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BLOOD PROFILE OF WEST AFRICAN DWARF GOATS FED PROVITAMIN A CASSAVA PEEL-CENTROSEMA LEAF MEAL BASED DIETS

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Abstract: The study was conducted to investigate the effects of feeding varying levels of provitamin A cassava peel-centrosema leaf meal at 0%, 10%, 20% and 30%, respectively as supplement to *Panicum maximum*, on the haematological and serum biochemical parameters of West African Dwarf (WAD) goats. The study lasted for 97 days during which haematological and serum biochemical parameters were evaluated in 36 bucks, which were randomly divided into four (4) groups of 9 goats each, with three goats constituting a replicate in a completely randomized design pattern. Blood samples were drawn from each animal on the last day of the trial and evaluated for haematological and biochemical profile analyzed statistically. The packed cell volume (PCV), haemoglobin (Hb), mean corpuscular haemoglobin concentration (MCHC) and mean corpuscular volume (MCV) significantly ($P<0.05$) ranged from 28.87% to 32.91%, 10.77 to 13.23 g/dl, 31.05 to 34.25% and 19.04 to 19.04 fl respectively, while red blood cell (RBC) and mean corpuscular haemoglobin (MCH) were significantly ($P>0.01$) similar and ranged from 10.97 to 11.01 $x10^6$/ul and 6.13 to 6.34 pg respectively. White blood cells and the differentials increased significantly ($P<0.05$) with increasing levels of the test ingredients. Total protein, albumin and globulin significantly ($P<0.05$) ranged from 66.21 to 73.32 g/l, 32.20 to 34.33 g/l and 34.01 to 38.99 g/l respectively. Serum creatinine increased significantly ($P<0.05$) as the level of test ingredient increased while serum urea and AST decreased significantly ($P<0.05$) as the levels of the test ingredient increased from A to D. Cholesterol however showed significant difference ($P<0.05$) but did not follow any specific trend with increasing or decreasing levels of provitamin A cassava peel-centrosema leaf meal. The study revealed that provitamin A cassava peel-centrosema leaf meal in the diets of West African Dwarf goats had no deleterious effects on the haematological and serum biochemical parameters of WAD goats and could therefore be included in ruminant diets up to 30%.

INTRODUCTION

The provitamin A cassava have been developed either through traditional plant-breeding or bioengineering (Sayre *et al*., 2011). The new provitamin A cassava varieties have potential of providing up to 25% of daily Vitamin A requirements of children and women (Aniedu and Omodamiro, 2012). Cassava with higher levels of provitamin A can help reduce vitamin A deficiency among undernourished communities that rely upon cassava for sustenance. Provitamin A cassava varieties are very high in carotenoids. More recently, protective effects of carotenoids against serious disorders such as cancer (Donaldson, 2004), heart disease (Sesso, 2003) and degenerative eye disease (Mozaffarieh *et al*., 2003) have been recognized, and have stimulated intensive research into the role of carotenoids as antioxidants and as regulators of the immune response system. These numerous advantages has led to high demand for the provitamin A cassava for different human or industrial uses thereby enhancing the availability of the peels which are grossly underutilized and were hitherto discarded as waste. The provitamin A cassava peels are made up of mainly polysaccharides and carotenoids; hence holds inestimable potentials as energy and vitamin A sources for goats. However, due to its crude protein deficiency, the need to supplement with readily available noncompetitive unconventional protein source like *Centrosema pubescens* leaf meal becomes imperative. Nworgu and Egbunike (2013) reported 23.24% Crude protein in *Centrosema pubescens* leaf meal; hence being a potential protein source in goat production.

Evaluation of the blood profile of animals may give some insight as to the potentials of a dietary treatment to meet the metabolic needs of the animal. Church *et al*. (1984) noted that dietary components have measurable effects on blood constituents such that significant changes in their values can be used to draw inference on the nutritive value of feeds offered to the animals. Haematological and serum biochemical studies are widely used for the diagnosis of animal diseases as well as investigation of blood damage and feed toxicity and protein quality (Jiwuba *et al*., 2016). Blood profile studies are important since blood is the major transport system of the body and evaluations of the blood profile usually furnish vital information on the body’s response to injury of all forms, feed quality and toxicity, the authors concluded. However, the assertion of Ikhimoya and Imasuen (2007) that most of the available information on haematological parameters of goats in

the humid tropics is based on disease prognosis from the region. Thus, data on haematological and serum biochemical profile of West African dwarf (WAD) goat offered agricultural biotechnological byproduct and leaf meal from non-conventional sources are scanty.

The sourcing for readily, locally available and nutritionally viable feed ingredients to enhance food production stimulated this research which aimed at evaluating the efficacy of provitamin A root cassava peel – centrosema leaf meal based diets on growth and haematological and serum biochemical parameters of West African dwarf goats.

MATERIALS AND METHODS

The experiment was carried out at the sheep and goat Unit, Federal College of Agriculture, Ishiagu, Ivo L.G.A., Ebonyi state, Nigeria. The College is located at about three kilometers (3km) away from Ishiagu main town. The College is situated at latitude 5.56^0N and longitude 7.31^0E, with an average rainfall of 1653 mm and a prevailing temperature condition of 28.50^0c and relative humidity of about 80%. Fresh provitamin A cassava peels varieties (TMS011368, TMS011412 and TMS1371) were obtained from National Root Crops Research Institute, Umudike, Abia State, Nigeria. The peels were subsequently dried to about 10% moisture content before milling and used in the formulation of provitamin A cassava peel - centrosema leaf meal based diets. Fresh green *Centrosema pubescens* leaves were harvested within the College. The *Centrosema pubescens* were shade-dried in batches, milled and also used at different levels in the formulation of provitamin A cassava peel - centrosema leaf meal based diets.

Thirty six (36) WAD goats of about 8 – 10 months of age and averaging 7.19kg in weight were selected from the College herd for this experiment. The goats were randomly divided into four (4) groups of nine (9) animals each with 3 goats constituting a replicate. The groups were randomly assigned the 4 experimental diets (A, B, C and D) in a completely randomized design (CRD). The animals were housed individually in well ventilated cement floored pens equipped with feeders and drinkers. Each animal received a designated treatment diet in the morning for 97 days. Feed offered was based on 3.5% body weight per day; the animals in addition were fed 2kg wilted *Panicum maximum* later in the day. Regular access to fresh drinking water was made available.

Experimental diets designated as A, B, C and Dwere formulated from provitamin A cassava peel, brewers dried grain, palm kernel meal, wheat offal, *Centrosema pubescens* leaf meal, bone meal, molasses and salt. Diet A served as a positive control and contained 0% of *Centrosema pubescens* leaf meal. Diets B, C and D contain 10%, 20% and 30% inclusion levels *Centrosema pubescens* leaf meal respectively as illustrated in Table 1. All feeds and test ingredients were analyzed for proximate compositions using the method of AOAC (2000). Gross energy was determined according to Nehring and Haelein (1973).

Table 1: Percentage Composition of the provitamin A cassava peel-centrosema leaf meal based diets

	Dietary levels (%)			
Ingredients (%)	**A**	**B**	**C**	**D**
Provitamin A Cassava peel	40.00	40.00	40.00	40.00
Centrosema pubescens leaf meal	0.00	10.00	20.00	30.00
Brewers dried grain	38.00	28.00	18.00	8.00
Palm kernel cake	18.00	18.00	18.00	18.00
Bone meal	2.00	2.00	2.00	2.00
Molasses	1.50	1.50	1.50	1.50
Common salt	0.50	0.50	0.50	0.50
Total	100	100	100	100

Five ml of blood samples were drawn from each animal on the last day of the study. The goats were bled through the jugular vein. The samples were separated into two lots and used for haematological and biochemical determinations. An initial 2.5 ml was collected from each sample in labelled sterile universal bottle containing 1.0 mg/ml ethyldiamine tetracetic acid (EDTA) and used for haematological analysis. Another 2.5 ml was collected over anti-coagulant free bottle and used for the serum biochemical studies. Serum biochemistry and haematological parameters were measured using Beckman Coulter Ac-T10 Laboratory Haematology Blood Analyzer and Bayer DCA 2000+ HbA1c analyzer, respectively. Mean corpuscular haemoglobin (MCH), mean corpuscular volume (MCV) and mean corpuscular haemoglobin concentrations (MCHC) were calculated.

The results were analyzed using the Special Package for Social Sciences Window 17.0. One -way analysis of variance (ANOVA) was employed to determine the means and standard error. Treatment means were compared using Duncan's new multiple range test (Duncan, 1955).

RESULTS AND DISCUSSION

The chemical compositions of the experimental diets, provitamin A cassava peel meal (PCPM) and *Centrosema pubescens* leaf meal (CPLM) used in this study is presented in table 2. The proximate

analysis revealed the presence of dry matter, crude protein, crude fibre, ether extract, nitrogen extract and ash. The proximate values for the provitamin A cassava peel meal showed a higher crude protein (CP), ash and ether extract (EE) and lower crude fibre (CF) values compared to the reports of Ahamefule *et al*. (2005) for cassava peel meal. The differences could be attributed to the improvements that have been carried on the yellow root cassava. The proximate compositions of the *centrosema pubescens* leaf meal in this study are comparable with the findings of Nworgu and Egbunike (2013) for the same leaf meal. The dry matter levels of the test diets (B, C and D) compared favourably well with the control diet (A). The CP, ash and ether extract of the test diets were higher than the control diet and tended to increase with the increasing levels of CPLM in the diets. The fibre content on the other hand is higher in the control diet and tended to decrease with increasing levels of CPLM. The nitrogen free extract and gross energy did not show any specific trend among the diets.

Table 2: The chemical compositions of yellow root cassava peel, Centrosema leaf meal and provitamin A cassava peel-centrosema leaf meal based diets

	Dietary levels (%)					
Parameters	**A**	**B**	**C**	**D**	**PCPM**	**CPLM**
Dry matter (%)	91.32	91.54	91.37	91.74	90.28	86.03
Crude protein (%)	12.11	12.78	13.41	14.32	9.23	20.44
Crude fibre (%)	14.36	14.65	13.13	13.07	12.93	10.32
Ether extract (%)	4.83	5.11	5.97	5.99	3.41	2.09
Ash (%)	9.01	9.63	10.17	10.19	9.74	6.95
Nitrogen free extract (%)	51.01	49.37	48.69	48.17	55.67	44.22
Gross energy	3.90	3.92	3.91	3.95	3.71	3.64

PCPM = Provitamin A cassava peel meal; CPLM = *centrosema pubescens* leaf meal

The haematology of West African dwarf (WAD) goat fed provitamin A cassava peel-centrosema leaf meal based diets is presented in Table 3. Packed cell volume, haemoglobin, mean cell haemoglobin concentration, mean cell volume, white blood cell neutrophil and lymphocyte differed ($P<0.005$) while red blood cell and eosinophil were similar ($P<0.05$) across the treatment groups. The packed cell volume (PCV) values (%) for the treatment groups fell within the normal range for WAD goats (21-35%), as reported by Daramola *et al*. (2005) and differed significantly ($P<0.05$) among the treatment groups. PCV is generally used as an index of toxicity and its value is influenced by breeds, age and sex. The significant increase in the concentration of PCV in the blood usually would suggest the absence of a toxic factor like haemagglutinin which have adverse effect on blood formation (Jiwuba *et al*., 2016). *Centrosema pubescens* had been reported to contain 20.01mg/kgDM iron (Nworgu and Egbunike, 2013) and provitamin A cassava implicated to contain high level of vitamin A; a fat-soluble vitamin, involved in a number of physiological processes like hematopoiesis (Guimarães *et al*., 2014). It is possible that traces of iron and the vitamin A still abiding in the treatment diets may perhaps have been responsible for the improved values.

The haemoglobin (Hb) value of the treatment groups differed ($P<0.05$) significantly with diet A animals having the lowest (10.77g/dl) and diet D animals the highest value of 13.23g/dl. The Hb concentration compared favourably with the normal range of 7-15g/dl reported by Daramola *et al*. (2005) for WAD goats. The high level of Hb of animals on diet D relative to other treatment groups may imply that the dietary protein was of higher quality, probably due to 30% fortification of the diets with *Centrosema pubescens* leaf meal. This agrees with the observation of Esonu *et al*. (2003)

that one of the possible sources of cheap protein in the diets is the leaf meal of some tropical legumes. Diets containing poor quality protein would usually influence poor transportation of oxygen from the respiratory organs to the peripheral tissues (Robert *et al*., 2000).

Mean cell haemoglobin concentration (MCHC) and mean corpuscular volume (MCV) of the WAD goats fed provitamin A cassava peel-centrosema leaf meal based diets differed significantly ($P<0.05$) while mean cell haemoglobin (MCH) values were similar ($P>0.05$) among the treatment groups. All the values however fell within the reference range for goats as reported by Fraser and Mays (1986). The normal range of MCV, MCHC and MCH recorded in this study for the WAD goats gave a clear indication of the absence of anaemia among the experimental groups. In humans, a close relationship between vitamin A deficiency and anemia has been long recognized (Semba and Bloem, 2002).

White blood cells (WBC) differed significantly ($P<0.05$) among treatment groups. The values obtained in this study (8.82-11.69 $\times10^9$/l)) were within normal range of 6.8-20.1 x10/ul for WAD goats and 4–13$\times10^9$/l as reported by Daramola *et al*. (2005) and Fraser and Mays (1986) respectively. WBC of the goats showed an increasing value in relation to an improvement of the immunological system. Vitamin A has been extensively studied for its influence on immunity due to its requirement for normal functioning of the immune system. Vitamin A deficient people are more prone to more severe infections and have a higher mortality than vitamin sufficient people (Beaton *et al*., 1992).

Lymphocyte and neutrophil differed ($P<0.05$) significantly and however fell within the reported reference range by Fraser and Mays (1986), while eosinophil were similar ($P>0.05$). The circulating lymphocytes in WAD goat, like in other ruminants were higher than Neutrophils. The percentage distribution of the White blood cell differentials in all the animals is in agreement with the reported values of Belewu *et al*. (2006). The linear increase in the WBC differentials could probably show that animals on diets A to D maintained an active immune system that defends the body against infection, allergic reactions, parasites and antigens**;** thus highlighted the activities of carotenoid in the development of immune system in the WAD goats. This conforms to Lin *et al*. (2006) who stated that Vitamin A is necessary for increasing the immunity through the production of antibodies and its deficiency would reduce immune response and increase susceptibility to infection. In addition, studies have found that beta-carotene supplementation substantially increases the proliferation of lymphocytes, a marker of immune function and immune cell surveillance (Moriguchi *et al*., 1996).

Table 3: Haematology of West African dwarf (WAD) goat fed provitamin A cassava peel-centrosema leaf meal based diets

	Dietary levels					Normal range
Parameters	**A**	**B**	**C**	**D**	**SEM**	
Packed Cell Volume (%)	28.87^c	30.35^b	30.53^b	32.91^a	2.49	22 - 38 (%)
Red Blood Cell x10^6/ul	10.94	10.71	10.97	11.01	0.41	8-18($\times10^6$ /μL)
Haemoglobin g/dl	10.77^d	11.63^c	12.40^b	13.23^a	0.48	8 - 12 g/dl
MCHC (%)	31.05^b	33.68^a	33.79^a	34.25^a	1.52	30-36 ((g/dl)
Mean cell Haemoglobin (pg)	6.13	6.27	6.29	6.34	0.19	5.2 - 8 (pg)
Mean cell volume (fl)	17.04^b	17.11^b	18.37^a	19.04^a	0.97	16 - 25 (fl)
White Blood Cell ($\times10^9$/l)	8.82^d	9.38^c	10.50^b	11.69^a	0.41	4-13($\times10^3$/μL)

Lymphocyte (%)	61.50^{c}	64.13^{b}	64.50^{b}	67.13^{a}	4.53	50 - 70 (%)
Neutrophil (%)	32.78^{c}	34.81^{b}	35.98^{b}	38.14^{a}	2.35	30 - 48 (%)
Eosinophil (%)	1.75	1.50	1.25	1.00	0.09	1 - 8 (%)

$^{a, b, c, d}$ means in the row with different superscripts are significantly different ($P<0.05$)

MCHC = Mean Cell Haemoglobin Concentration

The blood biochemistry of West African dwarf (WAD) goat fed provitamin A cassava peel-centrosema leaf meal based diets is presented in Table 4. Total protein, albumin, globulin and creatinine were significantly ($p<0.05$) influenced by the diets and tended to increase with an increasing levels of the test ingredients. Urea and AST also were significantly ($p<0.05$) affected and followed a specific pattern decreasing with increasing levels of the test ingredients. Bilirubin, ALT and ALP were not affected ($p>0.05$) by the treatment diets. Cholesterol however differed significantly ($p<0.05$) but did not follow any specific trend. The total protein, albumin and globulin were significantly ($P<0.05$) influenced by the treatment diets. The total protein fell within the normal range reported for healthy goats, indicating that the animals did not survive at the expense of body reserves. However, the statistically significant ($P<0.05$) differences observed among the treatment animals may related to reports of Jiwuba *et al*. (2016) that the serum protein is related to the amount of available dietary protein. The albumin and globulin values are slightly comparable to values reported by Oni *et al*. (2012) for WAD goats fed dried cassava leaves-based concentrate diets and fell within the reference range for healthy goats; an indication of proper functioning of the liver and high immunity response of the experimental animals respectively (Jiwuba *et al*. 2016). The normal range observed in this study for the total protein, albumin and globulin of WAD goats suggested that provitamin A cassava peel and *centrosema pubescens* leaf meal contained a tolerable levels of tannins known to diminish nutrient permeability in gut walls as well as increase excretion of endogenous protein which is subsequently passed out in the faeces and so may not alter protein metabolism (Mitjavila *et al*., 1977).

The creatinine and serum urea showed a significant ($P<0.05$) difference among the treatment groups. Creatinine was highest (1.34mg/dl/) in diet D animals and lowest (1.10mg/dl) in diet B animals and fell within the normal range (0.7-1.5mg/dl) of apparently healthy goats as reported by Fraser and Mays (1986); suggesting that there was no wasting or catabolism of muscle and that the animals did not survive at the expense of body reserve. Hence, Prvulovic et al. (2012) noted that creatinine level is direct correlation with muscle mass and kidney function in animals. This indicated that dietary protein was well used by the goats. The serum urea concentration in this study tended to decrease with increasing levels of the test ingredient with animals on diet A having the highest value and the lowest on diet D and however fell within reference range for apparently healthy goat. Increase in serum urea concentration may suggest an increase in orninthine, carbonyl transferase and orginase, which may indicate kidney damage (Ajabonna *et al*., 1999). The normal range of values implied therefore that the dietary proteins of the test diets were better utilized.

The diets in this present study did not significantly ($P>0.05$) influence the bilirubin and cholesterol values in the serum of the WAD goats, hence indicated the safety of the diets as supplements for WAD goats.

Enzymes are protein catalysts present mostly in living cells and are constantly and rapidly degraded although, renewed by new synthesis (Coles, 1986). Zilva and Pannall (1984) stated that normal enzyme level in serum is a reflection of a balance between synthesis and their release, as a result of the different physiological processes in the body. Aspartate aminotransferase (AST) is an enzyme abundantly found in liver and heart muscles and plays an important role in amino acid metabolism (Vojta *et al.*, 2011). There was significant ($P<0.05$) difference in AST levels among

the treatment groups and however fell within the normal range for apparent healthy goat as reported by Olaifa and Opara (2011). This result indicated the absence of necrosis, myocardial infarctions or hepatic metabolism; which are all indicators of good protein quality of the diets. This tends to suggest that the quality of protein in test diets were better than that of the control diet. The activities of the enzymes alanine transaminase (ALT) and alkaline phosphatase (ALP) studied were similar ($P>0.05$) among the treatment groups but however are within the reported reference range for goats. However ALT is an enzyme found in the highest amount in liver and typically used to detect liver injury (Pratt, 2010); thus indicating the safety of the diets for proper liver functioning. Guyton (1991) noted that ALP level in the blood is usually a good indicator of bone formation since osteoblasts secrete large quantities of this enzyme. The similarity of the ALP across the treatment could be said that the test diets did not adversely disrupt the activity of bone formation.

Table 4: Blood biochemistry of West African dwarf (WAD) goat fed provitamin A cassava peel-centrosema leaf meal based diets

	Dietary levels					
Parameters	**A**	**B**	**C**	**D**	**SEM**	**Normal range**
Total protein (g/l)	66.21^{b}	66.41^{b}	72.00^{a}	73.32^{a}	3.99	60 - 79 (g/l)
Albumin (g/l)	32.20^{b}	32.36^{b}	34.98^{a}	34.33^{a}	1.06	27 - 39 (g/l)
Globulin (g/l)	34.01^{c}	34.05^{c}	37.02^{b}	38.99^{a}	1.23	27 - 41(g/l)
Creatinine (mg/dl)	1.11^{b}	1.11^{b}	1.18^{a}	1.34^{a}	0.08	0.7 - 1.5 (mg/dl)
Urea (mg/dl)	17.85^{a}	15.52^{b}	16.21^{b}	13.11^{c}	0.39	10 - 27 (mg/dl)
Bilirubin (mg/dl)	0.18	0.20	0.19	0.22	0.02	0–1.71 (μmol/L)
Cholesterol (mg/dl)	62.71	63.07	61.40	62.32	3.04	40.1-127.3 (mg/dl)
AST (U/L)	27.12^{a}	24.74^{b}	20.91^{c}	19.61^{c}	7.87	12-38 (U/L)
ALT (U/L)	27.10	27.25	26.75	27.00	1.17	15.3-52.3 (U/L)
ALP (U/L)	81.25	82.75	81.75	82.50	4.26	61.3-283.3 (U/L)

$^{a, b, c, d}$ means in the row with different superscripts are significantly different ($P<0.05$)

AST = Aspartate aminotransferase
ALT = Alanine aminotransferase
ALP = Alkaline phosphatase

CONCLUSIONS

From this study, it could be concluded that provitamin A cassava peel-centrosema leaf meal based diets had no detrimental effect on haematological and serum biochemical parameters and therefore could be included in the diets of West African Dwarf goats up to 30%.

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Ethics approval and consent to participate

This paper followed all the guidelines for the care and use of laboratory animal model of the Federal College of Agriculture, Ishiagu, Ebonyi State, Nigeria.

Conflict of interest

The authors declare that they have no conflict of interest.

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