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Effect of Keeping Durations Prior to Processing of Bovine Blood on Its Proximate, Gross Energy and Amino Acid Compositions

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

The study was conducted to investigate the effect of keeping durations prior to processing of bovine blood on its proximate composition, gross energy and amino acid profile. Slaughterhouse blood used for this research was obtained from Ntak Inyang Central Abattoir located in Itu Local Government Area, Akwa Ibom State, Nigeria. The actual processing of fresh blood sample into blood meal took place at the Department of Animal Science Laboratory, University of Uyo, Annex. A Completely Randomized Design (CRD) was used. The experiment had four (4) treatments designated as (T_1, T_2 T_3 and T_4), with each differing from one another in keeping durations. Treatment$_1$ contained blood processed after collection at 0 hour serving as the control, T_2 blood was

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processed after 2hours, T_3 blood was processed after 4 hours and T_4 blood was processed after 6 hours. Blood kept for (6 hours) prior to processing was observed to have a significantly higher values ($P<0.05$) for crude protein (37.93%), crude fat (2.57%), crude fibre (0.25%), crude ash (7.91%), moisture (11.19%) and gross energy (2323Kcal/kg). The nitrogen free extract (NFE) values were also significantly, high ($P<0.05$) as the keeping durations prior to blood processing increased. Similarly, amino acid profile for both essential (EAAS) and non essential (NEAAS) revealed that T_4 (6 hours) had higher values ($P<0.05$) than other treatments. In specific terms, tryptophan (6.92%), Leucine (6.91%) and lysine (5.12%) for essential amino acids values were observed to be high in T_4 (6 hours) whereas arginine (5.98%) and glycine (4.10%) for non-essential amino acids also recorded high values. The significant variations in values of parameters (crude protein, gross energy and amino acid profile) as observed in T_4 (6 hours) might be due to the processing methods used in this study as well as the high moisture content potential of the blood meal. In conclusion, bovine blood meal processed after 6 hours of keeping duration is, therefore, recommended for end-users (farmers and feed millers), followed by T_3 (4 hours).

Keywords: Abattoir by-product; keeping durations; processing; bovine blood meal; nutritional content.

1. INTRODUCTION

Livestock sector has remained the major supplier of quality animal proteins, source of employment and dependable income generation opportunities to small, medium and large-scale operators. However, high cost of feeds and feed ingredients are combined negative factors capable of reducing the efficiency and capacity of livestock business, in addition to causing reduced animal protein intake among the populace. The cost of feed ingredients usually ranges from 65-70% of the total cost of production, especially for monogastrics [1] whose sources of animal proteins such as fishmeal in feeds formulations, are often expensive and scarce. This challenge has become a clarion call for animal nutritionists and other critical stakeholders to explore cheaper and alternative animal protein feedstuffs to fishmeal [2]. One of such resources is blood meal.

Blood is one of the abattoir by-products after slaughter and it is widely used in food preparations for human consumption; as a feed ingredient for animals and in a variety of laboratory, medical, industrial and agricultural applications [3]. Blood is a complex connective fluid containing a variety of suspended specialized cells, organic and inorganic substances which circulate in the vessels and capillaries of man and animals [4]. According to Wanasundara et al. [5,3], blood constitutes 3-5% of the live weights of animals, and approximately 50% of this can be collected at slaughter, with the remainder being retained in the capillary system. Blood is an important index which can be used for ideal physiological, pathological and nutritional diagnosis as well as for management practices in animals [6,7]. Okukpe et al. [8] reported that blood also contains myriad of metabolites and other constituents which provide a valuable medium for clinical and nutritional investigations of animals in feeding trials. As reported by Kececi and Col [9], blood chemistry is known to be influenced by several factors such as age, sex, breed, diet, disease, body condition, quality and quantity of feeds, levels of anti-nutritional factors, climate and season.

Several studies have revealed that blood and blood products that are used as food sources for both humans and animals are generally obtained from bovine and porcine species, as the use of blood from other sources is rate [3,10]. Bovine blood has been reported to consist of 80.9% water, 17.3% proteins, 0.23% lipids, 0.07% carbohydrates and 0.62% minerals [3,11]. Given the multiple nutritional, economical and environmental benefits derivable from processing blood into blood meal, such blood must be extracted from healthy animals and must be as safe as possible [12]. In addition, a closed draining system has often been suggested for the collection of blood after slaughter, although local operators are used to open draining system that encourage contact with microbial loads through carcass surface, wash water, vomit, faeces and urine [3]. According to Hardy [13], blood meal is a dry product processed from clean, fresh animal blood with no extraneous materials. Blood meal has a minimum content of 85% crude protein and it is very rich in lysine, methionine, histidine, arginine, leucine and tryptophan but low in isoleucine and glycine [1,14,15], whereas in contrary study, [16] and [13] reported on the low content of arginine and methionine in blood meal. It is also rich in zinc

and iron (more than 1500mg/kgDM) but low in calcium [17]. Blood meal is also reported to contain mostly proteins (about 90-95% DM), small amount of fat (less than 1% DM) and ash less than 5 %DM [17]. Compared with other animal protein sources, blood meal has a poor amino acid balance, and with low isoleucine content, diets for monogastrics must be formulated to contain enough isoleucine for the level of performance expected [17]. As posited by Anoh and Akpet [1], amino acid imbalance may have been the cause of reduced performances in early studies reported in the literature.

Studies have also reported that processing methods could be a serious factor in determining the percentage content of available amino acids, especially lysine in blood meal [18]. According to Mulik [17], the processing methods of drying the blood from slaughtered animals are known to affect the nutritional quality of the proteins in the blood meal. Mulik [17] also specified three (3) methods of processing including batch dry rendering, ring dried rendering and spray dried rendering, and attested to the potentials of both ring dried and spray dried blood meals in achieving greater content of total and available amino acids, with emphasis that the availability of lysine as a percentage of the total lysine is 84-89% as applicable to the ring dried blood meal as compared to 62-77% lysine from batch dried blood meal.

The aforementioned processing methods further sub-classified [17,18] into solar drying, oven drying, drum drying and flash drying, and harped on the efficiency of some methods in ensuring higher protein quality than the solar drying. Thus in principles, drying operates on an inverse relationship between the amount of heat applied and protein digestibility. For instance, lysine content and availability has been reported to decrease when the amount of heat increases of Batterham et al. [19].

Blood meal has been reported to be unpalatable especially when overcooked; for this reason, about 3-5% inclusion level is recommended for feed formulation in poultry in particular and in general, the outcome of blood meal varies depending on the amount added to the feed [20], but pertaining to ruminants, blood meal is mostly used as by-pass protein ingredients diets [16,20]. It also possess dark brown colour, characteristic smell and exhibits hydroscopic property, and needs to be dried to less than 10-12% moisture and stored in a dry place to prevent deterioration [17]. Additionally, blood meal should be free from adulterants, insects, fungal infestation, any offensive odour as well as spores of *Bacillus anthracis* and *Clostridium spp* [21].

Various reports on transforming whole blood into many food products are well documented by several investigators. Apart from being processed into blood meal for animal feeding production, value-addition is also applicable to food industries that utilize it for food additives such as emulsifiers, stabilizers, clarifiers, colourants, gelling agent, nutritional additives, egg albumin substitutes and pharmaceuticals [22,23].

Several studies have also reported on the contamination effects of discharging animal blood into water and land resources due to high content of biological oxygen demand (BOD), chemical oxygen demand (COD) and nitrogen content [23]. For instance, Siti Jamilah et al. [23] reported that the value of COD for blood is 400g per liter, BOD value is 200g per liter while blood contains approximately of 30g Nitrogen per liter. From the environmental management perspectives, the practice of recycling slaughterhouse wastes in the form of blood meal and other products has been considered an alternative solution to minimize harmful environmental hazards that often result from unsystematic blood disposal into land and water resources [22]. While Ofori and Hsieh [3] appraised the practice as having the potentials to minimize dangerous environmental pollution and attendant prohibitive cost of waste management associated with blood disposal into land and water, Lynch et al. [24] supported the practice for reasons of environmental decontamination and innovation to achieve resource utilization and sustainability.

However, despite the numerous benefits obtained from the processing of blood into several products, criticisms and biases from religious and consumers' groups have risen up concerns pertaining to their safe for use by humans and animals, hence highlighting that they may potentially be affected by zoonotic threats such as Hepatitis E Virus (HEV), transmissible spongiform encephalopathies (TES), protein allergens and toxins such as dioxin [10,25]. Nevertheless, this negative concern can be nullified if necessary hygienic practices and scientific applications are adopted in view of today's modernization of slaughterhouse operations while other authors [12] suggested specific cautions against

contamination from the points of bleeding techniques and drainage system employed during blood collection. According to Ofori and Hsieh [3,10], consumers' fear may be allayed with appropriate enforceable policies, one of which is the proper and unambiguous food labelling that indicate the protein source extracted from blood based ingredients as well as boosting consumers' confidence in patronizing these products as safe.

Given the fact that blood is a perishable by-product from abattoir, it needs to be processed within the shortest possible time after slaughter, otherwise physico-biochemical deterioration is inevitable. Therefore, the present study is aimed at assessing the effect of keeping durations on proximate composition, gross energy and amino acid profiles of bovine blood processed after 6 hours.

2. MATERIALS AND METHODS

2.1 Preparation of Blood Meal

Fresh blood was collected from cattle slaughtered at the Central Abattoir, located in Itu Local Government Area, Akwa Ibom State, Nigeria. After collection, the blood was processed by cooking after 0 hour, 2 hours, 4 hours and 6 hours at the Department of Animal Science Laboratory, University of Uyo, Annex. Each treatment was processed for 5 minutes at 100˚C. After cooking, samples were then oven-dried at low temperatures 55˚C for 3 days and then ground into blood meal. Uyo is situated on latitude 5˚17ˡ and 5˚ 27ˡ North of the equator and longitude 7˚27ˡ and 7˚58ˡ East of the Greenwich. Its temperatures ranges between 26˚c to 28˚c, annual rainfall regime ranges 2000m to 3000m and relative humidity ranges between 78% to 93% [26].

2.2 Chemical Analysis of Blood Meal

Dried blood meal sample was analysed in the Laboratory chemically for proximate constituents gross energy [27] and amino acid profile of the bovine blood meal was determined using Ninhydrin chemical reaction, through spectrophotometer determination of amino acid [28].

2.3 Statistical Analysis

Data obtained were subjected to one way analysis of variance (ANOVA) in a completely randomized design [29]. Mean separation was carried out by Duncan Multiple Range Test [30].

3. RESULTS AND DISCUSSION

The values obtained for proximate composition and gross energy for processed bovine blood meal is presented in Table 1 below. There were significant differences ($P<0.05$) among the parameters across the four (4) treatments. Treatment 4 (6 hours) indicated significant differences in values for crude protein (37.93%), crude fat (2.57%), crude fibre (0.25%), crude ash (7.91%), moisture (11.19%) and gross energy (2323kcal/kg) and these parameters were observed to be increasing as the keeping duration increased, nitrogen free extract (NFE) with ranges (35.14 - 40.62) also increased in line with increased keeping duration. Decreased moisture values of this study also are desirable advantageous in reducing microbial population which cannot thrive in a liquid less medium. Although the blood meal processed after 6 hours recorded the highest values for most of the parameters, its crude protein value of 37.93% has differed from the crude protein values of 85%, 80% and 77.1% earlier reported by some authors [14,18,31] while other parameters such as crude fat, crude fibre and crude ash also showed slight variations. These variations in values might be due to the processing methods used, the type and quality of forages available to the cattle from which the experimental blood was obtained in transforming raw blood into blood meal.

According to the literature, different authors are known to adopt different processing methods such as batch dry rendering, ring dried rendering and spray dried rendering, and each of these methods has different percentage values of protein content of blood meal irrespective of the keeping durations, as drying operates on an inverse relationship between the amount of heat applied and protein digestibility [17,18]. The crude protein value obtained in Treatment 4 of the present study was not appreciable owing to the cooking and oven drying methods employed, which have been reported to be less efficient in blood meal processing since both are known to supply greater heat, even within a shorter duration than other methods. Additionally, a method such as the spray dried rendering is reported to retain more nutrients in blood meal after processing [18]. However, the three (3) aforementioned methods are well advanced technologies, expensive and beyond the small-

scale operations when compared to cooking and oven drying method adopted for this present study.

The gross energy value of Treatment 4 (2323Kcal/kg) was also the highest among the treatments. The gross energy level showed that the blood meal processed after 6 hours can be considered as a rich source of energy, and can supply the energy required for livestock including poultry although the outcome of the blood meal varies depending on the amounts added to the feed [20]. Being hydroscopic in nature, the blood meal tends to have more gross energy values as the keeping duration increased due to the concentration or saturation effect from water loss. Therefore, the values are in agreement with the earlier report [17].

The results of essential amino acid composition of processed bovine meal are shown in Tables 2 below. There were significant differences ($P<0.05$) among bovine blood meals for essential amino acids (EAA) whose values obtained were significantly ($P<0.05$) higher than others particularly for blood meal processed 6 hours after collection. For blood meals processed after 6 hours. Methionine, histidine and isoleucine are in limited or short supply by blood meal as observed in this study research. Therefore, they are arranged in order of 1st, 2nd and 3rd limiting amino acid respectively, though this finding does not totally agree with some reports [18,31,32] who stated that isoleucine is the 1st limiting acid, followed by methionine as the 2nd limiting acid. It is also observed that tryptophan, leucine and lysine are in high supply in T_4, with the value of 6.92%, 6.91% and 5.12% respectively.

According to Ravindran et al. [33], blood meal has high tryptophan digestibility coefficient which is valuable as this particular essential amino acid is the 3rd limiting amino acid in broilers. In addition, the essential amino acid values from

Table 1. Proximate composition and gross energy of blood meal processed after 6 hours keeping duration

Parameters	T_1	T_2	T_3	T_4	
	(0 hour)	(2 hours)	(4 hours)	(6 hours)	SEM
Crude protein (%)	23.53^{d}	23.74^{C}	37.63^{b}	37.93^{a}	2.13
Crude fat (%)	1.59^{d}	1.64^{C}	2.51^{b}	2.57^{a}	0.14
Crude fibre (%)	0.07^{d}	0.05^{C}	0.22^{b}	0.25^{a}	0.03
Crude Ash (%)	5.91^{d}	5.84^{d}	7.83^{b}	7.91^{a}	0.30
Moisture (%)	33.76^{d}	33.84^{a}	11.19^{c}	11.14^{d}	3.41
NFE (%)	35.14^{d}	34.89^{c}	40.20^{b}	40.62^{a}	3.60
Gross Energy (Kcal/kg)	1197^{d}	1204^{c}	2317^{b}	2323^{a}	0.17

$^{a-d}$Means on the same row with different superscripts are significantly ($P<0.05$) different. SEM: Standard Error of Means. T_1 = Blood processed after 0 hour of collection, T_2 = Blood processed after 2 hours of collection, T_3 = Blood processed after 4 hours of collection and T_4 = Blood processed after 6 hours of collection

Table 2. Essential amino acid profile (%) of blood meal processed after 6 hours keeping duration

Parameters	T_1	T_2	T_3	T_4	
Essential Amino Acids	(0 hour)	(2 hours)	(4 hours)	(6 hours)	SEM
Alanine	1.83^{d}	1.95^{c}	4.00^{b}	4.24^{a}	0.34
Histidine	1.36^{d}	1.43^{c}	2.81^{b}	3.05^{a}	0.23
Isoleucine	1.24^{d}	1.37^{c}	2.72^{b}	3.06^{a}	0.24
Leucine	3.26^{d}	3.57^{c}	6.09^{b}	6.91^{a}	0.47
Lysine	2.12^{d}	2.22^{c}	4.74^{b}	5.12^{a}	0.42
Methionine	0.96^{d}	1.02^{c}	1.53^{b}	1.77^{a}	0.10
Phenylalanine	1.16^{d}	1.27^{c}	3.51^{b}	3.71^{a}	0.36
Threonine	1.75^{d}	1.86^{c}	3.61^{b}	3.82^{a}	0.29
Tryptophan	2.77^{d}	2.91^{c}	5.67^{b}	6.92^{a}	0.54
Valine	2.29^{d}	2.45^{c}	4.84^{b}	5.80^{a}	0.46

$^{a-d}$Means on the same row with different superscripts are significantly ($P<0.05$) different. T_1 = Blood processed after 0 hour of collection, T_2 = Blood processed after 2hours of collection, T_3 = Blood processed after 4 hours of collection and T_4 = Blood processed after 6 hours of collection

Table 3. Non-Essential amino acid profile of blood meal processed after 6 hours keeping duration

Treatments	T_1	T_2	T_3	T_4	
Non- Essential Amino Acids (%)	**(0 hour)**	**(2 hours)**	**(4 hours)**	**(6 hours)**	**SEM**
Arginine	2.07^{d}	2.17^{c}	4.87^{b}	5.98^{a}	0.51
Aspartic acid	3.80^{d}	4.08^{c}	7.91^{b}	8.54^{a}	0.65
Cystine	0.14^{d}	0.21^{c}	0.22^{b}	0.46^{a}	0.04
Glutamic acid	5.69^{d}	5.76^{c}	10.16^{b}	11.02^{a}	0.74
Glycine	1.32^{d}	1.45^{c}	3.90^{b}	4.10^{a}	0.39
Ornithine	0.02^{d}	0.06^{c}	0.14^{b}	0.21^{a}	0.22
Proline	0.94^{d}	1.04^{c}	2.02^{b}	2.21^{a}	0.17
Serine	1.79^{d}	1.93^{c}	3.76^{b}	3.92^{a}	0.30
Tyrosine	1.11^{d}	1.17^{c}	2.23^{b}	2.40^{a}	0.18

*$^{a-d}$ Means on the same row with different superscripts are significantly ($P<0.05$) different. **SEM**: Standard Error of Means. T_1 = Blood processed after 0 hour of collection, T_2 = Blood processed after 2hours of collection, T_3 = Blood processed after 4 hours of collection and T_4 = Blood processed after 6 hours of collection*

this study are adequate, compared to the recommendation [14] for poultry species. This observation might be due to high moisture retention which was above the normal range of 10-12% [17] before processing and its effect is reported to enhance low retention of available nutrients, especially amino acids.

Essential amino acid values from this study were within normal ranges compared to NRC [14] but lower than those reported by other authors [18, 31. Nevertheless, essential amino acid profile revealed that bovine blood meal (BBM) processed after 6 hours keeping duration enhances availability of limiting amino acids especially lysine (5.12%) and methionine (1.77%), both which can be compared with other animal protein sources such as fishmeal whose lysine value stands at 4.5% and methionine value at 1.80% while meat meal contains lysine value of 2.60% and methionine value of 0.75% [14,15], hence the beneficial effect of increased keeping durations has resulted in improved nutrient availability, particularly the limiting amino acids, owing to the principle of high moisture retention in blood processed after 6 hours.

Blood meal, as observed in this experiment, is rich in non-essential amino acids, though required in low quantities in diets of farm animals [14]. Non-essential amino acids, like the essentials, increased along the row as time or duration increased. The range of values of glycine (1.32 - 4.10%) and tyrosine (1.11% - 2.40%) observed in this study does not agree with the glycine value of 4.6g/100g and tyrosine value of 3.2g/100g earlier reported [17]. Even though the value of tyrosine (2.07%) reported by Olukayode et al. [31] agrees with results obtained from this study, the glycine value (4.59%) does not agree. Although, the value of arginine at 2.35g/100g and 3.34% reported by both authors falls within the range of 2.07% - 5.98% as observed in this study.

4. CONCLUSION AND RECOMMENDATION

Treatment that contained bovine blood meal processed after 6 hours of collection was the best as it had reasonable nutrient contents, especially the limiting amino acids, lysine and methionine. Therefore, further investigation is required to determine and compare the keeping durations of flesh blood and its nutritional contents prior to processing beyond 6 hours. If need be, the microbial effects of keeping duration of the blood should also be assessed.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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