia.

The profile of blood serum proteins such as albumin, globulin, total protein was not affected by cotton products and their inclusion levels. Likewise, Mena *et al*. (2004) reported that the serum concentration of albumin was not influenced by diets. In contrast to the present findings, Calhoun *et al*. (1990a) and Risco *et al*. (1992) reported a decrease in the serum albumin levels as the level of gossypol increased in the diet. The increases in plasma proteins and globulin levels at higher WCS and CSC in the diets agrees with the report of Solaiman *et al*. (2009) who reported that blood serum protein increased linearly ($P<0.05$) as the dietary cottonseed level increased in the diets of goats. This is probably due to increased levels of the cotton products as they are rich sources of proteins. Chineke *et al*. (2006) reported that increased protein intake leads to increased total proteins and globulins. On the other hand, Calhoun *et al*. (1990b) indicated that the total protein level decreased as the levels of gossypol increased in the diets of lambs receiving WCS-supplemented diets compared to goats fed a control diet. Likewise, Andreazzi *et al*. (1995) did not detect significant difference between the treatments for the blood metabolites in goats fed low levels of gossypol.

CONCLUSION

Feeding of increased levels of whole cottonseed and cottonseed cake to Red Sokoto bucks might have induced lymphocyte apoptosis as the lymphocyte counts of the bucks decreased with increased levels of the cotton products in the diets. Most haematological parameters were higher in animals fed CSC than in those fed WCS as well as with increased levels of the cotton products. It is concluded that up to 30% cottonseed cake and 10% whole cottonseed can be fed to bucks without adverse effects.

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