

Toxicological evaluation of graded levels of freshly harvested bamboo (*Bambusa arundinacea*) and tridax (*Tridax procumbens*) leaves on blood chemistry of rabbits

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Abstract

Bamboo is a perennial plant, readily available throughout the year and has been reportedly used in feeding livestock such as cattle, horse and sheep but known to possess antioxidant due to the presence of biologically active components groups, the phenolics. Thus the effects of bamboo (*Bambusa arundinacea*) leaves partially or totally replacing tridax (*Tridax procumbens*) leaves on blood constituents of rabbits were studied using weaner rabbits in a 70-days experiment. Six diets were formulated such that the control diet (T_1) had 0% leaves (concentrate/formulated feed) while the five other diets were offered at the ratio of 100:0 (T_2), 75:25 (T_3), 50:50 (T_4), 25:75 (T_5) and 0:100 (T_6) of freshly harvested *B. arundinacea* and *T. procumbens* leaves, respectively. Feeds were offered at 4% of the rabbits' live weight and apart from the control diet, the five other diets had 2% fixed concentrate and 2% experimental leaves ratio. Thirty-six weaned mixed sex and cross bred rabbits with an average weight of 1075 ± 15.5 g were allocated to a completely randomized design with six rabbits per treatment. The feeds were analysed for their antinutritional factors. The rabbits were slaughtered while the blood was harvested and used for the blood constituents. The percent ash was appreciably higher in bamboo than tridax leaves. The bioactive antioxidants flavonoids were detected in the leaves of bamboo and absent in tridax leaves. Both bamboo and tridax leaves had appreciably very low tannin. With exception of neutrophils, lymphocyte, eosinophil, basophil and MCHC, other haematological parameters were significantly ($P < 0.05$) affected. Diets had no effect on total protein, albumin, bilirubin, creatinine and ALP. It is concluded therefore that partial or total replacement of tridax with bamboo leaves had no deleterious effect on the health status of growing rabbits.

Keywords: Bamboo leaves, Tridax leaves, Haematology, Biochemical indices, Rabbits, Phytochemical screening

Introduction

World demand for meat has risen sharply during the last few decades (Dave, 2003). The key reasons for these increases in meat demand are increasing population, improving technology and increasing incomes. However, despite this overall improvement in technologies and incomes, per capita consumption of meat has lagged especially in the less-developed countries of the world because protein is the most costly food item (Osho and Asghar, 2004). Inadequate supply of proteins from

traditional livestock such as cattle, goat, sheep, pig and poultry has prompted the need to search for alternative protein sources that are cheap, readily available and posing minimal competition to man (Akinmutimi, 2007). As a contingent plan, the search for more economical source of animal proteins makes rabbit production attractive (Egbo *et al.*, 2001). Rabbits (*Oryctolagus caniculus*) have therefore become a viable option, because of their proverbial prolificacy, early maturity, fast growth rate, high genetic selection

potential, high feed conversion efficiency and economic utilization of space (Owen *et al.*, 2008; Yusuf *et al.*, 2009; Yahaya and George, 2013; Yahaya and Wekhe, 2014). The problem for most rabbit producers however, is the high cost of commercial pellets which may constitute as much as 70% of the total cost of production. This has necessitated the need to seek for alternative feed sources in forages. This is especially so because of the greater availability of forages and ability of rabbits to convert forage into meat for human consumption. Rabbit being a monogastric herbivore, cope with plant matter, such as grasses, legumes and can also thrive on forage diet although potential weight gains are not attained because of the poor nutritive value of tropical forage (Aregboola *et al.*, 1985, Bamikole and Ezenwa, 1999). Studies have proven that optimum performance is attained when forages are combined with formulated diets. Common leaf meals/forages of tropical browse plants that have found their way in rabbit nutrition include: *Leucaena leucocephala* (Herbert *et al.*, 2005), *Microdesmis spp* (Esonu *et al.*, 2002), Paw paw leaf meal (Bitto *et al.*, 2006), *Mucuna pruriens* (Emenalom *et al.*, 2009), *Azadirachta indica*. (Esonu *et al.*, 2006; Ogbuewu *et al.*, 2009), *Chromolaena odorata* (Osa, 2015), *Sporobolus virginicus sp.*, *Salicornia virginica sp.*, *Sportina spatinas sp.*, *Borrichia frutescens*, *Rhizophora mangle*, and *Languncularia racemosa*, the white mangrove (Forys and Humphrey, 1994; Bandaranayake, 2002), *Telfaria occidentalis* (Ugu), *Centrosema pubescens*, and *Leucaena leucocephala* (Makinde *et al.*, 2015). Despite the ability of the rabbits to thrive successfully on the forages, majority of the forages are usually in great abundance during the wet season. Unavailability of forages such as *Tridax*

procumbens during the dry season had become a great challenge for most producers making them to solely depend on concentrates during this period and hence increasing the cost of producing this highly nutritious animal. Bamboo is a perennial plant, readily available throughout the year and has been reportedly used in feeding livestock such as cattle, horse and sheep (Yeshambel *et al.*, 2012). Leaves of bamboo were reported to possess antioxidant potentials due to the presence of biologically active components groups, the phenolics (Nakajima *et al.*, 2003; Zhang *et al.*, 2005; Ajay, 2013) and also rich in proteins like gluteline, lysine, methionine, betain, cholin, proteolytic enzymes like nuclease and urease (Rathod *et al.*, 2011). The high crude protein (18.39-19.39%) and fibre content (26.78-33.19%) range of some bamboo leaves species makes it suitable for livestock feed (Antwi-Boasiako, 2011) and also compare well with other browse plants or forages used in livestock feeding. However, production-cost reduction is not only considered when utilizing forage as animal feed or additives but consideration is also given to its safety with regards to the potential toxicities that may be associated with it and also its effective utilization and overall digestibility. This is because although plants generally contain some bioactive and nutritional properties believed to be responsible for their desirable effects, they also contain some phytotoxins whose toxicological actions have always been ignored. Studies have shown that dietary components have measurable effects on blood components, hence, blood constituents are widely used in nutritional evaluation and survey of animals (Church *et al.*, 1984; Olorode *et al.*, 1995). The hematological and biochemical indices are an index and reflection of the effects of

dietary treatment on the animals in terms of the type and amount of feed ingested (Ewuola *et al.*, 2004). The objective therefore of the study is to evaluate the toxicological impact on blood chemistry of rabbits fed bamboo leaves in partial or total replacement of tridax leaves.

Materials and method

Experimental site

The experiment was conducted at the Rabbitry section of the Department of Animal and Environmental Biology, Adekunle Ajasin University, Akungba-Akoko, Ondo-State Nigeria.

Experimental animals, diets and management

Thirty-six weaned cross bred rabbits (New Zealand White x Chinchilla) and mixed sexes aged between 5-6 weeks and an average weight of 1075 ± 15.5 g were randomly allotted to six treatment groups with six rabbits (three bucks and three does) per treatment. Six diets were formulated and designated T₁, T₂, T₃, T₄, T₅ and T₆. Diet T₁ was the control and contained 100% concentrate/formulated feed, T₂ contained 50% concentrate with 50% (100% freshly harvested bamboo leaves (BL)) leaves, T₃ contained 50% concentrate with 50% (75% BL+25% freshly harvested tridax leaves TL) leaves, T₄ contained 50% concentrate with 50% (50% BL+50% TL) leaves, T₅ contained 50% concentrate with 50% (25% BL+75% TL) leaves and T₆ contained 50% concentrate with 50% (100% TL) leaves. At the start of the experiment, the rabbit were weighed individually and assigned to the six dietary treatments. The formulated diets and experimental leaves were analyzed for proximate composition according to AOAC (2000) and shown in Tables 1 and 2 respectively. Phytochemical screening (alkaloid, tannin, saponin, phytic acid and oxalate) of experimental leaves were

carried out following standard method as described by Trease (1989) and Sofowora (1993) as shown in Tables 2 and 3. The rabbits were stabilized on the diets for one week before starting the experiment. The rabbits were weighed at the start of the experiment and on weekly basis thereafter. The rabbits were offered 4% of their body weight on dry matter basis daily (2% concentrate and 2% forage). Left over feed were also weighed and subtracted from the total feed offered to obtain feed intake on daily bases. The experimental rabbits were housed each in a wooden hutch with a wire mesh floor (so that faeces and urine can drop) and window for ventilation. The condition of housing and management were same for all the experimental rabbits. Feeding and watering of the experimental rabbits were carried out on a daily basis. The experimental animals were managed well for optimum performance.

Data collection

At the end of the 70 days feeding trial, four rabbits per treatment were sacrificed and bled through the jugular vein and blood collected into two vacutainer tubes for each animal, one containing a calculated amount of ethylene diamine tetra-acetic acid (EDTA) for haematological study and the other sterile vacutainer tubes without EDTA. The second set of tubes were covered and centrifuged, serum separated out, decanted, deep-frozen for serum biochemical analyses. Determination of both haematological and serum biochemical variables were done following standard method as described by Lamb (1981).

Data analysis

Data were subjected to statistical analysis using analysis of variance of statistical package for social science (SPSS, 2000). Treatment means were compared using Duncan's option of the same software.

Result and discussion

Shown in Table 2 is the proximate composition of the diets fed to rabbits. The results from the proximate analysis of experimental leaves showed that, in bamboo leaf, the soluble carbohydrate was highest (34.54%) followed by crude protein (24.85%), Ash (15.29%), crude fibre (13.79%) and ether extract (2.71%) with similar trend in *Tridax procumbens* leaves. Phytochemical screening (Tables 3 and 4) revealed that both leaves are rich in oxalates, phytates, alkaloids, saponin but with very little tannins. Bamboo leaves are also rich in flavonoids. The toxic anti-nutritional components showed that in bamboo leaves, phytic acid was highest (14.32mg/g) followed by alkaloid (1.85%), saponin (1.65%), oxalate (1.58%) and tannin (0.005%), whereas in tridax leaves, phytic acid was highest (23.91mg/g),

followed by oxalate (3.78mg/g), alkaloid (2.35%), saponin (1.94%) and tannin (0.008%).

The result of the haematological variables of rabbits fed graded levels of freshly harvested bamboo and tridax leaves are shown in Table 5. The Packed cell volume (PVC) of rabbits on experimental leaf diets (T_2 - T_6) were not significantly different ($P>0.05$) from each other. Aside from Treatment T_3 (75%BL+25%TL), the WBC of rabbits fed experimental leaves was not significantly different ($P>0.05$) with the control T_1 (100% formulated feed). The lymphocytes and its differentials (neutrophils, lymphocytes, monocytes, eosinophils, basophils) and MCH were not significantly different ($P>0.05$) across the experimental diets. The red blood cells (RBC) of rabbits fed experimental leaves were significantly different from each other ($P<0.05$).

Table 1: Ingredient and nutrient composition of experimental basal diet

Ingredients	Quantity
Maize	47.24
Wheat Bran	38.00
Soya Bean Meal	12.49
Bone Meal	1.12
Periwinkle	0.58
Salt	0.35
Premix*	0.15
Total	100
Calculated Composition	
Crude Protein %	16.02
ME (Kcal/Kg)	2400.00
Energy : Protein	150:1
MC (%)	8.77
Ash (%)	6.96
EE (%)	5.05
CF (%)	3.27
CP (%)	17.08
NFE (%)	67.64

ME=Metabolizable Energy; Premix* supplied per type kg diet: Vit A, 100,00 IU; Vit. D 2,000,000 IU; Vit. E, 23,00mg; Vit K3 2,000mg; Vit. B, 3,000 mg; Vit. B2, 6,000 mg; Niacin, 50,00 mg; Calcium, 800 mg; Panthotenate, 10,000 mg; Vit. B6, 5,000 mg; Vit B12, 250 mg ; Folic acid, 100 mg; Biotin, 50 mg; choline chloride, 40,000 mg; Selenium, 120 mg and Anti oxidant, 120,00 mg . MC=Moisture Content; EE= Ether Extract; CF=Crude Fibre; CP=Crude Protein; NFE=Nitrogen free extract.

Table 2: Proximate composition of leaf meal

Leaf	MC (%)	Ash (%)	EE (%)	CF (%)	CP (%)	NFE (%)
<i>B. arundinacea</i>	11.82	15.29	2.71	13.79	21.85	34.54
<i>T. procumbens</i>	11.60	11.77	2.92	15.32	25.25	33.14

MC=Moisture Content; EE= Ether Extract; CF=Crude Fibre; CP=Crude Protein; NFE=Nitrogen free extract

Table 3: Quantitative screening of phytochemical constituents of experimental leaves

leaves	Alkaloid	Tannin	Phlobatannin	Cardiac glycosides	Terpenoids	flavonoids
BL	+	+	-	+	+	+
TL	+	+	-	-	-	-

(+): Present, (-): absent. BL: Bamboo leaves, TL: Tridax leaves

Table 4: Qualitative screening of phytochemical constituents of experimental leaves

	Phytic acid (mg/g)	oxalate (mg/g)	Tannin (mg/g)	Alkaloid (%)	Saponin (%)
Bamboo	14.32±0.25	1.58±0.05	0.005±0.0002	1.85±0.03	1.65±0.032
Tridax	23.91±1.52	3.78±0.08	0.008±0.0001	2.35±0.07	1.94±0.024

The Hb of rabbits fed T₂ (100% BL) and T₆ (100% TL) were not significantly different from each other (P>0.05) but numerically higher than the control. The MCHC and MCV of rabbits on experimental leaf diets were not significantly different (P>0.05) from each other.

The serum biochemical variables of rabbits fed graded levels of freshly harvested bamboo and tridax leaves are shown in Table 6. The total protein, albumin, albumin/globulin, bilirubin, creatinine and ALP were not significantly influenced (P>0.05) by the experimental diets. The globulin of rabbits on treatment T₂ (100% BL) was not significantly different (P>0.05) from the control diet T₁ (formulated feed). The cholesterol of rabbits fed experimental leaves, aside treatment T₄ (50%BL+50%TL) and though numerically higher were non-significant (P>0.05) with the control diet. Rabbit fed experimental leaves were not significantly different (P>0.05) and had higher values for AST and ALT when compared with the control.

The nutritional importance of a given leaf meal depends on the nutrients or anti-nutritional composition (Aletor and Omodara, 1994). The values for oxalate and phytate determined for both leaves were quite lower than 18.09±2.29mg/100g (oxalates) and 94.40±0.20mg/100g (phytates) as reported for *Caesalpinia*

pulcherrima (Pride of barbados) (Prohp *et al.*, 2006). Higher concentration of anti-nutrients such as phytates and oxalates has been shown to exert substantial effects on mineral bioavailability in foods (Weaver and Kannan, 2002). Oxalate salts are poorly soluble at intestinal pH and oxalic acid is known to decrease Ca absorption in monogastric animals (Allen, 1982). These anti-nutrients form complexes with nutritionally important minerals such as Ca²⁺, Mg²⁺, Cu²⁺, Fe²⁺, Mn²⁺, Co²⁺ and Zn²⁺ thereby preventing efficient absorption by the body systems (Aletor and Omodara, 1994). Alkaloids, flavonoids, saponins and tannins are known to have antimicrobial activity as well as other physiological activity (Sofowora, 1980; Evans, 2005). In fact flavonoids have a wide range of biochemical and pharmacological activities in mammalian and other biological systems. They possess anti-inflammatory, anti-oxidant, anti-allergic, hepato-protective, anti-thrombic, anti-viral and anti-carcinogenic activities (Middleton *et al.*, 2000).

The present study was carried out to establish baseline information on the toxicological effect of replacing tridax with bamboo leaves on haematology and serum biochemical characteristics of rabbits. Haematological parameters are good indicators of the physiological status of animal and its changes are of value in assessing the response of animals to various

physiological situations (Esonu *et al.*, 2006). It could also serve as a baseline for comparison in conditions of nutrient deficiency, physiology and health status of animals (Daramola *et al.*, 2005).

The effect of replacing tridax with bamboo leaves on haematological parameters such as white blood cells (WBC), number of platelets, Red blood cells (RBC), haemoglobin (Hb), Mean Cell Volume (MCV) and Mean corpuscular haemoglobin concentration (MCHC) were significantly different in the rabbits across the treatment groups and values were neither here nor there but are in agreement with normal range for healthy rabbits (Archetti, 2008). Values obtained for all the treatment groups indicated nutritional adequacy of the treatment diet and also the potential of bamboo leaves to effectively replace tridax leaves. PCV range of 27.50-37.25% observed in this study was lower than what were reported by Sese *et al.* (2014) (41-47%) for rabbits fed graded levels of mucuna leaf meal, Ojebiyi *et al.* (2013) (36.50-38.70%) for rabbits fed concentrates, *Aspilia Africana* and *Tridax procumbens* and fairly similar to Shah *et al.* (2007) and Njidda and Isidahomen (2010) (31-38%) for rabbits fed sesame seed meal. The values reported for these blood parameters were within the normal physiological range for rabbits (Omoikhoje *et al.*, 2006; Archetti, 2008; Amata, 2010). Strong influence of diet on haematological traits especially on PCV and Hb is observed as indicators of the nutritional status of animals (Kabata *et al.*, 1991). According to Isaac *et al.* (2013), PCV is involved in the transport of oxygen and absorbed nutrients. Increased PCV shows a better transportation and thus results in increased primary and secondary polycythemia. Chineke *et al.* (2016) further posited that high PCV reading indicated an increase in

the number of RBCs or reduction in circulating plasma volume. Adejumo (2004) reported that PCV and Hb were positively correlated with the nutritional status of animal. Hence the values for PCV in this study (100% inclusion levels (T_3)) had no toxicity effect on the health status of the rabbits. It also indicated that bamboo leaves can be used to replace Tridax leaves. The normal PCV indicates the absence of normocytic anaemia which is reported to be characterized by a normal MCV and MCH and only detected by a decreased number of RBC or PCV (Coles, 1986). The result is corroborated by the normal RBC which further elucidated the absence of haemolytic anaemia and depression of erythropoiesis. The RBC values in the present study were higher than the values of $4.31-4.43 \times 10^{12}/l$ reported by Sogunle *et al.* (2007) for rabbits fed cassava peel meal (CPM) diets and lower than the values of $5.94-6.41 \times 10^{12}/l$ reported by Olafadehan (2009) for rabbits fed processed cassava peels. The haemoglobin concentrations (Hb) of rabbit fed leaf meal were within the reported range of 9.5-13.0g/dl indicated by Archetti (2008) for healthy rabbits. The adequate haemoglobin concentration for all the experimental rabbits fed leaf meal is probably an indication that replacing tridax with bamboo leaves in the diet supported haemoglobin synthesis which according to Sirois (1995), among other factors, primarily affected by protein intake. The result suggests absence of microcytic hypochromic anaemia which is due to iron deficiency and improper utilization for the formation of haemoglobin. The WBC values reported in this study were lower than the values of $6.05-9.30 \times 10^6/l$ reported by Sokunbi and Egbunike (2000) for rabbits fed neem (*Azadirachta indica*) leaf meal. Higher WBC count of rabbits on T_3 diet compared to other diets could probably be

due to the blend of anti-nutrients present in both leaves. This is further corroborated by the relatively high lymphocyte count of this diet compared to the four other leaf meal diets. However, obtained values are within the range $3.3-12.2 \times 10^9/l$ for normal rabbit (Archetti, 2008). This observation implies that the diets supported haemopoietic tissues with resultant production of adequate WBC. It has been reported that toxic substance in feed tends to suppress haemopoietic tissues with consequent production of lower WBC. Since none of the rabbits suffered from leucopenia, it appears that replacing tridax with bamboo leaves did not affect the immune status of the rabbits because the WBC functions primarily as a defense system as it contain lymphocyte that have a central role in the immunological defense mechanism of the body (Eroschenko, 2000). Values obtained for basophil, eosinophil, lymphocyte and monocyte are within the normal range of healthy rabbits (Mitruka and Rawnsley, 1977; Kerr, 1989; Archetti *et al.*, 2008). Frandson (1986) reported that the number of neutrophils in the blood increases rapidly when acute infection is present, hence a blood count showing this increase is useful in diagnosis of infections. He reported further that eosinophils which normally are scarce increase in numbers in certain chronic diseases, such as infection with parasites and also in allergic reactions. The low values of basophils and monocytes agrees with the statement that basophils and monocytes are normally present in small to moderate number in the peripheral blood system of rabbits (Bernischke *et al.*, 1987; Odeys, 1996). The rabbits were generally healthy all through the duration of the experiment. Variations in blood platelets may probably be as a result of small sample size tested or platelet clumping in the blood of the rabbit tested (Hewitt *et al.*, 1989).

However, values gotten were in agreement with the normal range of healthy rabbit according to Hewitt *et al.* (1989). Platelet work to form blood clots and prevent loss of too much blood when an injury is sustained and also help in wound healing. Blood platelets are implicated in blood clotting. Low platelet concentration suggests that the process of clot-formation (blood clotting) will be prolonged resulting in excessive loss of blood in the case of injury.

Serum proteins have been explored extensively in nutritional studies to distinguish normal state from abnormal conditions in animals. It is also an indication of the protein retained in the animal body (Akinola and Abiola, 1991; Esonu *et al.*, 2001). Dietary components have shown to have measurable effects on blood components (Harper *et al.*, 1999; Awosanya *et al.*, 2000). The non-significant effect of experimental diets on total protein and albumin of the rabbits indicate the ability of the diets especially the experimental leaves, to support the production of these blood components. Serum creatinine were within the normal range of 0.5-2.6mg/dL (except T₅ (25% BLM + 75% TLM)) reported by Hewitt *et al.* (1989). Total protein and creatinine contents have been shown to depend on the quantity and quality of dietary protein (Eggum, 1970; Iyayi, 1998; Awosanya *et al.*, 1999; Esonu *et al.*, 2001) with muscle wasting reportedly shown to be linked to excess creatinine in the blood of animals and is normally due to creatinine phosphate catabolism during the process (Bell *et al.*, 1992). This is not supported in the present study, thus ruling out the possibility of muscle wasting. Serum urea values of experimental rabbits did not follow a particular pattern and fall within the normal range of healthy rabbits of 8.1-25.0mg/dL (Hewitt *et al.*, 1989). The high serum urea concentration value recorded for

Table 5: Haematological variables of rabbits fed graded levels of freshly harvested Bamboo and Tridax leaves

Parameters	Units	T1	T2	T3	T4	T5	T6
PCV	%	24 ¹ 27.50±7.14 ^{b 2}	37.25±4.72 ^a	31.50±4.04 ^{ab}	34.75±3.30 ^a	33.00±2.71 ^{ab}	34.50±0.58 ^a
WBC	10 ⁹	24 3.85±2.41 ^b	4.75±1.46 ^{ab}	6.38±1.18 ^a	5.55±1.03 ^{ab}	4.73±0.30 ^{ab}	3.55±0.86 ^b
Neutrophils	%	24 51.50±3.11	51.25±1.71	48.75±5.19	50.50±2.52	50.50±2.89	52.75±1.89
Lymphocytes	%	24 44.00±2.58	43.75±1.50	45.75±4.43	45.75±2.50	45.00±2.58	43.50±1.73
Monocytes	%	24 2.50±0.58 ^{abc}	2.75±0.96 ^{ab}	3.25±0.96 ^a	1.50±0.58 ^c	1.75±0.50 ^{bc}	2.00±0.00 ^{bc}
Eosinophil	%	24 1.25±0.50	1.25±0.50	1.25±0.50	1.25±0.50	1.75±0.50	1.00±0.00
Basophils	%	24 0.75±0.50	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00	0.75±0.50
Platelet	10 ⁹	24 189.25±129.30 ^b	196.50±138.37 ^b	325.75±74.09 ^{ab}	405.75±92.06 ^a	306.75±74.75 ^{ab}	287.75±143.77 ^{ab}
RBC	10 ¹²	24 4.01±1.07 ^b	5.79±0.72 ^a	4.95±0.49 ^a	5.49±0.38 ^a	5.17±0.46 ^a	5.10±0.29 ^a
Haemoglobin	g/dl	24 8.55±2.42 ^b	11.98±1.63 ^a	10.13±1.34 ^{ab}	11.13±1.35 ^a	10.63±0.96 ^{ab}	11.23±0.33 ^a
MCHC	%	24 ¹ 30.96±1.07 ^{b 2}	31.28±0.74 ^{ab}	32.14±1.17 ^{ab}	31.98±1.77 ^{ab}	32.19±0.75 ^{ab}	33.03±0.97 ^a
MCH	Pg	24 21.28±0.63	20.67±1.07	20.46±1.57	20.23±1.42	20.57±0.66	22.04±0.96
MCV	fl	24 68.75±0.73 ^a	66.14±4.12 ^{ab}	58.60±13.04 ^b	63.29±2.25 ^{ab}	63.90±0.76 ^{ab}	67.29±3.97 ^{ab}

^{abc}; Means with different superscripts along the same row are significantly different (P>0.05) from each other; T₁=formulated feed (FF) (100%), T₂=50% FF + 50% (100% Bamboo Leaves (BL)), T₃=50% FF + 50% (75% BL+25% Tridax leaves (TL)), T₄=50% FF+50% (50% BL+50% TL), T₅=50% FF+50% (25% BL+75% TL), T₆=50% FF+50% (100% TL). MCV=Mean Cell Volume; MCH=Mean Corpuscular Haemoglobin; MCHC= Mean Corpuscular Haemoglobin Concentration; WBC= White blood cell; RBC= Red blood cell; PCV=Packed Cell volume

Table 6: Serum biochemical variables of rabbits fed graded levels of freshly harvested Bamboo and Tridax leaves

Parameters	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆
Total protein (mg/dL)	7.46±0.87	7.70±2.12	5.28±0.66	6.00±0.62	6.77±1.14	5.98±2.31
Globulin (mg/dL)	2.48±0.44 ^a	1.95±0.78 ^a	1.65±0.58 ^b	1.61±0.29 ^b	1.93±0.49 ^{ab}	1.55±0.24 ^b
Albumin (mg/dL)	4.98±0.44	5.78±2.19	3.63±0.90	4.39±0.52	4.79±1.44	4.43±2.44
Albumin/globulin	2.04±0.22	3.26±1.59	2.55±1.50	2.79±0.59	2.70±1.41	2.98±1.85
Cholesterol (mg/dL)	37.99±1.99 ^b	64.64±9.20 ^{ab}	42.62±7.45 ^{ab}	65.61±4.76 ^a	54.30±6.00 ^{ab}	37.14±1.39 ^{ab}
Glucose (mg/dL)	14.50±0.71 ^b	22.00±2.83 ^{ab}	17.50±3.54 ^b	27.00±7.07 ^a	14.50±3.54 ^b	15.50±0.71 ^b
Bilirubin (mg/dL)	0.12±0.03	0.49±0.48	0.77±0.82	0.41±0.57	0.15±0.04	0.28±0.18
Creatinine (mg/dL)	2.07±0.66	2.52±0.60	2.16±0.32	2.27±0.21	2.95±0.70	2.51±1.15
Urea (mg/dL)	16.09±4.27 ^c	13.08±3.29 ^c	23.69±6.29 ^b	17.77±4.32 ^{bc}	15.87±5.82 ^c	33.52±2.83 ^a
ALP (IU/L)	24.02±5.15	28.09±5.32	27.19±11.00	30.27±10.15	30.21±13.41	24.87±13.01
AST (IU/L)	39.00±4.88 ^b	50.88±14.93 ^{ab}	53.75±25.42 ^{ab}	57.25±30.39 ^{ab}	66.13±23.92 ^a	53.00±22.56 ^{ab}
ALT (IU/L)	18.05±1.68 ^b	28.75±7.12 ^a	26.50±8.37 ^{ab}	26.85±8.20 ^{ab}	29.55±5.02 ^a	24.10±2.65 ^{ab}

^{abc}: Means with different superscripts along the same row are significantly different ($P > 0.05$) from each other; T₁=formulated feed (FF) (100%), T₂=50% FF + 50 % (100% Bamboo Leaves (BL)), T₃=50% FF + 50% (75%BL+25%Tridax leaves (TL)), T₄= 50% FF+50% (50% BL+50%TL), T₅=50% FF+50 % (25%BL+75%TL), T₆= 50% FF+50 % (100%TL). ALP: alanine transaminase; AST: aspartic transaminase

T₆ could probably be attributed to an increase in activities of urea enzymes omithine, carbonyl transferase and originase (Ajagbona *et al.*, 1992) resulting from anti-nutrition factors contained in the test feedstuff. This agrees with Ogbuewu *et al.* (2015) who reported significant high serum urea values (55.50-77.30mg/dL) for rabbit bucks fed neem leaf based diets. It also support the findings of Bawa *et al.* (2006) who reported high serum value range of 60-70mg/dL for growing rabbits fed differently processed neem seed meal. The cholesterol level in this study was significantly affected by the treatment diets and did not also follow a particular trend. However, values are within the range of normal healthy rabbit (25-60mg/dL) (Mitruka and Rawnsley, 1977). Clinical data strongly support a relationship between diets and serum cholesterol levels (Konjucfa *et al.*, 1997). The findings in this study disagree with the results of Ogbuewu *et al.* (2015) and Obikaonu *et al.* (2011) who observed a downward trend in serum cholesterol values with increasing levels of leaf meals. Since cholesterol levels were within the normal range, possibilities of anorexia, diabetes, liver dysfunction and malabsorption of fat which are the symptoms of abnormal cholesterol levels in the blood are ruled out. Serum bilirubin, ALP and ALT values were similar among the dietary groups. Serum bilirubin values reported here were within the recommended range of 0-1.0mg/dL (Mitruka and Rawnsley, 1977). Rabbits fed experimental leaves had higher values of AST than the control diet. The AST and ALT concentrations are within the normal range of 42.5-98.01IU/L and 48.5-78.9IU/l respectively reported for rabbits (Mitruka and Rawnsley, 1977). An increase in serum ALT and AST above the normal range has been reported to signify necrosis and

myocardial infaction or response to the presence of a number of toxic factors (Sigma, 1985). The normal ALT levels indicate that the activities of osteoblast were not affected because the blood level of ALT is usually a good indicator of the rate of bone formation (Guyton, 1991).

Conclusion

The study showed that fresh bamboo leaves could be fed wholly or partially along with concentrate and other commonly used forages with no deleterious or toxicological effects on blood components of rabbits

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