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Research Article

Blood Profile of Friesian X Bunaji Dairy Calves Fed Diets Containing Different Ratios of Soymilk

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Abstract

Blood is particularly sensitive to changes in nutrition and environmental temperature. The study investigates the response of dairy calves to soymilk as replacement for cow milk. Soybean was sourced, cleaned and soaked in clean water for 72 hours. The water was changed twice after every 24 hours of soaking. Thereafter, the soybean was rinsed, sun-dried for 8 days, milled, sieved and then taken to the laboratory for chemical analyses. Results showed that the soymilk contained 87.11% moisture, 12.89% total solid (TS), 3.30% fat, 9.59% solid-non-fat (SNF), 5.55% protein and 0.82% ash. While the milk obtained from Friesian x Bunaji cows had moisture, TS, fat, SNF, protein and ash contents of 88.30%, 11.70%, 2.69%, 9.01%, 3.29% and 0.67%, respectively. The protein content of the different ratios of soy: cow milk diets increased with increased levels of soymilk and varies from 3.29% in 0:100 to 5.23% in 75:25 ratio of soy: cow milk. The TS, fat, SNF and ash contents also followed similar pattern. Sixteen Friesian x Bunaji dairy calves with body weight of 34.8±0.7kg were randomly assigned to four dietary treatments (which consisted of 0:100, 25:75, 50:50 and 75:25 ratios of soy: cow milk) with four calves per treatment in a Completely Randomized Design. Each calf received two litres of the mixture of soy: cow milk daily. *Digitaria smutsii* hay and clean drinking water were provided to the calves' ad libitum. Results showed significant (P<0.05) difference in packed cell volume (PCV) across the treatments. Calves fed 75:25 ratio of soy: cow milk had higher (19.53) PCV percentage compared with those on the control diet which had the lowest (15.01%). While for white blood cell (WBC) calves fed diets containing 75:25 and 50:50 ratios of soy: cow milk had higher (11.65 and 12.46x109/l) WBC compared with those on 0:100 and 25:75 which had lower (10.28 and 10.66 x109/l) WBC values, respectively. Haemoglobin concentration also followed similar pattern. Serum total protein was significantly (P<0.05) higher (6.32g/dl) in calves fed 75:25 ratio of soy: cow milk though not statistically different from calves fed 50:50 (6.28g/dl) and 0:100 (6.1228g/dl). The lowest serum protein (5.51g/dl) was observed in calves fed 25:75 ratio of soy: cow milk. Albumin, glucose and creatinine also followed similar pattern. But the amount of globulin was significantly (P<0.05) higher (2.44g/dl) in calves fed 0:100 ratio of soy: cow milk (the control) compared with calves on the other treatments. Calves fed 50:50 ratio of soy: cow milk had the highest (9.79mg/dl) blood urea nitrogen (BUN) compared with calves on 25:75, which had the lowest (7.13mg/dl) concentration of BUN but statistically similar to those on 0:100 (7.60mg/dl). All the blood parameters measured were within the normal range for healthy calves. Therefore, it was concluded that soymilk can be used to replace cow milk in the diet of calf up to 75% inclusion level without any detrimental effect on health status of the animals.

Introduction

The low productivity of ruminants in developing countries is characterized by high calf mortality, poor growth rate, delay in onset of puberty, and long interval between successive parturition all of which are largely attributable to poor feed resources, feeding and management [1]. During the initial stage of life, dairy calves depend solely on milk for their nutritional needs and gradually adapt to solid feeds as the rumen becomes functional. But due to shortage of liquid milk, calves cannot get the required amount of milk from their dam resulting in underfeeding or starvation with a consequence of stunted growth and mortality [2]. Suitable substitutes for milk such as soymilk can improve the nutrition and survivability of infant pre-ruminants [3].

Milk replacer is an ideal liquid feed for pre-ruminants. In developed countries, alternatives to whole milk (called milk replacers) are formulated using by-products of milk processing industry [4]. Commercial milk replacers are scarce and expensive because they are mostly imported. Soybean (*Glycine max L.*) is readily available, less expensive and can be cultivated locally. Soybean has potential for use in milk replacer formulations because of its high nutritive value. Soybean is a good source of high quality, relatively inexpensive protein. Soybean flour contains up to 40% protein compared with 1.0 to 5.6% protein content of most animal milk [2] and is also rich in minerals and vitamins such as iron, zinc, copper, thiamine, riboflavin, niacin and panthothenic acid. Most of these are well known haematinics – a general term for those nutrients, including iron, folic acid, and vitamin B12, required for the formation and development of blood cells in the bone marrow [5].

Soybean protein is often used in calf milk replacers and starters instead of milk proteins because of its low cost and its essential amino acid content which is similar to cow's milk. Soymilk is cheaper than cow milk therefore, successful replacement of cow milk with soymilk may reduce feed cost. Roy *et al.* (2016) reported that soy proteins are widely used in milk replacer formulations [2]. However, to ensure proper growth and health of the calf, processing and subsequent solubility of the protein source are crucially important for determining the suitability of alternative proteins in milk replacers. Soaking as a processing method could be employed to improve the feeding value of soymilk. This technology can easily be adopted by farmers to enhance the survival of calves and have excess milk for sale [6].

Blood is an important and reliable medium for assessing the health status of animals [7]. Blood is frequently used for assessing threat to homeostasis [8]. The examination of blood provides the opportunity to clinically investigate the presence of metabolites and other constituents in the body of animals and it plays a vital role in the physiological, nutritional and pathological status of an animal [9]. Blood analyses are vital tools that help detect any deviation from normal in the animal's body [10]. The study therefore investigates the effect of feeding different ratios of soymilk on some haematological and serum biochemical parameters of Friesian x Bunaji dairy calves.

Materials And Methods

Location of the Study

The study was conducted at the National Animal Production Research Institute (NAPRI), Ahmadu Bello University, Shika-Zaria, Nigeria. Shika is located within the Northern Guinea Savanna ecological zone of Nigeria between latitude 10°11'N and longitude 7°8'E, at an altitude of 650m above sea level [11]. The area receives a mean annual rainfall of 1150mm which commences from May and last till October. Following the wet season is a period of dry cool weather known as 'harmattan' which marks the onset of the dry season. This extends from mid-October to March when the hot weather sets in. At this period, the mean minimum and maximum temperatures range from 12 – 28°C during the cold 'harmattan' season and 20 – 36°C in the hot season. The mean relative humidity is 21% and 72% during the 'harmattan' and the rainy seasons respectively [12].

Source of Feed Materials

The soybean variety used for this study was Samsoy II and was purchased from an open market in Giwa Local Government Area of Kaduna State, Nigeria. While the other feed materials (*Digitaria smutsii* hay, concentrate diets and Friesian x Bunaji milk) were obtained from Dairy Research Programme of NAPRI, Shika-Zaria, Nigeria.

Preparation of Soyflour

Soybean was cleaned by winnowing and hand picking of stones and debris. The cleaned soybean was soaked in excess water in plastic containers for 72 hours. The water was changed twice after every 24 hours during the soaking period. After which the soybean was rinsed with clean tap water and sun-dried for 8 days. The dried soybean was milled into flour and sieved with the aid of 0.04mm sieve. The resultant soyflour was stored in polythene bags and samples were taken to the laboratory for chemical analyses. The flowchart of the processing method is shown in figure 1 below.

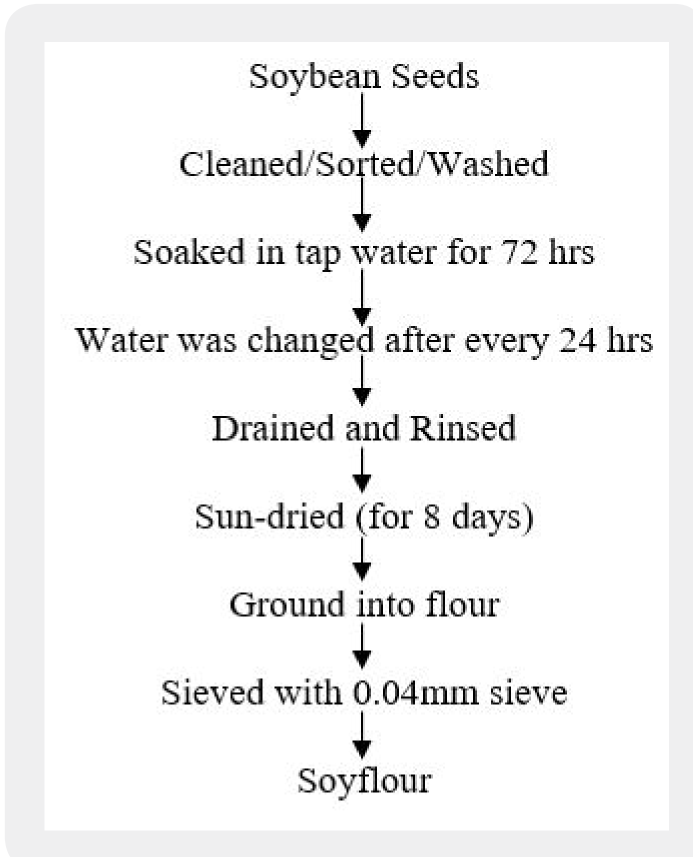

Figure 1: *Flowchart for Soybeans Processing into Soyflour*

Source: Revised from Pele *et al.* (2016).

Experimental Diets

Ten (10) litres of Friesian x Bunaji milk from the Dairy Research programme of NAPRI was collected at 08:00am and used for the morning feeding, while another 10 litres was collected at 04:00pm and used for the evening feeding, respectively. The experimental diets were prepared immediately after fresh milk collection. Soymilk was prepared in batches according to standard procedure. To formulate one litre of soymilk, 125g of the 72 hrs soaked soyflour was dissolved in 1000ml of clean tap water according to methods described by Sarker *et al.,* (2015) [13]. Thereafter, 6 litres of soymilk was prepared by dissolving 750g of the 72 hrs soaked soyflour in 6000 ml of clean tap water. The mixture was homogenized by constant stirring to prevent coagulation. The soymilk was added to fresh cow milk in a stainless container at the ratio of 0:1000mls (which served as the control); 250:750mls; 500:500mls and 750:250mls soymilk: cowmilk, respectively before feeding to the calves.

Experimental Animals, Design and Management

Sixteen (16) Friesian x Bunaji dairy calves of mixed sexes (8 males and 8 females) aged between 2 - 3 weeks, with live weight of 34.8±0.7kg from the Dairy Research Programme of the National Animal Production Research Institute (NAPRI), Ahmadu Bello University (ABU), Shika-Zaria were used for the growth trial. The calves were identified with ear tags, weighed and randomly distributed into four (4) dietary treatments consisting of 4 calves per treatment in a Completely Randomized Design (CRD). The calves were housed in an open-sided, well-ventilated building (calf-pen) with a concrete floor and equipped with both feeding and watering troughs. Each animal received two (2) litres of whole milk and mixture of soymilk and cow milk daily (one litre of the liquid feed was provided between 08:00 - 09:00am, and another 1 litre was provided in the evening between 04:00 - 05:00pm. In addition, the experimental animals were given weighed quantities of concentrate diet and *Digitaria smutsii* hay to stimulate rumen development. The concentrate diet was made up of maize, maize offal, cotton seed cake (CSC), bone meal, common salt (NaCl) and vitamin-mineral premix (Table 1). Fresh and clean drinking water was also provided daily *ad libitum*. Proper sanitary measures were obeserved to protect the calves against parasitic infestations and other contagious diseases. The growth trial was carried out for a period of 98 days (between October, 2016 to February, 2017).

Table 1: *Ingredient Composition of Concentrate Diet fed to Friesian x Bunaji Calves*

Ingredients	Percentage (%)
Maize	22.00
Maize Offal	22.75
Cotton Seed Cake	52.00
Bone Meal	2.00
Common Salt (NaCl)	1.00
Vitamin-Mineral Pre-mix	0.25
Total	**100.00**

Blood Collection

Blood samples were collected from all the experimental animals (16) at the end of the experiment. Five (5) mls of blood sample was drawn through the jugular vein using hypodermic needles with syringe and was emptied into two separate sets of vacutainer tubes. Two (2) mls of the collected blood sample was poured into a plastic bottle containing anti-coagulant [ethylene diaminetetracetic acid (EDTA)]. The vials were capped immediately and the contents were tilted twice gently to mix the blood with the EDTA as reported by Aye (2013) [14]. The remaining 3mls was emptied into another plastic bottle that do not contain EDTA. For blood samples collected, the heparinised blood was used for haematological studies which include: packed cell volume, haemoglobin, red blood cell, white blood cell, lymphocytes, neutrophils and basophils.

Laboratory Analyses

Milk and soymilk samples were analyzed for gross composition (moisture, total solids, solid-not-fat, fat, protein and ash). The total solid (TS) was determined according to the method described by Bradley Jr (2003) by drying 5g of milk samples to a constant weight at 105°C for 3 hours [15,16]. Fat content was estimated by the Gerber method (AOAC, 2005). Solid-not-fat (SNF) was calculated as the difference between TS and fat content (SNF = TS - fat).

Milk protein (Nitrogen x 6.38) was determined by semi-micro distillation method using Kjeldahl and Makhamps apparatus and ash content was determined by igniting the dried samples at 500°C [15]. For blood samples collected, the heparinised blood was used for haematological studies which include: packed cell volume, haemoglobin, red blood cell, white blood cell, lymphocytes, neutrophils and basophils. While the blood samples in the set of bottles containing no anti-coagulant were kept in the refrigerator at a temperature of about −4°C for about 3 hours to aid sedimentation. The samples were later spun in a centrifuge at 3000 rpm for 10 minutes and the serum was separated and stored at −10°C for analysis. The sera for each of the animals were then used to assay for the serum metabolites (Total Protein, Globulin, Albumin, Glucose, Blood Urea Nitrogen and Creatinine) according to methods described by Etana *et al.* (2011) [17].

Results

Chemical Composition of Friesian x Bunaji Milk and Soymilk (on wet basis)

The result of the chemical composition of milk from Friesian x Bunaji cows and soymilk is presented in Table 2. The cow milk had a total solid, fat, solid-non fat, protein and ash content of 11.70, 2.69, 9.01, 3.29 and 0.67%, respectively. While soymilk had a total solid, fat, solid-non fat, protein and ash content of 87.11, 12.89, 3.30, 9.59, 5.55 and 0.82%, respectively.

Table 2: *Chemical Composition of Friesian x Bunaji Milk and Soymilk*[*]

Parameters (%)	Friesian x Bunaji Milk	Soymilk (soaked for 72 hrs)
Moisture	88.30	87.11
Total Solid	11.70	12.89
Fat	2.69	3.30
Solid-Not-Fat	9.01	9.59
Protein	3.29	5.55
Ash	0.67	0.82

[*] = Wet basis

Chemical Composition of Different Ratios of Soy: Cow Milk fed to Calves

Table 3 shows the results of the chemical composition of the different ratios of soy:cow milk fed to Friesian x Bunaji calves. The moisture content differed significantly ($P<0.05$) and ranged from 81.98 in the diet containing 75:25 ratio to 88.30% in 0:100 ratio (the control diet). There was a significant (P>0.05) increase in total solids as the level of soymilk inceased in the diet. The fat, solid-non fat, protein and ash content also follow similar trend. Total solids, fat and solid-not-fat were higher in diet containing 75:25 ratio (18.02, 4.75 and 13.27%) but not significantly (P>0.05) different from 50:50 ratio (16.52, 3.75 and 12.77%), respectively. However, protein and ash contents in the diet containing 75:25 ratio of soy:cow milk were higher (5.23 and 1.45%) compared with (3.29 and 0.67%) of protein and ash in the control diet.

Table 3: *Chemical Composition of Different Ratios of Soy:Cow Milk fed to Calves*

Parameters (%)	Ratios of Soy to Cow Milk (%)				
	0:100	**25:75**	**50:50**	**75:25**	**SEM**
Moisture	88.30^{a}	84.80^{b}	83.48^{c}	81.98^{d}	0.21
Total Solids	11.70^{b}	15.20^{ab}	16.52^{a}	18.02^{a}	0.83
Fat	2.69^{b}	3.02^{ab}	3.75^{a}	4.75^{a}	0.53
Solid-Not-Fat	9.01^{b}	12.18^{ab}	12.77^{a}	13.27^{a}	0.39
Protein	3.29^{b}	4.35^{ab}	4.40^{ab}	5.23^{a}	0.67
Ash	0.67^{c}	0.99^{b}	1.08^{b}	1.45^{a}	0.09

^{abcd} means with different superscripts within the same row differed significantly (P<0.05).
SEM = Standard Error of Mean.

Effect of Feeding Different Ratios of Soy: Cow Milk on Some Haematological Parameters of Friesian x Bunaji Calves

The result of the effect of feeding different ratios of soy:cow milk on haematological parameters of Friesian x Bunaji calves is presented in Table 4. There was significant (P<0.05) difference in the packed cell volume (PCV) across the treatments. Calves fed 75:25 ratio of soy:cow milk had higher PCV percenatage (19.53), while calves fed the control diet had the lowest PCV value (15.01%). The result of white blood cell (WBC) showed significant (P<0.05) difference across treatments. Calves fed 75:25 and 50:50 ratios of soy:cow milk were significantly (P<0.05) higher and those fed 0:100 and 25:75 ratios of soy:cow milk had significantly (P<0.05) lower and similar WBC. Haemoglobin concentration also followed similar pattern. While for MCHC, calves fed 0:100 and 25:75 ratios of soy:cow milk had significantly (P<0.05) higher and similar MCHC of 29.91 and 28.58g/dl, respectively followed by (25.08g/dl) in calves fed 50:50 ratio of soy: cow milk, the least MCHC value (21.66g/dl) was recorded in calves fed 75:25 ratio of soy: cow milk.

Table 4: *Effect of Feeding Different Ratios of Soy:Cow Milk on Some Haematological Parameters of Friesian x Bunaji Calves*

Parameters	Ratios of Soymilk to Cow Milk (%)					
	0:100	25:75	50:50	75:25	SEM	Range[1]
PCV (%)	15.01[c]	18.67[ab]	17.67[b]	19.53[a]	0.85	24 - 46
WBC ($x10^9$/l)	10.28[b]	10.66[b]	11.65[a]	12.46[a]	0.49	4 - 12
RBC ($x10^{12}$/l)	6.52	7.22	6.44	7.40	0.51	0 - 20
Hgb (g/dl)	9.81[b]	9.61[b]	10.43[a]	10.78[a]	0.25	8 - 15
MCV (fl)	49.93[b]	49.15[b]	54.55[a]	50.27[b]	0.71	40 - 60
MCH (pg)	15.47[a]	15.36[ab]	15.18[b]	15.91[a]	0.25	11 - 17
MCHC (g/dl)	29.91[a]	28.58[a]	25.08[b]	21.66[c]	1.02	30 - 36
Differential WBC (%)						
Lympocytes	68.88[a]	66.44[ab]	67.08[a]	64.78[b]	1.48	45 - 75
Neutrophils	26.82[a]	18.11[c]	20.97[b]	24.40[a]	1.42	15 - 33
Basophils	0.68[b]	0.94[a]	0.81[ab]	1.10[a]	0.08	0 - 2

[abc] means with different superscripts within the same row differed significantly (P <0.05).
PCV = Packed Cell Volume; WBC = White Blood Cells; RBC = Red Blood Cells; Hgb = Haemoglobin;
MCV = Mean corpuscular volume; MCH = Mean corpuscular haemoglobin;
MCHC = Mean corpuscular haemoglobin concentration; SEM = Standard Error of Mean.

[1]**Source:** Merck Veterinary Manual (2016).

Effect of Feeding Different Ratios of Soy: Cow Milk on Some Serum Biochemical Indices of Friesian x Bunaji Calves

The results of feeding different ratios of soy:cow milk on some serum biochemical indices of Friesian x Bunaji calves are presented in Table 5. Serum total protein differed significantly (P<0.05) across the treatments and was highest (6.32g/dl) in calves fed 75:25 ratio of soy: cow milk though not statistically different from calves fed 50:50 (6.28g/dl) and 0:100 (6.12) ratios of soy:cow milk. The lowest serum protein (5.51g/dl) was observed in calves fed 25:75 ratio of soy: cow milk. Albumin, glucose and creatinine also followed similar pattern. But the amount of globulin was highest (2.44g/dl) in calves fed 0:100 ratio of soy:cow milk (the control) and lowest in calves fed 75:25 ratio of soy:cow milk though not signifcantly (P>0.05) different from calves fed 25:75 and 50:50 ratio of soy:cow milk. There was a significant (P<0.05) difference in blood urea nitrogen (BUN) across the treatments. Calves fed 50:50 ratio of soy:cow milk had the highest BUN of (9.79mg/dl) while calves fed 25:75 ratio of soy:cow milk had the lowest BUN concentration of (7.13mg/dl) and was statistically similar with calves fed the control diet (7.60mg/dl).

Table 5: *Effect of Feeding Different Ratios of Soy:Cow Milk on Serum Biochemical Indices of Friesian x Bunaji Calves*

Serum Indices	Ratios of Soy to Cow Milk (%)					
	0:100	**25:75**	**50:50**	**75:25**	**SEM**	**Range[1]**
Total Protein (g/dl)	6.12^{a}	5.51^{b}	6.28^{a}	6.32^{a}	0.17	6.7 - 7.5
Albumin (g/dl)	3.68^{b}	3.34^{c}	4.11^{a}	4.32^{a}	0.15	2.5 - 3.8
Globulin (g/dl)	2.44^{a}	2.17^{b}	2.17^{b}	2.00^{b}	0.09	3.0 - 3.5
Glucose (mg/dl)	81.72^{b}	73.87^{c}	76.38^{bc}	96.20^{a}	2.87	40 - 100
Blood Urea Nitrogen (mg/dl)	7.60^{c}	7.13^{c}	9.79^{a}	8.99^{b}	0.30	10 - 25
Creatinine (mg/dl)	0.47^{b}	0.46^{b}	0.60^{a}	0.58^{a}	0.03	0.5 - 2.2

^{abc} means with different superscripts within the same row differed significantly (P<0.05).
g/dl = gram per decilitre; mg/dl = milligram per decilitre; SEM = Standard Error of Mean.

[1]**Source:** Merck Veterinary Manual (2016).

Discussion

Chemical Composition of Friesian x Bunaji Milk and SoyMilk (wet basis)

Milk is a complex mixture of fat, proteins, carbohydrate, minerals, vitamins and other miscellaneous constituents dispersed in water [18]. Milk composition including moisture, protein, fat, total solid and solid-non fat are an important indicators of milk quality [19]. The percentage of total solid obtained for Friesian x Bunaji milk was lower than the value reported by (Ahmed *et al.*, 2002) in Holstein Friesian cow milk in Pakistan [20]. A similar result was obtained by (Ibeawuchi and Dalyop, 1995) in White Fulani cow milk [21]. The low value recorded for total solid content in this study when compared with other researches may be due to low nutrition and other management practices adopted. The milk protein content of Friesian x Bunaji milk obtained in this study was comparable with values reported by (Ndubueze *et al.,* 2006) for grazing White Fulani cows [22]. And even for Friesian x Bunaji cattle in Nigeria [23]. The total solids of cow milk obtained in this study was slightly below the value reported by (Sarker *et al.*, 2015) which might be due to the difference in breed and feed [13].

The milk fat content obtained in this study was slighlty lower than the value reported by Roy *et al.,* (2016) [2]. Higher milk fat was also reported by Sarker *et al.,* (2015) [13]. The low milk fat content observed in this study might be attributed to age of cows from which the milk was collected, breed of the cows, stage of lactation and nutrition. The breed of cow whose milk were used were a cross between tropical (Bunaji or White Fulani) and temperate (Holstein Friesian), so the milk fat will not be as high as those from pure breeds of Friesian or other dairy cattle like Jersey. Fayeye *et al.,* (2013) reported that Jersey breed of dairy cattle had higher milk fat than Holstein Friesian breed [24]. Egbowon (2004) reported that milk fat content decreases at mid-lactation and continue to decrease until the end of lactation . Also, underfeeding can cause lower proportion of milk components [25]. Similarly, Nickerson (1999) reported that genetic factor, nutrition, environment and milking management practices have important effect on milk composition and quality [20].

The protein content of the 72 hours soaked soymilk observed in this study on wet basis (5.55%) was slightly above the value (4.0%) reported by Sarker *et al.,* (2015) for soymilk obtained by boiling soybeans at 100°C for 15 minutes [13]. But similar to the values of (4.5%) reported by Odumodu (2010) for soybeans soaked for 18 hours [26]. The total solids (12.89%), fat (3.30%) and ash (0.82%) obtained in this study for soymilk were slightly higher than the (10.70%), (2.70%) and (0.60%) reported by Sarker *et al.,* (2015) for total solids, fat and ash, respectively [13]. This study found that soaking increased the protein content of the soybeans. This is consistent with the report of Pele *et al.,* (2016) who found that soaking increased the CP content of soybeans soaked for 12 and 24 hours [27].

Chemical Composition of Different Ratios of Soy: Cow Milk Fed to Calves

The mixture of soy:cow at different ratios showed significant ($P<0.05$) increase in nutritional content as compared with cow milk or soymilk alone. The similar fat content in the diets containing different ratios of soymilk might be attributed to the low fat content in cow milk which had not affect the total fat content in the mixture much. However, the protein content of the mixture of soymilk and cow milk were higher than those of cow milk alone. The non-significant ($P>0.05$) difference observed in the protein contents of the diet containing soymilk in addition to cow milk might be due to the low protein content of cow milk used. Other authors reported higher fat and protein contents in cow milk but the low levels obtained in this study may be attributed to breed of cow, feeding and stage of lactation.

Effect of Feeding Different Ratios of Soy: Cow Milk On Some Haematological Parameters of Friesian X Bunaji Dairy Calves

Haematology is the study of morphology and physiology of blood [7]. Packed cell volume (PCV) which is also known as haematocrit (HT or Hct) or Erythrocyte Volume Fraction (EVF) is the percentage (%) of red blood cells in blood [9]. The PCV obtained in this study (15.01%), (18.67%) and (17.67%) for calves fed 0:100, 25:75 and 50:50 ratios of soymilk and cow milk, were lower than 27.30%, 26.90% and 27.50% reported by Sarker *et al.,* (2015) for goat kids fed the aforementioned ratios of soymilk and cow milk [13]. The PCV values obtained in this study fell within the normal range of 24 - 46% for healthy calves as reported by (Merck Veterinary Manual, 2016) [28]. This implies that the inclusion of soymilk in the diets of calves was ideal and adequate and the animals were not anaemic. Increased PCV shows a better transportation and thus results in an increased primary and secondary polycythemia [9]. Furthermore, Chineke *et al.,* (2006) and Etim *et al.,* (2014a) reported that high PCV values indicated either an increase in number of red blood cells or reduction in circulating plasma volume [7,29].

Bello and Tsado (2013) stated that PCV within the normal range suggests that there was no toxic factor (such as haemaglutinin) in the feed of the animal which may adversely affect blood formation. White blood cells (WBC) followed similar pattern but were still within the normal range (4 - 12%) reported by (Merck Veterinary Manual, 2016) [28,30]. Although calves fed 75:25 ratio of soymilk to cow milk had slightly higher WBC which might be attributed to the high levels of soymilk used in the diet which means higher consumption of possible residuals of ANFs by the calves on that treatment. Higher concentration of WBC in the blood of an animal implies that the animal is trying to fight the effect of some undesirable elements in the diet consumed. Etim (2010) reported that normal range of WBC indicate that the animals are healthy because increase in WBC counts above the normal range is an indication of allergic conditions, anaphylactic shock and certain parasitism [11]. White blood cells are known to act as defensive mechanism against toxic factors in the body.

The WBC counts obtained in this study agrees with the findings of Sarker *et al.,* (2015) who observed elevated serum antibody in calves fed soy products [13]. Reports had also shown that animals with high concentration of WBCs are capable of generating antibodies in the process of phagocytosis and have high degree of resistance to disease [32]. However, the WBC counts obtained in this study fall within the normal range of (4 - 12 x10^9/l) reported by (Merck Veterinary Manual, 2016) [28]. Red blood cells (RBC) serve as a carrier of haemoglobin in the body of the animal. And the haemoglobin reacts with oxygen carried in the blood to form oxyhaemoglobin during respiration [9]. The values of RBCs in this study were within the normal range of (0 – 20 x10^{12}/l) reported for healthy calves (Merck Veterinary Manual, 2016) [28]. This could be due to the fact that soybean is a rich source of minerals and vitamins (such as iron, zinc, copper, thiamine, riboflavin, niacin and patholenic acid) which are well-known hematinics and are essential in the formation of RBCs [33,34]. Haemoglobin, Mean corpuscular volume (MCV), Mean corpuscular haemoglobin (MCH) and Mean corpuscular haemoglobin concentration (MCHC) were all within the normal ranges of 8 - 15g/dl, 40 - 60 fl, 11 - 17pg and 30 - 36g/dl, respectively reported for healthy calves (Merck Veterinary Manual, 2016) [28]. This means that the addition of soymilk in the diets of calves increased the protein content and its utilization.

This finding is consistent with the report of Bello and Tsado (2013) who found that haemoglobin values within normal range imply that the dietary proteins were of high quality [30]. The differential WBC (lymphocytes, neutrophils and basophils) observed in this study fall within the normal range for healthy calves. This means that the levels of ANFs in the processed soybeans did not affect the immune system of the animals. This agrees with Chineke *et al.,* (2006) who found that lymphocytes, neutrophils and basophils offer protection against toxins and infectious organisms in the body of the animal [29]. Thus, significantly high differential WBC may imply the presence of ANFs in the blood. Etim *et al.,* (2014b) corroborated that the immune status of an animal is a function of lymphocytes and neutrophils [32]. And that when these cells fall within the normal range, it's an indication that the feeding pattern did not affect the immune system.

Effect of Feeding Different Ratios of Soy: Cow Milk On Some Serum Biochemical Indices of Friesian X Bunaji Dairy Calves

Blood biochemical indices are considered important in evaluating the health status of animals as a result of feeding, disease or low production [35]. Total protein significantly ($P<0.05$) increased with increased levels of soymilk in the diets of the calves. The serum total protein (5.51 to 6.12 g/dl) obtained in this study were within the range (6.12 to 6.49 g/dl) reported by Geiger *et al.,* (2014) in Holstein calves fed varying milk replacers with or without direct-fed microbial supplementation [36]. The improvement of serum total protein observed in this study might have been associated with improved dietary intake as a result of the inclusion of soymilk. However, the serum TP concentration obtained in this study fall within the normal range (6.7 - 7.5 g/dl) reported by (Merck Veterinary Manual, 2016) for healthy animals [28]. This is in line with the reports of Hagawane *et al.,* (2009) who indicated that TP concentrations are usually used as an appraisal of nutritive status of animals reflecting improved feed intake and metabolism [35].

The albumin concentration is also an important indicator of nutritional status of animals. Albumin performs many functions in the body of an animal including maintaining the osmotic pressure that causes fluid to remain within the blood stream instead of leaking out into the tissues [35]. The serum albumin (3.34 to 4.32 g/dl) obtained in this study were similar to the range (4.1o to 4.80 g/dl) reported by in Holstein calves fed milk replacer containing different amount of energy and protein [37]. But the values of albumin obtained in this study were slighlty above the normal range (2.5 – 3.8 g/dl) for healthy animals which suggest that the calves were not dehydrated. This is because, albumin levels are dependent on the state of hydration of the body.

An animal that is dehydrated will have an artificially high albumin level, which would return to normal when the dehydration is corrected [35]. This study showed that the inclusion of soymilk at different ratios in the diet of calves did not lead to dehydration. Globulins as a component of serum TP are known to play important role in blood clotting, osmotic function, blood viscosity, defensive function, buffering action, capillary permeability, conservative and transport function as well as serve as reserve proteins [35]. The globulin concentrations obtained in this study were within the normal range (3.0 - 3.5 g/dl) for healthy calves [28].

Blood glucose concentration is regarded as one of the indicators of energy status in ruminants [34]. The liver is the pre-dominant site of glucose synthesis in the ruminant. Glucose supply is crucial for maintenance and productive functions in growing ruminants [35]. The blood glucose concentration observed in this study which ranged between 73.87 to 96.20mg/dl was higher than the range of (40.00 to 49.75mg/dl) reported by Roy *et al.,* (2016) [2]. But is consistent with the values (70.75 to 77.75 mg/dl) reported by Geiger *et al.,* (2014) in Holstein calves fed varying milk replacers with or without direct-fed microbial supplementation [36]. The high blood glucose observed in this study could be attributed to the treatment effect of soymilk used in the diet of the calves. This increment could also be attributed to the physiological shift in the primary energy source from glucose to volatile fatty acids (VFA) when the rumen in young calves becomes functional [38]. However, the blood glucose levels in this study were within the normal range (40 – 100 mg/dl) reported by (Merck Veterinary Manual, 2016) for healthy calves [28].

The reported range of blood urea nitrogen (BUN) in this study (7.13 to 9.79 mg/dl) was lower than 17.37 to 23.00mg/dl reported by Roy *et al.,* (2016) for calves fed soybeans based milk replacer [2]. The BUN in this study increased significantly (P<0.05) with increased levels of soymilk in the diet. The significant (P<0.05) increase in blood urea nitrogen levels (9.79 mg/dl) obtained in calves fed 50:50 ratio of soymilk and cow milk could be attributed to the high utility value of the diet. Similarly, other workers also demonstrated increased BUN in calves with increase levels of soymilk in the diet [39]. The concentrations of BUN obtained in this study fall within the normal range of 10 - 25mg/dl reported by (Merck Veterinary Manual, 2016) for healthy calves [28]. Higher concentration of BUN is an index of renal dysfunction [39]. However, Harris (2009) reported that BUN may be a better indicator of hydration than renal function, and an elevated BUN would imply dehydration [40]. This study found that calves fed the different ratios of soymilk were not dehydrated because an elevated level of BUN would imply dehydration. Significantly lower ratios denote acute tubular necrosis, low protein intake, starvation or severe liver disease as reported by Hagawane *et al.,* (2009) [35].

Serum creatinine is an important indicator of renal health because it is an easily measured by-product of muscle metabolism that is excreted unchanged by the kidneys [40]. The serum creatinine (0.47 to 0.60 mg/dl) obtained in this study were lower than the values (1.50 to 2.2 mg/dl) reported by Lee *et al.,* (2009) in Holstein calves fed milk replacer containing different amount of energy and protein. It was found that the values of creatinine obtained in this study were within the normal range (0.5 – 2.2 mg/dl) reported for health calves (Merck Veterinary Manual, 2016) [28,37]. This supports the findings of Hammon *et al.,* (2002) who reported that serum creatinine concentration in calves fed high protein milk replacer were within the safe range, suggesting normal liver and renal functions in calves [38].

Conclusion

Soaking of soybeans for 72 hours improved the nutritive value. All the blood parameters measured were within the normal range for healthy calves. Therefore, soymilk can be used to replace cow milk in the diet of calf up to 75% inclusion level without any detrimental effect on health status of the animals. Further study is recommended on the use of other ingredients as milk substitute and to compare the results with this study.

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