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Research Paper

Effects of sesame meal substitution on carcass parameters and meat quality of growing male lambs

Ata M and Obeidat BS.

Online J. Anim. Feed Res., 15(3): 126-131, 2025; pii: S222877012500015-15 DOI: <u>https://dx.doi.org/10.51227/ojafr.2025.15</u>

Abstract

This study was established to determine how Awassi male lamb carcass and meat quality features would change if soybean meal were substituted with sesame meal. Twenty-four lambs started with 15.7 \pm 0.33 kg BW were chosen and allocated randomly to two dietary treatments; the 0% sesame meal (CON diet) or the 12.5% sesame meal (SM12.5 diet). Lambs were placed in experimental pens separately that



crossref

equipped with plastic waterers feeders to allow free access to diets and water throughout the experiment. Lambs were slaughtered at day 84 for measuring carcass traits and meat quality. Fasted and carcass weights were measured as dressing percentage was calculated. Non-carcass parts were separated from the carcass and weighed. Carcass features examination included measuring carcass linear dimensions, leg cuts and longissimus dorsi muscle characteristics. Meat quality was evaluated after two weeks for color (L*a*b* co-ordinates), pH, water holding capacity, values of shear force and cooking loss. Lambs consumed SM12.5 had more (P < 0.05) fasting live weight (kg), and weights of hot and cold carcasses compared to CON group. Other carcass measurements were not affected by the SM inclusion (P > 0.05). Similarly, meat quality parameters did not differ between the two dietary treatments. Therefore, according to these findings, feeding SM to Awassi lambs would not have an adverse impact on the quality of their meat or carcass characteristics. However, performance was enhanced as the fasting weight, as well as hot and cold carcass weight was improved.

Keywords: Awassi lambs, Carcass Characteristics, Dietary treatments, Meat Quality, Sesame meal.

[Full text-PDF]

Research Paper

Prevalence and economic consequences of umbilical lesions in livestock farming

Ayvazoğlu Demir P, Aydin E, Yildiz U, Aydin U, and Aksoy Ö.

Online J. Anim. Feed Res., 15(3): 132-139, 2025; pii: S222877012500016-15 DOI: <u>https://dx.doi.org/10.51227/ojafr.2025.16</u>

Abstract

The aim of this study is to determine the incidence of umbilical lesions in calves within livestock enterprises and to assess the economic losses they incur. The analysis is based on clinical data from 815 umbilical lesion cases recorded over a 17-year period, supplemented by findings from a Delphi survey conducted with expert veterinarians. The results indicated that umbilical lesions predominantly occur in March and April, and 53% of affected calves are younger than one month old. It was observed that 72.8% of umbilical lesions occurred in Simmental calves. Additionally, umbilical urachus fistula was more commonly diagnosed in females, while omphalitis was more prevalent in males. The incidence rate of umbilical lesions was calculated at 38.6%, with a surgical success rate of 73.9%. Postoperative complications were observed in 28.9% of



cases, and 53.8% of the calves were culled due to growth retardation or further complications. The estimated cost of surgical intervention and postoperative treatment for umbilical lesions was determined to be \$101.3 per calf. However, this cost increased to \$245.6 in cases complicated by postoperative complications and developmental delays. Despite the fact that umbilical lesions can be largely prevented through simple postnatal hygiene measures, their incidence remains high, leading to substantial economic losses in Türkiye.

Keywords: Calves, Delphi survey, Economic losses, Hygiene measures, Postnatal infections, Surgical intervention.

[Full text-PDF]

Research Paper

Response of heat stressed broilers to ameliorating effects of synthetic vitamin C and *Citrus sinensis* extract

Ayoola MO, Oladejo OA, Adeleye BE, Oguntunji AO, Alabi OM, Lawal TE and Aderemi FA.

Online J. Anim. Feed Res., 15(3): 140-149, 2025; pii: S222877012500017-15 DOI: <u>https://dx.doi.org/10.51227/ojafr.2025.17</u>

Abstract

Heat stress is a critical environmental factor in the poultry industry, negatively impacting performance and causing economic losses. High temperatures reduce productivity and increase mortality. This study used 120 Abor Acre broilers (28 days old, 1.050 ± 0.4 kg) fed standard commercial feed. At day 35, 30 birds were randomly assigned to 4 treatments: T₁ (water as control), T₂ (30 mg/L synthetic vitamin C based on manufacturers' dosage), T₃ (30 ml/L orange extract), and T₄ (50 ml/L orange extract), each in three replicates. Birds were exposed to artificial heat stress (36 ± 2°C) for 15 days using an automated heater, with unrestricted access to feed and treatment solutions. Physiological data were recorded periodically, and blood samples were collected on days 0, 5, 10 and 15 for serum biochemical analysis. Data were analyzed using



the SAS procedure for repeated and non-repeated measures. Results showed significantly higher final body weight (FBW) and average body weight gain (ABWG) in T_4 compared to other groups (P < 0.05). Average daily feed intake (ADFI) was highest in T_4 chickens, and feed conversion ratio (FCR) was lowest, indicating superior feed efficiency. Rectal temperature, body temperature, pulse rate, and respiratory rate were significantly highest in T_1 group (P < 0.05). Serum biochemical markers (AST, ALT, glucose, and creatinine) were highest in T_1 , whereas T_4 recorded the lowest values. T_1 broilers had the highest external and internal thermal gradients, while T_4 had the lowest. Glycogen reserves were highest in T_4 group. The study concludes that sweet orange extract, especially at a 50% concentration, is a viable organic alternative to synthetic vitamin C (30 mg/L) for mitigating heat stress in broilers. It enhances growth performance, feed efficiency, and physiological stability under heat stress, making it a sustainable solution for poultry production in tropical climates.

Keywords: Broiler, Heat stress, Serum, Sweet orange extract, Vitamin C.

[Full text-PDF]

Research Paper

Effect of probiotic supplement on growing lambs of the lacon breed during the suckling period

Tsvihun A, Karatieieva O, Ponko L, Yakovchuk V, Yulevich O and Ponichtera P.

Online J. Anim. Feed Res., 15(3): 150-158, 2025; pii: S222877012500018-15 DOI: <u>https://dx.doi.org/10.51227/ojafr.2025.18</u>

Abstract

Dairy lambs are susceptible to many pathogens that can affect their subsequent performance. The use of probiotics in the rearing of Lacon lambs during the suckling period has been identified as a means of maintaining intestinal microbial balance. Studies have shown that probiotics can be used as an alternative therapy that prevents the use of antibiotics and, thus, can reduce the emergence and spread of antibioticresistant bacteria as well as antibiotic residues in dairy products, meat and milk. In addition, it increases the growth rate of the animal and its stress resistance under various conditions. In this regard, we conducted a comprehensive assessment of the impact using the therapeutic and probiotic supplement Immunobacterin-D on the growth and development of Lacon dairy lambs and their clinical parameters during the suckling



period. It was found that the use of the probiotic supplement Immunobacterin-D during the suckling period of Lacon dairy lamb provided an increase in average daily gains by 16.1%, an increase in the content of total protein by 15.8% and an increase in the level of globulin proteins by 35.4%, which indicates a high resistance of the lambs' organism. Thus, the use of the probiotic Immunobacterin-D in feeding lambs had a positive effect on their growth rate, development, improved their health and adaptive capacity, as they showed greater resistance to elevated ambient temperatures compared to those that did not receive the supplement. The use of the probiotic increases the stress resistance of lambs, which can be used as one of the effective approaches in the breeding system of Lacon sheep.

Keywords: Breeding system, Gastrointestinal microbiota, Intestinal microbial balance, Sheep, Stress resistance.

[Full text-PDF]

Determination of brewery yeast-treated crop residues as animal feed resource

Getachew M, Amare T, and Yimer E.

Online J. Anim. Feed Res., 15(3): 159-167, 2025; pii: S222877012500019-15 DOI: <u>https://dx.doi.org/10.51227/ojafr.2025.19</u>

Abstract

The study evaluates the effects of brewery yeast to improve the nutritional value and in vitro digestibility of selected crop residues in Dessie Town, Amhara region, Ethiopia. The experiment was laid out in a completely randomised design (CRD) with a 3×5 factorial arrangement, i.e., three-selected crop residues (Teff straw, maize, and sorghum stover) with five levels of fermentation periods (0, 3, 6, 9, and 12 days). The mean crude protein (CP) values of brewery-spent yeast (BSY) treated Teff straw, maize stover, and sorghum stover treated crop residues were 7.85%, 14.3%, and 14.78%, respectively. Among proximate and detergent values of the interaction effect of fermentation period and crop residue type, dry matter (DM), crude protein (CP), estimated metabolizable energy (EME), acid detergent lignin (ADL), and



neutral detergent fiber (NDF) content were significantly (p < 0.01) varied at different fermentation periods. While ash, organic matter (OM), and acid detergent fiber (ADF) content were highly significant (p < 0.001) variations between the interaction of crop residues and the fermentation period. The value of in vitro dry matter digestibility (IVDMD) and in vitro organic matter digestibility (IVOMD) of BSY treated crop residue was significantly (p < 0.001) varied for the interaction of crop residue type and fermentation period. The EME, IVDMD, and IVOMD of BSY treated crop residues were observed in the range of 7.54-10.64 MJ/kg, 54.44%-61.46%, and 47.14%-66.48%, respectively. Therefore, the study investigates how brewery yeast can enhance the nutritional quality and in vitro digestibility of certain crop residues in Dessie Town, Ethiopia. It followed a random design with three crop residues (Teff straw, maize stover, and sorghum stover) and six fermentation periods. Treated residues showed varying crude protein values and significant differences in in vitro digestibility and energy content. Therefore, utilizing brewery-spent yeast (BSY) presents a cost-effective and sustainable approach to enhancing the nutritional value and in vitro digestibility of crop residues. Further studies may explore the scalability of BSY treatments and their effects on livestock performance.

Keywords: Brewery spent yeast, Crop residue, In vitro digestibility, Nutritional value.

[Full text-PDF]

Research Paper

The effect of nitrogen and sulfur in making oncom of cassava pulp by *Neurospora sitophila* and its impact on in vitro digestibility and fermentability in sheep diet

Hernaman I, Dhalika T, Tanuwiria UH, Ayuningsih B, Budiman A, Hidayat R, Rosani U, Mutaqin BK, Nugraha MR, and Ismiraj MR.

Online J. Anim. Feed Res., 15(3): 168-174, 2025; pii: S222877012500020-15 DOI: <u>https://dx.doi.org/10.51227/ojafr.2025.20</u>

Abstract

This study focused on the modification of oncom making based on cassava (*Manihot esculenta*) pulp to improve the nutritional profile through fermentation by *Neurospora sitophila* and enrichment with a mixture of urea (0, 2.5, 5, 7.5, and 10% of dry matter) and a nitrogen:sulfur ratio of 15:1 (oncom cassava pulp= OCP). Hence an investigation was carried out on the *in vitro* digestibility and fermentability of OCP when included in sheep diets. *In vitro* evaluation (diet oncom cassava pulp; DOCP) was carried out on diets consisted of a mixture of 50% grass and 50% concentrate containing 2.5%OCP (which selected based on the highest analysed crude protein and crude fiber levels when compared to the nutrient contents of other enriched OCP levels, P < 0.001) with compositions of 0, 10, 20, and 30% of DOCP.



Hememan I, Dhalika T, Tanuwina UH, Ayuningah B, Budiman A, Hidayat R, Rosani U, Mutagin BK, Nugraha MR, and IaminajMR (2025). The effect of nitrogen and sulfur in making oncom of casasra pulp by *Neurospore attophila* and its impact on an vitro digestibility and fermentability in sheen did: Oxing J dom End (2017). PCN International International Statement (2017).

Results showed that the higher use of urea and nitrogen:sulfur ratio caused a decrease in crude fiber and gross energy and an increase in nitrogen-free extract (NFE) levels (P < 0.001). Using 2.5%OCP in the diet resulted in significantly different N-ammonia (N-NH3), volatile fatty acid (VFA), in vitro dry matter digestibility (IVDMD), and in vitro organic matter digestibility (IVOMD) (P < 0.05), except for total gas and ruminal pH values. The highest IVDMD and IVOMD were obtained in 30% DOCP with a close N-NH3 and VFA and within the normal range. In summary, CP fermentation by *Neurospora sitophila* which is enriched with a mixture of 2.5% urea and nitrogen:sulfur at a 15:1 ratio can be used in sheep diet up to 30%.

Keywords: Chemical additives, Feed, Fermentation, Neurospora sitophila, Rumen.

Review

Estimation of body weight from biometric traits of chickens using regression statistical method: a systematic review

Tsenane JA, Tyasi TL, and Tada O.

Online J. Anim. Feed Res., 15(3): 175-185, 2025; pii: S222877012500021-15 DOI: <u>https://dx.doi.org/10.51227/ojafr.2025.21</u>

Abstract

Live body weight is a vital tool when placing price on the chickens for profit and to assess decisions to be made in selection of animals and other husbandry practices. However, the chicken farmers, especially the resource limited farmers, lack weighing scales to perform these animal husbandry practices. The objective of this study was to systematically review the articles published on the estimation of live body weight from biometric traits of chickens using regression statistical methods. Databases such as Google Scholar, ScienceDirect, PubMed, and Web of Science, with the combination of the following keywords: "Body weight" or "body mass", "biometric traits" "zoometric measurements" or "Morphological traits" or "linear body measurements" or "Morphometric



traits" or "body parameters" or "growth traits" or "growth performance traits", chicken or poultry or "*Gallus gallus domesticus*" or fowl, regression. There were limited articles that aimed to predict live body weight using regression statistical method across the world. A total of fourteen articles were published between the years 2009 and 2024. The results indicated that stepwise linear regression method was mostly used by 39% of the articles included, followed by multiple regression method and simple linear regression method by 22% of the included articles each. The results indicated that the highest coefficient of determination (R2 = 0.970) was recorded on the model of combination of body length (BL) and breast length (BRL) using multiple linear regression method on chickens. The limitation is that some articles did not include either sex or age of the animals that can make it difficult to make conclusion for different sexes and ages. This systematic review concludes that the multiple linear regression statistical method is the best in estimating live body weight in chickens with combination of BL and BRL. As a practical suggestion, it would be best for breeders to select chickens with the highest measurements of BL and BRL to improve the live body weight of the chickens.

Keywords: Biometric traits, Body weight, Chicken, Linear body measurements, Regression.

[Full text-<u>PDF</u>]

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EFFECTS OF SESAME MEAL SUBSTITUTION ON CARCASS PARAMETERS AND MEAT QUALITY OF GROWING MALE LAMBS

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Supporting Information

ABSTRACT: This study was established to determine how Awassi male lamb carcass and meat guality features would change if soybean meal were substituted with sesame meal. Twenty-four lambs started with 15.7 \pm 0.33 kg BW were chosen and allocated randomly to two dietary treatments; the 0% sesame meal (CON diet) or the 12.5% sesame meal (SM12.5 diet). Lambs were placed in experimental pens separately that equipped with plastic waterers feeders to allow free access to diets and water throughout the experiment, Lambs were slaughtered at day 84 for measuring carcass traits and meat quality. Fasted and carcass weights were measured as dressing percentage was calculated. Non-carcass parts were separated from the carcass and weighed. Carcass features examination included measuring carcass linear dimensions, leg cuts and longissimus dorsi muscle characteristics. Meat quality was evaluated after two weeks for color (L*a*b* co-ordinates), pH, water holding capacity, values of shear force and cooking loss. Lambs consumed SM12.5 had more (P < 0.05) fasting live weight (kg), and weights of hot and cold carcasses compared to CON group. Other carcass measurements were not affected by the SM inclusion (P > 0.05). Similarly, meat quality parameters did not differ between the two dietary treatments. Therefore, according to these findings, feeding SM to Awassi lambs would not have an adverse impact on the quality of their meat or carcass characteristics. However, performance was enhanced as the fasting weight, as well as hot and cold carcass weight was improved.



INTRODUCTION

Towards meeting their demands for energy, crude protein, and other nutrients, sheep are fed a variety of conventional feed ingredients. Although the price of these ingredients is significant (about 70% of ruminants' production cost), which is not represented in the sale cost of these sheep, therefore affects the livestock holders' profits (Obeidat and Gharaybeh, 2011). Several studies that have evaluated the use of non-conventional feed in sheep diets found that by-products reduced production costs, which in turn increased productivity and returned on in animal production investments (Ata and Obeidat, 2020; Ominski et al., 2021).

One of the significant byproducts of sesame seed oil production is sesame meal (SM). Sesame by-products which produced annually reached 1 million tons worldwide (Weiss, 2000) and about 3600 tons of SM in Jordan as reported by the Ministry of Agriculture (2017).

Sesame meal is high in crude protein content (approximately 46% CP) as reported by Obeidat et al. (2019); sesame meal (SM), on the other hand, could be attained after the extraction of oil from sesame seed by using compressing techniques. According to other researchers, SM chemical composition is impacted by how it is processed (Sá et al., 2022). Additionally, the sesame seeds' dry matter content increased and their moisture content declined as a result of the roasting techniques utilized during processing (Salamatullah et al., 2021). On the other hand, if a lot of tiny sesame seeds escape when being hulled, it might indicate a higher fat content (Bonos et al. 2017). The plant variety and the method of oil withdrawal are frequently linked to variations in the fat and protein content of oilseed byproducts (Elleuch et al., 2007).

Sesame by-products, as reported by researchers, showed an improvement in small ruminants' performance lacking any negative outcome on carcass and meat composition (Hassan et al., 2013; Ghorbani et al., 2018; El-Tanany et al., 2021). The study hypothesis was that feeding SM by partially replacing soybean meal will improve lamb performance, carcass, and meat characteristics. Since few studies has been conducted regarding the effect of feeding SM on Awassi lambs' carcass and meat quality traits, this current research was carried out to investigate how replacing soybean meal with SM would affect Awassi lamb's carcass traits and quality of meat.

MATERIALS AND METHODS

Ethical approval

The methodology and guidelines followed in this experiment was authorized by the Animal Care and Use Committee Institution at the Jordan University of Science and Technology in the current search before to its start (Research study number for ethical approval: 16/04/12/39AAB; Deanship of research; Jordan University of Science and Technology, 2025). The committee guidelines were derived from the animal welfare well-established concepts known as the three Rs; Replace, Reduce, Refine; which reflect specify principles and considerations that can be used as tools when balancing between harm and benefit while using experimental animals during the research period (Curzer et al., 2015).

Study procedures and sample analysis

The current study all set of methods were described by Obeidat et al. (2022). Briefly, twenty-four Awassi male lambs body weight (BW) equivalent to 15.7 ± 0.33 kg were divided evenly between the two treatments in random order (12 animal per treatment; N=12). Both the control diet (CON), which consisted of 0% sesame meal, and the diet (SM12.5), which contains 12.5% sesame meal, were used as treatments. Diets were combined to ensure that lambs received their required nutrients as they were formulated to be isonitrogenous (crude protein (CP) content was designed to be 15.6% of dietary DM) (Table 1).

The lambs were weighed, their ears were marked with plastic tags, and a veterinarian checked them to verify they were healthy and clear of diseases before the trial began. Separate cement pens measuring 1.5 by 0.75 meters were used for the lambs to be housed in as each pen provided with plastic feeders (10 L) and waterers (7 L). Lambs had free access throughout the experiment to water and diets.

Every two weeks during the trial, the feed was mixed, and samples were collected to determine its chemical composition. The trial was conducted for 84 days, of which the first week was spent to get lambs acquainted to their pens and feed while the other 77 days were used for data assortment.

	Diets 1	CON	SM40 5	CM
Item		CON	51112.5	5111
Ingredients (% DM)				
Barley grain, whole		47	45.5	-
Soybean meal, 440 g/kg CP (solvent)		21	10	
Sesame meal		0	12.5	-
Wheat straw		30	30	
Salt		1	1	-
Limestone		0.9	0.9	-
Vitamin-mineral premix ²		0.1	0.1	
Feed cost/ton (US\$) ³		418	375	
Nutrients (% DM)				
Dry matter		90.3	90.5	93.9
Crude protein		15.6	15.6	41.5
Neutral detergent fiber		30.0	30.5	12.6
Acid detergent fiber		20.3	19.88	5.3
Ether extract		1.7	3.45	14.5
Metabolizable energy, Mcal/kg ⁴		2.28	2.37	3.53

Table 1 - Ingredients and chemical composition of diets-containing sesame meal (SM) fed to Awassi lambs.

vitamin D3, 200,000 IU; vitamin E, 75 mg, vitamin K3, 200 mg; vitamin B1, 100 mg; vitamin B5, 500 mg; lysine 0.5%; DL-methionine, 0.15%; manganese oxide, 4000 mg; ferrous sulphate, 15,000 mg; zinc oxide, 7000; magnesium oxide, 4000 mg; potassium iodide, 80 mg; sodium selenite, 150 mg; copper sulphate, 100 mg; cobalt phosphate, 50 mg, dicalcium phosphate, 10,000 mg. ³ Calculated based on the prices of diet ingredients of the year 2022. ⁴ Estimated based on tabular values of NRC (2007).

Slaughtering procedures, carcass, and meat quality evaluation

All lambs at day 84 were slaughtered for carcass and meat characteristics measurements at the Agriculture and Production facilities Center at the university. Slaughtering procedure followed in this study are documented by Abdullah and Musallam (2007); whereas lambs were handled and slaughtered by trained personnel approximately after 18 hours of fasting. Live weight was recorded before starting the procedures followed by recording hot carcass weight directly after slaughtering while after a full day of chilling carcass at 4°C the weight was documented. Calculating the dressing percentage of all carcasses performed through dividing the carcass weight (jold) by animal live weight (fasted). Immediately after slaughtering, non-edible parts were separated from the carcass and weighed. Subsequently 24 hours later, linear dimensions were recorded following the slaughtering procedure using the chilled carcass parts. Carcasses were dissected to four parts (loin, rack, shoulder, and leg cutes) to be examined. Two weeks before starting the meat quality evaluation, the loins' cut longissimus dorsi muscle was separated and kept in vacuum-packed at 20 °C.

Cooking loss, shear force, water holding capacity (WHC), color values (CIE L*a*b* coordinates), and pH values were all measured as indicators of meat quality. All meat quality parameters were measured following the procedures of Abdullah and Musallam (2007). In a fridge set at 4°C, frozen longissimus dorsi muscles were allowed to defrost overnight whereas still in their plastic bags. The pH was measured after thawing and muscles were divided into slices of particular thickness to be used for meat quality measurement. Allowing all slices to be oxygenated for 2 h at 4 °C, slices were spread on a tray and coated with a porous cloth. Slices color was measured (slices thickness were 15-mm). For Cooking loss (CL) measurements, slices with 25-mm thick were used, which were evaluated before being cooked, sealed in bags made of plastic, and cooked for 1 hour and 30 min in a 75 °C water bath to be re-weighed for determine the amount of water lost. To determine shear force values, cooked slices were split into 6 smaller samples (cores) with size of 1 cm3 and left at 4 °C overnight. With Warner-Bratzler (WB) shear blade (Model 235), cooked meat cores were sheared using the triangular cutting slot placed on the Salter, parallel to the muscle fiber direction for calculating the force (kg) needed to shear the cores. Evaluating the WHC performed with the technique outlined by Grau and Hamm (1953) and it was described as follow: WHC % = (initial weight – final weight) × 100/initial weight.

Statistical analysis

Data was examined using SAS PROC MIXED methods for analysis, with considering diet as the fixed effect (SAS version 8.1, 2000, SAS Institute Inc., Cary, NC) and individual lambs were the random variable. The probability of rejecting a false null hypothesis, was calculated as $1-\beta$ (1 - Type II error probability) as α level is 0.05. Least square means separation was performed by using Tukey test and significance level was determined at p<0.05.

RESULTS

Sesame meal, as well as, the experimental diets were chemically analyzed as presented in Table 1. Sesame meal shown to be rich in CP content. The ME energy and EE content were high in the SM (3.53 Mcal/kg, 14.5%; respectively). The proximate analysis for the experimental diets showed similar DM, CP, and neutral detergent fiber (NDF) content, while diet containing sesame meal (SM12.5) had greater EE and ME content compared to the control diet (CON). Feed cost (US\$/ton) was calculated based on the ingredients prices and was less for diet containing SM compared to CON (375 US\$/ton, and 418 US\$/ton; respectively).

The effect of SM on lambs' carcass traits were summarized in Table 2. live weight, carcass weight (hot and cold) was greater (P < 0.05) for lambs consumed SM12.5 compared to CON group. Dressing percentage, non-carcass parts, cut weight, tail fat, as well as, dissected leg features did not change (P > 0.05) by the presence of SM to the diet. Carcass leaner dimensions were not affected (P > 0.05) also by sesame meal addition as illustrated in Table 3.

Meat quality characteristics were slightly changed with SM inclusion as presented in Table 4. Meat pH was similar (P > 0.05) between the two diets. Meat color coordinates were similar (P > 0.05) regarding the whiteness, redness, and yellowness.

	Diet ¹	CON	SM12.5	SEM4	P value
Item		(n = 12)	(n = 12)	CEM	, value
Fasting live weight (kg)		31.11 ^b	33.18 ^a	0.700	0.0397
Hot carcass weight (kg)		14.63 b	15.84 a	0.377	0.0359
Cold carcass weight (kg)		13.93 b	15.05 a	0.231	0.0442
Dressing percentage		44.68	45.30	0.870	0.6167
Non-carcass components (kg) ²		1.39	1.45	0.038	0.3366
Carcass cut weights (kg) ³		12.33	13.06	0.303	0.1205
Fat tail (kg)		1.67	1.73	0.129	0.7205
Leg weight (g)		2220	2297	73.4	0.4346
Subcutaneous fat (g $/100$ g)		12.2	11.4	0.70	0.4466
Intermuscular fat (g $/100$ g)		1.9	2.1	0.10	0.2570
Total fat (g / 100 g)		14.1	13.5	0.71	0.5595
Total meat (g/ 100 g)		55.8	55.8	1.14	0.9931
Total bone (g / 100 g)		22.3	22.4	0.62	0.8456
Meat to bone ratio		2.51	2.52	0.080	0.8800
Meat to fat ratio		4.15	4.21	0.241	0.8707

Table 2 - Effects of sesame meal (SM) on carcass, non-carcass components, dissected leg carcass cut weights and percentages of Awassi lambs.

¹ Diets were: the control diet (CON) or 12.5% SM (SM12.5) of dietary dry matter (DM). ² Non-carcass components (Heart, liver, spleen, kidney, and lungs and trachea). ³ Carcass cut (shoulder, racks, loins, and legs). ⁴ SEM = Standard error of the mean; ^{a,b} within a row means without a common superscript difference (P < 0.05).

Table 3 - Effects of feedin	g sesame meal (SM) on carcass leane	r dimensions of	Awassi lambs.
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	Diet ¹	CON	SM12.5	SEM4	Byoluo	
Item		(n = 12)	(n = 12)	3EIMI*	r value	
Leg fat depth (L3) (mm)		2.35	2.50	0.241	0.6434	ĺ
Tissue depth (GR) (mm)		8.67	8.50	0.438	0.7884	
Rib fat depth (J) (mm)		1.75	1.90	0.141	0.4684	
Eye muscle width (A) (mm)		49.36	49.24	0.729	0.9083	
Eye muscle depth (B) (mm)		19.76	20.02	0.317	0.5745	
Eye muscle area (cm2)		8.84	9.03	0.325	0.6618	
Fat depth (C) (mm)		1.50	1.50	0.143	1.000	
Shoulder fat depth (S2) (mm)		1.30	1.40	0.158	0.6643	
1 Diets were: the control diet (CON)	or 12 5% SM (SM12 5) of	dietary dry matter (DN	1) 2 SEM = Standard	error of the mean		

Table 4 - Effects of feeding sesame meal (SM) on meat quality characteristics of Awassi lambs.

	Diet ¹	CON	SM12.5	CEM4	Dyelue		
Item		(n = 12)	(n = 12)	SEIVIT	P value		
pH ²		5.73	5.75	0.007	0.0619		
Cooking loss $(g/100 g)$		39.1	50.7	4.61	0.1058		
Water holding capacity (g/ 100 g)	26.7	28.2	0.88	0.2467		
Shear force (kg/cm ²)		8.0	7.8	0.39	0.7269		
Color coordinates							
L* (whiteness)		37.18	36.51	0.585	0.4293		
a* (redness)		3.41	2.11	0.502	0.0757		
b* (yellowness)		18.19	1.71	0.400	0.2185		
¹ Diets were: the control diet (CON) or 12.5% SM (SM12.5) of dietary dry matter (DM). ² pH measured after thawing. ³ SEM = Standard error of the mean							

DISCUSSION

Sesame meal and formulated diets chemical composition values, in this current trial, were slightly within the range values of what previously reported by other researchers (Awawdeh et al., 2019; Obeidat et al., 2022). The greater diets' EE and ME content reported in this study might referred to the process of SM preparation and kinds of seeds which harvested from various plants (Elleuch et al., 2007). Feed cost, on the other hand, was reduced by 10 % compared to CON when SM was included. This result is comparable to which reported by Obeidat et al. (2022); authors noticed that the inclusion of sesame meal to the lambs' formulated diet decreased feed cost per ton (US\$) by 10%.

Omer et al. (2019) noticed a reduction of feed cost ranged from 27 to 38% by increasing the quantity of SM added to the formulated diets compared to the CON. Our findings may contribute to the enhancement of economic effectiveness attained via utilizing alternative feeds by Awassi lambs.

An improvement in fasting live weight and carcass weights with lambs consumed SM included diet was noticed in this current study. In agreement with our results, other researchers reported increasing final weight and carcass weight with lambs fed sesame by-products containing diets (Fitwi and Tadesse, 2013; Bonos et al., 2017). Those previous studies attributed the improvement of final and carcass weight to the increase of feed intake and utilization of diets containing sesame by-products. In this current study, greater fasting and carcass weight revealed from improved SM diet intake and digestibility which was reported in previous study (Obeidat et al., 2022).

Other carcass characteristics (dressing percentage, cuts weight, non-carcass components, dissected leg, fat tail and leaner dimensions) where not differ with the group consumed SM diet from the CON group which reflects that the addition of SM had no adverse effect on Awassi lambs' carcass traits. Fitwi and Tadesse (2013) reported opposite results to our study; they noticed an increase in dressing percentage, rib-eye area, and non-carcass component weight with the addition of sesame seed cake to sheep diet.

Meat quality was not affected with sesame by-products addition to diets as reported previously (Bölükbaş and Kaya, 2022; Kaya et al., 2022). Minor changes on meat quality were noticed by the addition of SM, in this current study, through a slightly increase in meat pH with the SM group. It was expected that including sesame by-products in sheep diets could alter rumen function as changing the pH and subsequently the composition of the meat, despite the fact that considerable amounts of sesame oil are rich in PUFA and may improve rumen microbial population (Bauman and Griinari et al., 2003; Aldai et al., 2012).

Regarding meat color, redness slightly increased with SM group. Awawdeh et al. (2019) noticed changing in meat color with addition of alternative feed to lambs' diets. The researchers referred variations in redness as being caused by altered mechanisms for pigment synthesis rather than a direct result of hemoglobin or myoglobin concentration in meat (Priolo et al., 1998).

The goal of incorporating alternative feeds into livestock diets is to lower costs without affecting the quality of the meat or the carcass traits. According to the current findings, adding SM up to 12.5% of diets had no effect on the carcass and meat characteristics. Both preceding and this current study have demonstrated that SM might be included in animal diets without negatively affecting the meat quality and carcass traits.

CONCLUSION

The current results verified that adding sesame meal to lambs' fattening rations had no negative impact on the meat's quality or the carcass's features. Additionally, the cost of feed was decreased by 10% for the SM-based diet during the time and settings of this study which led to increase the economic efficiency.

DECLARATIONS

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Authors' contributions

Mysaa Ata: Writing, reviewing, and editing; Belal Obeidat: Writing & editing, data curation; formal analysis, methodology, supervision.

Data availability

The datasets used and/or analysed during the current study available from the corresponding author on reasonable request.

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Consent to publish

All authors agree to the publication of this manuscript.

Competing interests

The authors have not declared any conflict of interest.

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PREVALENCE AND ECONOMIC CONSEQUENCES OF UMBILICAL LESIONS IN LIVESTOCK FARMING

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Supporting Information

ABSTRACT: The aim of this study is to determine the incidence of umbilical lesions in calves within livestock enterprises and to assess the economic losses they incur. The analysis is based on clinical data from 815 umbilical lesion cases recorded over a 17-year period, supplemented by findings from a Delphi survey conducted with expert veterinarians. The results indicated that umbilical lesions predominantly occur in March and April, and 53% of affected calves are younger than one month old. It was observed that 72.8% of umbilical lesions occurred in Simmental calves. Additionally, umbilical urachus fistula was more commonly diagnosed in females, while omphalitis was more prevalent in males. The incidence rate of umbilical lesions was calculated at 38.6%, with a surgical success rate of 73.9%. Postoperative complications were observed in 28.9% of cases, and 53.8% of the calves were culled due to growth retardation or further complications. The estimated cost of surgical intervention and postoperative treatment for umbilical lesions was determined to be \$101.3 per calf. However, this cost increased to \$245.6 in cases complicated by postoperative complications and developmental delays. Despite the fact that umbilical lesions can be largely prevented through simple postnatal hygiene measures, their incidence remains high, leading to substantial economic losses in Türkiye.



Keywords: Calves, Delphi survey, Economic losses, Hygiene measures, Postnatal infections, Surgical intervention.

INTRODUCTION

Ensuring the healthy birth and postnatal development of calves is a critical factor for the sustainability and profitability of livestock enterprises. In particular, adequate colostrum intake and the implementation of early hygiene protocols significantly reduce mortality and morbidity rates while also enhancing growth performance, milk yield, and reproductive efficiency, thereby contributing to long-term economic gains in both the dairy and beef cattle sectors (Godden et al., 2019; Avcioğlu et al., 2024; Keller et al., 2024).

In recent years, the expansion of cattle farming in Türkiye has led to a notable rise in the number of calves admitted to veterinary clinics (Demir and Gültekin, 2024). Among the most common health issues in newborn calves are umbilical lesions, including omphalitis, umbilical abscess, urachal fistula, omphalophlebitis, umbilical hernia, omphaloarteritis, and umbilical eventration (Guerri et al., 2020). These conditions are reported to be highly prevalent among calf surgical diseases (Ganga et al., 2011; Hayat et al., 2019; Yurdakul et al., 2021).

The etiology of umbilical lesions is influenced by both hereditary and environmental factors. Key environmental contributors include improper umbilical cord cutting and sanitation, poor housing conditions, compromised general health, insufficient colostrum intake, and inadequate postnatal care and feeding (Sağlıyan et al., 2016; Ayvazoğlu et al., 2020; Avcioğlu et al., 2024).

Beyond the direct financial burden associated with treatment costs and calf mortality, umbilical lesions can lead to developmental delays and reductions in meat, milk, and reproductive efficiency. These losses not only affect individual farm profitability but also have broader implications for the livestock sector. This study was conducted with the aim of estimating the incidence of umbilical lesions in calves and the economic loss incurred.

MATERIALS AND METHODS

In this study, data from 815 calves of different breeds and sexes, admitted with complaints of umbilical lesions to the Department of Surgery at Kafkas University Veterinary Clinic in Kars during the period 2007–2023, were utilized. The data were categorized based on anamnesis information provided by animal owners.

In the analysis of the obtained data, frequency tables and descriptive statistics were employed. The statistical relationships between umbilical lesion diagnoses and categorical variables were determined using the chi-square test, Mann-Whitney U test, and Kruskal-Wallis test. The Bonferroni test was used as the Post-Hoc test in determining which

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groups the difference was derived from The IBM SPSS Statistics 20 software package was utilized for all statistical analyses. Furthermore, expert opinions regarding umbilical lesions were collected through a face-to-face Delphi survey conducted with 25 expert veterinarians working in the region. The survey questionnaire included items related to technical and economic parameters. Based on the data obtained from the Delphi survey, three distinct scenarios (optimistic, pessimistic, and expected) were established.

The estimated economic loss associated with umbilical lesions in calves was calculated using a combination of technical and economic parameters derived from both field data and expert opinions. The technical and economic data, as well as the formulas used for determining the estimated economic losses, are presented in the Table 1 (1USD\$= 32.4 TL in 17.04.2024 - TCMB).

Table 1 - Key Parameters Used in the Estimation		
Parameters	Value (Min-Max)	References
1. Number of calf in Türkiye	5.950.181	TURKSTAT (2024)
2. Total number of calf in Kars province	164.894	TURKSTAT (2024)
3. Average market price calf (\$)	577.0 (420-775)	TURKSTAT (2024)
4. Morbidity Rate of Newborn Calves (%)	24.7	Ayvazoğlu Demir et al. (2019)
5. Mean incidence rate of umbilical lesions in calves (%)	38.6 (5.0-70.0)	Expert opinion
6. Surgical intervention cost (\$/head)	34.9 (16.4-81.9)	Expert opinion
7. Treatment and medication cost (\$/head)	27.9 (10.9-54.6)	Expert opinion
8. Economic loss due to reformed (culled) animals (\$)	144.25 (105-194)	Expert opinion
9. Postoperative additional care and nutritional expense (\$/day/head)	1.64 (1.02-3.16)	Calculation
10. Duration of postoperative recovery (Days)	23.5 (9-40)	Expert opinion

Estimated Loss=(NUL×PUL×ROp×RRef×CRef)+(NUL×PUL×COp+T)+(NUL×PUL×SOp×TRec×CEC) NUL = Total number of calves diagnosed with umbilical lesions; PUL = Proportion of calves affected; ROp = Probability of surgical intervention; RRef = Proportion of animals culled post-treatment due to complications; CRef = Cost associated with reformed (culled) animals; COp+T = Combined cost of surgical intervention and postoperative medical treatment; SOp = Success rate of the surgical procedure; TRec = Average recovery period following surgical intervention (days); CEC = Additional daily cost of postoperative care and nutrition per calf; Estimated Loss (No Surgery)=(NUL×MRate×CCalf)+(NUL×RRef×(1-MRate)×CCalf/4) NUL = Total number of calves diagnosed with umbilical lesions; MRate = Mortality rate due to untreated umbilical lesions; CCalf = Market value of a calf; RRef=

Proportion of calves culled due to disease-related complications

RESULTS AND DISCUSSION

In neonatal calves, the umbilical cord typically undergoes closure by the fifth day post-birth under normal conditions. provided no complications arise (Hides and Hannah, 2005; Yurdakul et al., 2021). However, in instances where appropriate care and nutritional management are not ensured, umbilical lesions become an unavoidable consequence. Umbilical infections constitute a significant proportion of surgical conditions observed in neonatal calves and represent one of the most frequently encountered health issues in this age group.

The annual distribution of calves presenting with umbilical lesions at Kafkas University Veterinary Faculty Research Hospital is illustrated in Figure 1, which indicates that between 2007 and 2015, the number of calves admitted with umbilical lesions fluctuated between 20 and 30 cases per year. However, a notable increase was observed after 2016. This trend may be linked to the sharp rise in red meat prices in Türkiye since 2016, which has led to increased livestock valuation by producers and heightened attention to animal health (Aksoy et al., 2018).

In this study, the diagnostic distribution of umbilical lesions was as follows: 46.5% of cases were identified as umbilical hernia, 38.5% as omphalitis, 10.9% as umbilical abscess, and 4.1% as urachus fistula. Regarding lesion types, the majority of diagnosed cases consisted of umbilical hernia and omphalitis, findings that are in agreement with those of Yurdakul et al. (2021). However, some studies have suggested a higher prevalence of omphalitis and urachus infections (Moscuzza et al., 2014; Marchionatti et al., 2016). The frequent occurrence of omphalitis is likely due to insufficient postnatal umbilical care, inadequate colostrum intake, and substandard shelter hygiene (Ayvazoğlu Demir et al., 2019; Yurdakul et al., 2021). The association between calf sex and the occurrence of umbilical lesions is summarized in Table 2, which reveals that 31% of the calves admitted to the veterinary hospital were female, while 69% were male. Among these, 72.7% of female calves were diagnosed with urachus fistula, and 71.3% of male calves with omphalitis, indicating a statistically significant association between sex and umbilical lesion type (P<0.01). This association was further supported by the Mann-Whitney U test, which confirmed a significant difference between sex and lesion type (U = 45.419, P = 0.03, P<0.05).

In this study, it was determined that a notable proportion of the affected calves were male (69%), and this difference was statistically significant, aligning with previous studies that also reported a higher frequency of umbilical lesions in male calves (Ayvazoğlu Demir et al., 2019; Sağlıyan et al., 2016). Similarly, Kharb et al. (2021) found that 66.7% of umbilical lesion cases occurred in male calves, while Fazili et al. (2013) reported that umbilical hernias were more common in males, suggesting a potential sex-related predisposition. The higher susceptibility observed in male calves may be attributed to anatomical factors, particularly the close proximity of the umbilical cord to the urethra, which increases the likelihood of urinary contamination and subsequent infection. An analysis of the breed distribution of umbilical lesions revealed that 72.8% of the affected calves were Simmental, 13.1% Montofon, 8.3% Simmental crossbreds, and 5.8% indigenous breeds. The prevalence of umbilical lesions by breed is detailed in Table 3.



Table 2 - Occurrence of umbilical lesions by sex						
Diagnosis	Female	Male	Total			
Omphalitis	90 (28.7%)	224 (71.3%)	314 (38.5%)			
Hernia	112 (29.6%)	267 (70.4%)	379 (46.5%)			
Abscess	27 (30.3%)	62 (69.7%)	89 (10.9%)			
Urachus fistula	24 (72.7%)	9 (27.3%)	33 (4.1%)			
Total	253 (31.0%)	562 (69.0%)	815 (100%)			

Table 3 - Ratio of umbilical lesions by calf breeds						
Diagnosis Breeds	Omphalitis	Hernia	Abscess	Urachus fistula	Total	
Simmental	232 (39.1%)	281(47.4%)	55 (9.3)%	25 (4.2%)	593 (72.8%)	
Simmental crossbred	21 (30.9%)	37 (54.4%)	9 (13.2%)	1 (1.5%)	68 (8.3%)	
Montophone	38 (35.5%)	44 (41.1%)	22 (20.6%)	3 (2.8%)	107 (13.1%)	
Local breed	23 (48.9%)	17 (36.2%)	3 (6.4%)	4 (8.5%)	47 (5.8%)	
Total	314 (38.5%)	379 (46.5%)	89 (10.9%)	33 (4.0%)	815 (100%)	
X ² =21,094; P =0,012; P <0,05						

According to Table 3, Simmental and Simmental crossbred calves exhibited a higher incidence of umbilical hernia, while omphalitis was more frequently diagnosed in indigenous breeds, and umbilical abscess was more common in Montofon calves. Chi-square analysis revealed a statistically significant difference in lesion distribution across breeds (P<0.05). Breed-based analysis revealed that 72.8% of the calves presenting with umbilical lesions were of the Simmental breed. This result is consistent with previous studies by Ayvazoğlu Demir et al. (2019) and Yurdakul et al. (2021), which reported umbilical lesion rates of 50–55% in Simmental calves. The high prevalence of umbilical lesions in Simmentals may be attributed to their increasing population in Türkiye as well as their greater susceptibility to disease compared to indigenous cattle breeds.

The monthly distribution of umbilical lesion cases recorded between 2007 and 2023 is illustrated in Figure 2, which shows that the incidence of umbilical lesions peaked in March and April, coinciding with the calving season. Seasonal analysis revealed that 40.5% of cases were reported in spring, 23.5% in winter, and 20.7% in summer. These findings

align with expert opinions collected through the Delphi Survey, where veterinarians consistently reported an increase in umbilical lesion cases during March and April. This observation is consistent with other studies reporting the highest incidence of umbilical lesions between February and April (Aydemir et al., 2022; Avci et al., 2024). Similarly, another study observed that calves born in unhygienic environments during the calving season were more susceptible to umbilical infections, highlighting the importance of proper management during this critical time (Kharb et al., 2021). The seasonal distribution of umbilical lesions is presented in Table 4, which shows that omphalitis, hernia, and abscess cases were more frequently diagnosed in spring, whereas hernia and abscess were prevalent in autumn and summer. Urachus fistula was most commonly observed in winter. The statistical analysis confirmed a significant relationship between seasonality and lesion diagnosis (P<0.01). Similarly, the Kruskal-Wallis test yielded a statistically significant association between season and lesion type (K=9.794, P=0.02, P<0.05).

Age-based analysis revealed that 53.0% of the affected calves were younger than 30 days, 20.5% were between 31-60 days old, and 26.5% were older than 60 days. The correlation between age and umbilical lesion prevalence is presented in Table 5, which indicates that omphalitis was predominantly observed in calves younger than 30 days (47.7%), while umbilical hernia was more frequently diagnosed in those older than 90 days (72.3%). Abscesses were more common in calves aged between 30-60 days and those older than 90 days. The statistical analysis revealed a significant difference between age groups and lesion type (P<0.05). Additionally, the Kruskal-Wallis test confirmed a statistically significant association between age and umbilical lesions (K=27.630, P=0.00, P<0.01).

Regarding treatment outcomes, 89.6% of the calves admitted with umbilical lesions recovered successfully, while 10.4% experienced postoperative complications. The relationship between disease diagnosis and surgical success is illustrated in Table 6, which demonstrates that the complication rates for omphalitis, hernia, abscess, and urachus fistula were approximately 10%. However, chi-square analysis did not indicate a statistically significant relationship between lesion type and prognosis (P>0.05).

Further evaluation of clinical severity revealed that 7.6% of affected calves exhibited mild symptoms, 84.5% had moderate symptoms, and 7.9% presented with severe symptoms. The association between disease prognosis and surgical success is summarized in Table 7, which indicates that 89.6% of calves underwent successful surgical intervention. Statistical analysis revealed no significant correlation between disease severity and surgical success (P>0.05). To estimate the economic losses associated with umbilical lesions, the results of the Delphi Survey conducted with expert veterinarians are provided in Table 8.





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	Diagnosis	Hamla	Abaaaa	Line along flatula	Tatal	
Season	Umphalitis	Hernia	ADSCESS	Urachus fistula	Iotai	
Winter	75 (39.3%)	79 (41.4%)	22 (11.5%)	15 (7.9%)	191 (23.5%)	
Spring	143 (43.3%)	155 (47.0%)	27 (8.2%)	5 (1.5%)	330 (40.5%)	
Summer	56 (33.1%)	83 (49.1%)	23 (13.6%)	7 (4.1%)	169 (20.7%)	
Autumn	40 (32.0%)	62 (49.6%)	17 (13.6%)	6 (4.8%)	125 (15.3%)	
Total	314 (38.5%)	379 (46.5%)	89 (10.9%)	33 (4.0%)	815(100%)	
X ² =22.777; P=0.007; P<0.01						

Table 4 - The rates of umbilical lesions by seasons								
Day	Diagnosis	Omphalitis	Hernia	Abscess	Urachus fistula	Total		
<30 day		206 (47.7%)	169 (39.1%)	34 (7.9%)	23 (5.3%)	432 (53.0%)		
31-60 day		66 (39.5%)	68 (40.7%)	28 (16.8%)	5 (3.0%)	167 (20.5%)		
61-90 day		30 (28.8%)	61 (58.7%)	11 (10.6%)	2 (1.9%)	104 (12.8%)		
>90 day		12 (10.7%)	81 (72.3%)	16 (14.3%)	3 (2.7%)	112 (13.7%)		
Total		314 (38.5%)	379 (46.5%)	89 (10.9%)	33 (4.0%)	815 (100%)		
X ² =74.039 P =0,00 P <0.01								

Table 6 - Success of the operation with the diagnosis of the disease of the calves

Diagnosis	Success of the operation	Success	Complication	Total	
Omphalitis		282 (89.8%)	32 (10.2%)	314	
Hernia		340 (89.7%)	39 (10.3%)	379	
Abscess		79 (88.8%)	10 (11.2%)	89	
Urachus fistula		29 (87.9%)	4 (12.1%)	33	
Total		730 (89.6%)	85 (10.4%)	815	
X2=0,190 P=0.979 P>0.05					

Table 7 - The success of the operation with the prognosis of the disease of the calves

	Success of the operation	Fuener	Complication	Total
Prognosis		Juccess	complication	Total
Slight		57 (91.9%)	5 (8.1%)	62 (7.6%)
Middle		613 (89.0%)	76 (11.0%)	560 (84.5%)
Severe		60 (93.8%)	4 (6.2%)	64 (7.9%)
Total		730 (89.6%)	85 (10.4%)	815 (100%)
Y2 =1 834 P=0 400 P >0 05				

Table 8 - Technical parameters of the economic losses due to umbilical lesions in Türkiye

Variable	Mean (Min-Max)
Mean umblical lesions rate in calf (%)	38.6 (10-60)
Live weight loss in calves due to umbilical lesions (%)	16.6 (10-50)
Rate of growth retardation in animals due to umbilical lesions (%)	46.6 (10-90)
Mortality rate of calves in case of untreated disease (%)	67.8 (30-90)
Recovery rate of calves in case of untreated disease (%)	12.5 (5-50)
Probability of operation for calves with umbilical lesions (%)	92.6 (80-98)
Operation success rate in calves (%)	73.6 (50-98)
Postoperative recovery time (Day)	23.5 (9-40)
Probability of return of the calf after the operation (%)	71.6 (50-95)
Recovery time of the calf after the operation (Month)	1.5 (1-2)
Probability of any postoperative complication (%)	28.9 (5-70)
Recurrence rate of the disease within 1 year after treatment (%)	10.7 (0-20)
Rate of reformed calves after navel operation (%)	53.8 (10-70)

Variable	Loss per calf (\$/calf)	Estimated average loss	(%)	Optimistic scenario	Pessimistic scenario
Reformed animal loss	144.3 (105-195)	1.129.791	45.98	161.772	1.716.769
Operation cost	34.9(16.39-81.97)	508.072	20.68	252.519	1.030.942
Post-operative medication cost	27.9 (10.93-54.65)	406.167	16.53	168.397	687.336
Extra postoperative care cost	38.5 (9.18-86.4)	412.943	16.81	138.606	543.329
Total	-	2.456.973	100.00	721.295	3.978.375
* The umbilical lesion rate was calculated as 3	38.6%.				

Table 9 - Estimated cost of operations performed due to umbilical lesions in Kars Province (\$)*

According to the survey findings, the incidence of umbilical lesions average prevalence of 38.6% (10%- 60%). Consistent with these findings, prior studies have reported prevalence rates ranging between 1.3% and 66.15% (Pamuk et al., 2009; Hayat et al., 2019; Avci et al., 2024). Notably, comparative analyses have demonstrated a higher prevalence of umbilical infections in Türkiye relative to European countries (Svensson et al., 2003; Wieland et al., 2016; Yanmaz et al., 2017). Expert veterinarians reported that 95.6% of the calves diagnosed with umbilical lesions underwent surgical intervention. The success rate of these procedures was recorded at 73.9% (ranging from 50% to 98%), with an average recovery period of 23.5 days. Approximately 71.6% of calves recovered fully within 30-60 days.

Although medical treatment options exist for umbilical lesions, their efficacy remains limited. The Delphi survey results indicated that the recovery rate of untreated calves was as low as 12.5%, while the mortality rate reached 67.8%. Given these findings, surgical intervention remains the most definitive treatment approach, particularly for umbilical hernias (Yanmaz et al., 2017; Yurdakul et al., 2021).

The study further revealed that 53.8% of affected animals were culled due to complications. In the absence of treatment, umbilical lesions, including omphalitis, omphalophlebitis, omphaloarteritis, urachus fistula, umbilical abscess, and umbilical hernia, may lead to severe systemic complications through hematogenous dissemination, potentially affecting vital organs such as the lungs, kidneys, joints, and other tissues (Marchionatti et al., 2016; Yurdakul et al., 2021). Notably, umbilical infections can lead to pyemia, resulting in conditions such as arthritis and septicemia, thereby increasing treatment costs and contributing to economic losses due to mortality in non-responsive cases.

Economic analyses based on survey data estimated the average cost of surgical intervention for umbilical lesions in Kars at \$101.3 per calf. However, in cases complicated by postoperative complications and developmental delays, this cost may rise to \$245.6 per animal. According to birth statistics, an estimated 164.894 calves were born in Kars in 2023, of which approximately 15.721 were affected by umbilical lesions. Consequently, the annual economic loss attributable to the treatment of umbilical lesions in Kars was calculated at approximately \$2.46 million, with variations ranging from \$721.295 in the most optimistic scenario to \$3.98 million in the pessimistic case (Table 9).

Furthermore, in the absence of treatment, economic losses were projected to increase dramatically to \$6.88 million, factoring in a 67.8% mortality rate and an estimated 32.2% reduction in the value of affected breeding stock. When the same methodology was applied at the national level, it was estimated that 567.302 of the 5.950.181 calves born in Türkiye in 2023 may have developed umbilical lesions. Accordingly, the total economic burden associated with surgical intervention was estimated at \$88.66 million. In scenarios without intervention, this figure could rise substantially to \$248.28 million, due to increased mortality and productivity losses.

Taken together, these findings emphasize that umbilical lesions in neonatal calves constitute a significant source of economic loss, both in terms of direct treatment expenditures and indirect losses associated with impaired growth and early culling. Notably, a review of the literature revealed a lack of previous studies that quantitatively assessed the economic burden of umbilical lesions, further underlining the relevance and contribution of the current study.

CONCLUSION

The findings of this study demonstrate that umbilical lesions in calves impose a significant burden on the livestock sector, affecting both animal health and economic sustainability. While surgical intervention helps mitigate financial losses to some extent, umbilical lesions continue to generate substantial costs due to treatment expenses, postoperative care, complications, transport costs, labor inefficiencies, and production losses. However, these lesions can be largely prevented through simple and cost-effective management strategies. Raising awareness among livestock owners regarding preventive measures, ensuring that affected animals receive timely treatment at specialized veterinary centers, and refraining from using calves with congenital umbilical hernia for breeding purposes can substantially reduce the economic losses associated with umbilical lesions in the Turkish livestock industry. Implementing these strategies can enhance both animal welfare and the overall profitability of livestock enterprises.

DECLARATIONS

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Data availability

The datasets used and/or analysed during the current study available from the corresponding author on reasonable request.

Authors' contribution

P. Demir Ayvazoğlu, and E. Aydın: designed the research, supervision, writing, and editing. U. Aydın and U. Yıldız: investigation, collecting the data. Ö. Aksoy: writing, and review.

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RESPONSE OF HEAT STRESSED BROILERS TO AMELIORATING EFFECTS OF SYNTHETIC VITAMIN C AND Citrus sinensis EXTRACT

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Supporting Information

ABSTRACT: Heat stress is a critical environmental factor in the poultry industry, negatively impacting performance and causing economic losses. High temperatures reduce productivity and increase mortality. This study used 120 Abor Acre broilers (28 days old, 1.050 ± 0.4 kg) fed standard commercial feed. At day 35, 30 birds were randomly assigned to 4 treatments: T₁ (water as control), T₂ (30 mg/L synthetic vitamin C based on manufacturers' dosage), T₃ (30 ml/L orange extract), and T₄ (50 ml/L orange extract), each in three replicates. Birds were exposed to artificial heat stress (36 \pm 2°C) for 15 days using an automated heater, with unrestricted access to feed and treatment solutions. Physiological data were recorded periodically, and blood samples were collected on days 0, 5, 10 and 15 for serum biochemical analysis. Data were analyzed using the SAS procedure for repeated and non-repeated measures. Results showed significantly higher final body weight (FBW) and average body weight gain (ABWG) in T4 compared to other groups (P < 0.05). Average daily feed intake (ADFI) was highest in T₄ chickens, and feed conversion ratio (FCR) was lowest, indicating superior feed efficiency. Rectal temperature, body temperature, pulse rate, and respiratory rate were significantly highest in T₁ group (P < 0.05). Serum biochemical markers (AST, ALT, glucose, and creatinine) were highest in T_1 , whereas T_4 recorded the lowest values. T_1 broilers had the highest external and internal thermal gradients, while T₄ had the lowest. Glycogen reserves were highest in T₄ group. The study concludes that sweet orange extract, especially at a 50% concentration, is a viable organic alternative to synthetic vitamin C (30 mg/L) for mitigating heat stress in broilers. It enhances growth performance, feed efficiency, and physiological stability under heat stress, making it a sustainable solution for poultry production in tropical climates.

Keywords: Broiler, Heat stress, Serum, Sweet orange extract, Vitamin C.

INTRODUCTION

Heat stress is one of the most significant stressors impacting poultry production having significant effects on the health and productivity of poultry. Excessive heat is a major threat in both hot climates and temperate regions (Hu et al., 2022; Sun et al., 2023). The broiler industry faces the challenge of heat stress which adversely impacts the growth rate, production performance, immune response, gut function and gut microbiota, redox balance, energy bioavailability in cells, and attainment of the body's homeostasis, culminating in huge economic losses to the poultry industry (Saeed et al., 2019; Uyanga et al., 2021).

A major decrease in feed efficiency, feed intake and body weight gain due to heat stress has been reported in many studies conducted on birds and other animals (Vandana et al., 2021; Ayoola et al., 2023; Chen et al., 2024). During stress conditions, the most important thing any living being must do is to survive rather than growth (Hawkins and Storey 2020; Akinmoladun et al., 2023). A study on broilers revealed that both recurring and continuous environmental stress (heat stress) significantly compromises growth performance by reducing protein digestibility up to 9.7%. Broilers under heat stress have shown increased metabolizable energy intake (20.3%) and heat production (35.5%) and decreased energy retention (20.9%) and lastly energy efficiency (32.4%) (De Souza et al., 2016).

Environment temperature is one of the factors that affect gut health including gut microbial ecosystem, gut health relating to nutrient absorption affect the numerous nutrient that is used for growth (Patra and Kar, 2021). Heat stress affects the physiological parameters in animals and the responses vary depending on the time and degree of thermal challenge (Ayoola et al., 2020).

Vitamins are essential nutrients that are required for various biochemical and physiological processes in the body (Johnson and Bales, 2022). Sweet orange (*Citrus sinensis*) is a main source of vitamin C but also contain ample amount of calcium, folic acid, potassium, thiamine, niacin and magnesium (Gupta et al., 2020). Dietary supplementation of vitamin C (ascorbic acid) aids in limiting stress metabolic signs, improved growth performance, enhanced immunological status and reduced mortality (El-Senousey et al., 2018, NCBI, 2021). Ascorbic acid serves as an antioxidant and has anti-

inflammatory effects which is effective for poultry in cases of inflammation, oxidative stress, and infection (El-Senousey et al., 2018).

The current research trend in livestock production focuses on sustainable agriculture and application of organic products which involves using natural materials, drugs, or food supplements rather than synthetic products. In a heat stress environment, vitamin C which is an active ingredient in OE contributes to the energy supply of poultry birds by corticosterone biosynthesis (Oguntunji et al., 2019, Akinmoladun et al., 2023). The present study aims to evaluate the ameliorating effect of natural source of ascorbic acid (*Citrus sinensis*) extract on heat stress in broiler chicken as compared to the use of synthetic vitamin C based on manufacturers' dosage. The outcome of this study is expected to contribute to sustainable broiler production in the tropics.

MATERIALS AND METHODS

Study area

The study was carried out at the Poultry Unit, Teaching and Research farm, Bowen University, Iwo, Osun State. The laboratory work was conducted at the Animal Science laboratory, Bowen University, Iwo. The use and handling of animals were in accordance with the approval granted by the Bowen University ethics committee (BUEC). The study area is located in Derived Savanna Agro-Ecological Zone characterized with environmental temperature range of 15 to 28°C and mean annual rainfall of 1400 mm. The coordinate of the study area is Latitude 7°37'30" N and Longitude 4°11'40" E.

Birds and management

One hundred and twenty healthy abor acre broilers aged 28 days old, weighing 1.050±0.4 kg served as subjects of the study. The birds were raised in a standard poultry house and fed standard commercial feed from day-old. All necessary medications were administered to the birds. Vaccinations, medications, and other related management practices were done as recommended by the National Veterinary Research Institute (NVRI), Jos, Nigeria.

Ethical considerations

The methods/procedures used in this study were in concomitant with those outlined in the Animals ARRIVE guidelines and were carried out by the U.K. Animals (Scientific Procedures) Act, 1986 and associated guidelines; EU Directive 2010/63/EU for animal experiments; or the National Institutes of Health guide for the care and use of laboratory animals (NIH Publications No. 8023, revised 1978). The study was conducted with the approval of the Bowen University Ethics Committee.

Experimental unit

Sweet oranges were sourced from a local market and the juice was extracted. Fresh orange extract (OE) free of seeds and other foreign particles was stored in an air-tight jar and refrigerated for subsequent use. At age 35 days, thirty birds were randomly assigned to each treatment of oral supplementation as T_1 (control-water), T_2 (30 mg/L synthetic vitamin C based on manufacturers dosage), T_3 (30ml/L of OE), and T_4 (50ml/L of OE) in three replicates and birds were subjected to regulated artificial induced heat stress simulation pen maintained at 36 ± 2°C for 24 hours, 15 days using an electric automated heater. Throughout the trial, birds had unrestricted access to feed and treatment solution.

Data collection

Meteorological data

Meteorological data within the pen were monitored and recorded. Temperature within the pen was maintained at 38°C, while average relative humidity was 72.5% during the experiment. Feed intake, body weight, and feed conversion ratio were monitored weekly.

Physiological parameters

Data on physiological parameters were taken as described by Ayoola et al (2023) on days 0, 5, 10 and 15. Each parameter were measured and described as:

The respiratory rate (RR): Determined by counting the number of flank movements in one complete breathing in a minute or vent per minute using a stopwatch".

The skin temperature (ST°C): An infrared thermometer was placed at the shaved area under the wings. Nubee NUB8380, California, USA model was positioned at a 15 cm distance from the animal to and measurement was taken after the sound of alarm from the thermometer.

Rectal temperature (RT°C): A digital clinical thermometer was used in taking the measurement. The sensory tip of the thermometer was disinfected, and inserted via the vent of birds (depth of 1 cm). Reading on thermometer was taken after the indicator sound.

Panting rate (PR °C): This is a reading of times a bird panted within a minute using a stopwatch.

From the measured data: The body thermal gradients was calculated as:

Internal thermal gradient = $(RT^{\circ}C - ST^{\circ}C)$; External thermal gradient = $(RT^{\circ}C - ambient temperature)$; Total thermal gradient = $(ST^{\circ}C - ambient temperature)$ (Yahav and Hurwitz, 1996)

Serum biochemical data

On days 0, 5, 10 and 15 of experiment, five birds were randomly selected from each replicate carefully to avoid stress. Blood samples were collected via wing veins from the selected birds. Blood samples for serum analyses were collected into plain bottles, centrifuged and serum was stored at -20°c until further analysis.

Statistical analysis

Data obtained for vitamin supplement on growth performance and muscle glycogen were analyzed with completely randomized design (CRD). The equation was of the form: Yij = μ + Ti + ε ij

Where: Yij = individual observation; μ = General mean; Ti = Treatment effect (i= 1.....4); ε_{ij} = experimental error.

The interactive effects of vitamin supplements and periods on physiological and serum biochemical parameters were analyzed with a two factorial, 4 x 4 factorial experiment in a CRD.

The statistical model used was: Yijk = μ + Ai + Bj + (AB)ij + ε ijk;

Where: Yij = individual observation; μ = population mean; Ai = Effect of factor A (i= 1.....4); Bj = Effect of factor B (i= 1.....4); (AB)ij = Interactive effect of factors A and B; Eijk = experimental error

The mean differences were determined using Duncan multiple range test (DMRT) at 5% probability All statistical analyses were performed with the SAS-2002, version 16.

RESULTS

The feed composition fed to the experimental birds is as presented in table 1. As reported in table 2, the growth performance results showed no significant differences (P > 0.05) in initial body weight (IBW) among treatments. However, final body weight (FBW) and average body weight gain (ABWG) were significantly higher (P < 0.05) in the T₄ group (50% citrus-sweet orange) compared to T₁ (ordinary water). T₂ (synthetic vitamin) and T₃ (30% citrus-sweet orange) also outperformed T₁ but were not different from each other. Average daily feed intake (ADFI) was highest in T₄ and significantly higher (P < 0.05) than all other treatments, while the feed conversion ratio (FCR) was lowest in T₄, indicating the best feed efficiency. T₁ had the highest FCR, showing the poorest feed efficiency. No mortalities were recorded across treatments.

Table 1 - Gross composition of basal diet	
Feed ingredients	Quantity (kg)
Maize	45
Palm kernel cake	8
Wheat bran	12
Groundnut cake	11
Soyabean meal	14
Fish meal	2
Oyster shell	1
Bone meal	3
Vitamin premix*	2.5
Salt	1.2
Methionine	0.15
Lysine	0.15
Total	100
Calculated values	
Metabolizable Energy (ME)	2800.5 kcal*kg ⁻¹
Crude protein (CP)	20.1%
Crude fibre (CF)	4.98% n
Ether Extract (EE)	5.5%
*: Premix to provide the followings per kg of feed: Vitamin A: 500 iu, Vit.D3: 1200 mg, Vit.E: 11 mg, Vit.K	3: 2 mg, Riboflavin: 20 mg, Nicotinic

*: Premix to provide the followings per kg of feed: Vitamin A: 500 iu, Vit.D3: 1200 mg, Vit.E: 11 mg, Vit.K3: 2 mg, Riboflavin: 20 mg, Nicotinic acid: 10 mg, Panthothenic acid: 7 mg, Cobalamin: 0.08 mg, Choline chloride: 900 mg, Folic acid: 1.5 mg, Biotin: 1.5 mg, Iron: 25 mg, Manganese: 80 mg, Copper: 2 mg, Zinc: 50 mg, Cobalt: 1.2 mg and Selenium: 0.1 mg.

Table 2 - Performance c	haracteristics of b	roilers fed control a	ind experimental	diets for 15 days
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Parameter	T1	T ₂	Тз	T4	SEM	
IBW (kg)	1.52	1.55	1.53	1.54	1.16	
FBW (kg)	1.71 °	1.80 ^b	1 .78 [♭]	1.83 ª	0.58	
ABWG (kg)	0.19°	0.25 ^b	0.25 ^b	0.29ª	0.21	
ADFI (g)	256.55°	303.13 ^b	308.27 [♭]	331.25 ª	2.12	
FCR	1.88 ^a	1.69 ^b	1.72 ^b	1.59 °	-	
Mortality (%)	0	0	0	0	-	

^{abc} means with the same superscripts along a row are not significantly (P>0.05) different from each other. * IBW: initial body weight, FBW: final body weight, ABWG: average body weight gain, ADFI: average daily feed intake, FCR: feed conversion ratio. T_1 = control-water), T_2 = 30 mg/L Synthetic Vitamin C (based on manufacturers dosage), T_3 = 30ml/L of orange extract, and T_4 = 50ml/L of orange extract.

In table 3, rectal temperature (RTC) of birds in T_1 had the highest RTC by day 15 (43.76 ± 4.43 °C), which was significantly higher (P < 0.05) than in T_2 , T_3 , and T_4 . By day 5 onward, RTC in T_2 , T_3 , and T_4 decreased, with T_4 showing the lowest RTC at day 15 (39.06 ± 4.71 °C). Body temperature (BTC) followed a similar trend, with T_1 showing significantly higher (P < 0.05) BTC by day 15 (41.65 ± 4.28 °C), compared to T_2 , T_3 , and T_4 . Birds on T_1 exhibited a significantly higher panting rate by day 15 (56.63 ± 14.16 breaths/min), compared to T_2 , T_3 , and T_4 (P < 0.05). T_2 , T_3 , and T_4 had consistently lower PR values, with T_4 showing the lowest panting rate (48.15 ± 10.51 breaths/min by day 15). Consequently, birds on T_1 had significantly higher RR from day 5 onward (25.81 ± 11.01 breaths/min at day 15) compared to the other treatments (P < 0.05). T_2 , T_3 , and T_4 had lower RR values, with no significant differences among them.

Table 4 reported the biochemical parameters measured in the study across treatments. AST levels, T_1 consistently exhibited the highest values, particularly at day 15 (177.5 ± 4.12 U/L), indicating greater liver stress. In contrast, treatments T_2 , T_3 , and T_4 resulted in significantly lower AST levels, with T_4 showing the most improvement (151.67 ± 3.77 U/L at day 15). ALT levels followed a similar trend. T_1 had significantly higher ALT levels by day 15 (10.65 ± 0.16 U/L), reflecting greater liver strain. The ALT levels in T_2 , T_3 , and T_4 were significantly lower, with T_4 and T_2 showing the best results. Glucose levels were also highest in T_1 (195.1 ± 32.0 mg/dL at day 15), showing a higher stress response in terms of energy metabolism. T_2 , T_3 , and T_4 significantly lowered glucose levels, with T_4 showing the greatest reduction (162.1 ± 32.0 mg/dL), indicating better glucose management and potentially reduced stress. Creatinine levels, indicative of kidney function, were significantly higher in T_1 (3.03 ± 1.06 mg/dL at day 7), suggesting kidney stress. T_2 , T_3 , and T_4 had significantly lower creatinine levels, with T_4 showing the lowest levels by day 15 (0.960 ± 1.10 mg/dL).

As reported in Figure 1, the ETG results show that T_1 experienced the highest heat stress, peaking at day 5 and gradually declining. T_2 and T_3 also peaked at day 5 but at lower levels than T_1 . T_4 consistently exhibited the lowest ETG, with a smaller peak and faster reduction, demonstrating superior heat stress management. The ITG results showed in Figure 2, reported that T_1 had the highest initial ITG which gradually decreased over time, indicating poor internal heat regulation. T_2 exhibited a steady decline, with a sharp drop after day 10. T_3 showed a delayed reduction in ITG, peaking around day 10 before declining. T_4 consistently maintained the lowest and most stable ITG levels throughout the study, demonstrating superior internal heat management. The TTG results in Figure 3 show that T_1 had the highest peak around day 5, indicating the greatest heat stress, with a gradual decrease over time. T_2 and T_3 had similar trends, peaking slightly lower than T_1 and steadily declining, reflecting moderate stress reduction. T_4 displayed the lowest TTG, with a sharp decline after day 5, reaching near-zero by day 15, indicating the best heat stress management.

The glycogen levels in the treatments demonstrate significant differences (P < 0.05) in glycogen storage among the groups. For glycogen 1, T_1 , T_2 and T_3 have comparable glycogen levels, ranging from 0.750 to 0.850, all marked with the same significance level. In contrast, T_4 significantly increased (P < 0.05) glycogen storage to 1.45, indicating superior glycogen accumulation.

Similarly, glycogen 2 results show T₁, T₂, and T₃ with similar glycogen levels ranging from 0.825 to 0.950. T₄ again demonstrated a significant increase in glycogen storage at 1.34 These findings suggest that higher supplementation of citrus-sweet orange enhances glycogen reserves in broilers, indicating its potential as an effective nutritional strategy for improving energy storage and overall performance in poultry production under heat stress conditions.

Treatment	_	_	_	_	_
Parameters	Day	T1	T2	Тз	T4
	0	41.09 ± 3.16	40.79 ± 3.23	40.10 ± 4.72	41.50 ± 3.54
Rectal temperature (RTC °C)	5	43.76 ± 4.43 ^a	42.10 ± 3.42 ^b	42.50 ± 4.18 ^b	42.41 ± 4.15 ^b
Rectai temperature (RTC °C)	10	42.70 ± 5.13 ^a	40.76 ± 3.33 ^b	40.76 ± 3.63 ^b	40.01 ± 4.23 ^b
	15	42.55 ± 4.17 ª	40.26 ± 4.61 ^b	40.22 ± 4.53 ^b	39.06 ± 4.71 ^b
Body temperature (BTC ^o C)	0	38.78±3.16	38.67 ± 4.23	38.53 ± 3.64	38.53 ± 4.20
	5	41.65 ± 4.28 ^a	40.03 ± 3.31 ^b	40.67 ± 3.11 ^b	40.45 ± 3.22 ^b
	10	40.67 ± 3.66 ^a	38.33 ± 4.33 ^b	38.51 ± 4.81 ^b	38.13 ± 3.42 ^b
	15	40.00 ± 3.52 ^a	38.11 ± 5.71 ^b	38.67 ± 4.91 ^b	38.14 ± 3.72^{b}
	0	32.00 ± 1.00	33.21 ± 0.00	32.30 ± 0.00	32.00 ± 0.00
Panting rate	5	56.63 ± 14.16 ^a	52.10 ± 11.26 ^b	52.63 ± 12.66 ^b	52.10 ± 12.81 ^b
(PR breath/min)	10	54.63 ± 12.11 ^a	51.21 ± 10.46 ^b	51.41 ± 11.32 ^b	51.23 ± 13.26 ^b
	15	52.81 ± 15.06ª	48.14 ± 11.16 ^b	48.13 ± 14.51 ^b	48.15 ± 10.51 ^b
	0	20.63 ± 1.23	20.63 ± 1.16	20.63 ± 1.26	20.63 ± 0.16
Respiratory rate	5	24.63 ± 7.22 ^a	22.93 ± 9.06 ^b	23.01 ± 7.06 ^b	22.20 ± 9.11 ^b
(RR breath/min)	10	25.81 ± 11.01 ^a	21.03 ± 10.01 ^b	21.34 ± 8.16 ^b	20.77 ± 7.56 ^b
	15	25.12 ± 12.36ª	21.23 ± 11.51 ^b	21.41 ± 8.04 ^b	20.64 ± 11.31 ^b
^{abc} means with the same superscri	pts along a r	ow are not significantly (I	P > 0.05) different from e	each other. $T_1 = control$	water), $T_2 = 30 \text{ mg/L}$

Table 3 - Effect of	treatments (vitamin	supplement) x p	periods on physiolo	ogical parameters of	heat-stressed broiler
chickens					

^{abc} means with the same superscripts along a row are not significantly (P > 0.05) different from each other. $T_1 = control-water$), $T_2 = 30$ mg/l Synthetic Vitamin C (based on manufacturers dosage), $T_3 = 30$ ml/L of orange extract, and $T_4 = 50$ ml/L of orange extract.

Table 4 - Effect of treatments (vitamin supplement) x periods on serum blood biochemical	parameters of heat-stressed
broiler chickens	

Treatment	Dev	Ŧ	-	Ŧ	Ŧ
Parameters	Day	11	12	13	14
	0	135.2 ± 2.15	136.2 ± 2.15	134.5±2.15	134.9 ± 2.15
AST (88-208 U/L)	5	177.5 ± 4.12 ª	156.1 ± 4.15 ^b	157.5 ± 5.56 ^b	155.3 ± 4.71 ^{bc}
	10	170.2 ± 3.11 ^a	154.5 ± 4.17 ^b	152.5 ± 5.12 ^b	153.5 ± 3.62 ^b
	15	166.1 ± 4.49 ^a	152.3 ± 5.71 ^b	150.4 ± 5.88 ^b	151.67 ± 3.77 ^b
	0	4.18 ± 0.1	4.67 ± 0.1	4.53 ± 0.1	4.53 ± 0.16
ALT (9-37 U/L)	5	10.65 ± 0.16 ª	6.03 ± 0.16 ^b	7.07 ± 0.16 ^{bc}	5.45 ± 0.16 ^{bc}
	10	8.03 ± 0.26 ^a	5.68 ± 3.12°	6.0 ± 0 ^b	5.02 ± 1.44°
	15	5.13 ± 0.18 ^a	4.18 ± 0.1°	5.07 ± 0.16 ^{ab}	4.85 ± 0.16°
	0	170.8 ± 32	170.5 ± 32	170.9 ± 32	171.2 ± 32.0
Chucana	5	195.1 ± 32 ª	181.2 ± 26.2 ^b	183.1 ± 20.15 ^b	178.1 ± 17.25°
Glucose	10	182.1 ± 32 ª	177.2 ± 23.2 ^b	178.1 ± 18.13 ^b	169.0 ± 18.27°
	15	175.1 ± 32.01ª	162.1 ± 31 ^b	163.1 ± 31 ^b	162.1 ± 32 ^b
	0	0.86 ± 0.12	0.85 ± 0.12	0.82 ± 1.12	0.88 ± 1.12
Overtining	5	3.03 ± 1.06ª	2.63 ± 1.01 ^b	2.61 ± 1.06 ^b	2.60 ± 1.01 ^b
Greatinine	10	1.60 ± 0.05 ^a	1.25 ± 0.05 ^b	1.25 ± 0.1 ^b	1.24 ± 0.10 b
	15	1.020 ± 0.1	0.950 ± 0.12	0.910 ± 1.11	0.960 ± 1.1

^{abc} means with the same superscripts along a row are not significantly (P > 0.05) different from each other; * AST: aspartate aminotransferase, ALT: alanine aminotransferase. T₁ = control-water), T₂ = 30 mg/L Synthetic Vitamin C (based on manufacturers dosage), T₃ = 30 ml/L of orange extract, and T₄ = 50 ml/L of orange extract.







Table 5 - Muscle glycogen results of heat stressed broilers to ameliorating effects of synthetic vitamin C and Citrus sinensis in humid tropics.

	Treatments	т.	T.	T.	т.	
Muscle glycogen parameters		11	12	13	14	
Glycogen 1		0.750 ± 0.2 ª	0.800 ± 0.05 ª	0.850 ± 0.05 ª	1.45 ± 0.050 b	
Glycogen 2		0.825 ± 0.25 ª	0.870 ± 0.025 ª	0.950 ± 0.02 ª	1.34 ± 0.035 b	
^{abc} Means along the same row with similar superscripts are not significantly ($P > 0.05$) different using Duncan's test as post hoc analysis. $T_1 = control-water$), $T_2 = 30 \text{ mg/L}$ Synthetic Vitamin C (based on manufacturers dosage), $T_3 = 30 \text{ml/L}$ of orange extract, and $T_4 = 50 \text{ml/L}$ of orange extract.						

DISCUSSION

The present study investigates the use of organic materials, specifically sweet orange extract (OE), as an alternative to synthetic Vitamin C and its relationship with periods in mitigating heat stress in broiler chickens. The growth performance results showed that broilers on supplemented OE T₄ group, exhibited the best growth performance, indicating a positive influence of OE on feed intake and nutrient utilization. These findings are consistent with previous research that highlights the role of vitamin C in improving feed efficiency and growth performance under heat stress conditions (Sumanu et al., 2024; Mohamed et al., 2024). The improvement in growth performance in the OE-supplemented groups can be attributed to the antioxidant properties of sweet orange. Citrus fruits, including sweet orange, are rich in vitamin C, flavonoids, and carotenoids, which have been shown to reduce oxidative stress by scavenging free radicals (Ayoola et al., 2010). Under heat stress, oxidative stress is a major factor that impairs the physiological functions of poultry, leading to reduced feed intake and poor growth (Sahin et al., 2002). The antioxidant properties of sweet orange likely helped to mitigate this oxidative damage, allowing the birds to maintain better growth and nutrient absorption. Additionally, vitamin C plays a role in corticosterone biosynthesis, a hormone that supports energy metabolism during stress (Oguntunji et al., 2019, Ayoola et al., 2023). By enhancing energy metabolism, OE likely helped broilers maintain higher growth rates despite the heat stress conditions.

Heat stress leads to physiological changes in broilers, including elevated rectal temperature, heart rate, and respiratory rate, all of which indicate the birds' efforts to dissipate excess heat and maintain homeostasis (Yahav, 2009). The present study found that broilers on T_4 had significantly lower rectal temperatures, respiratory rates, and panting rates compared to the T_1 , indicating improved thermal regulation. These results suggest that OE supplementation helped in reducing the heat load on the birds. The lower rectal temperatures in the OE groups may be due to the improved antioxidant defense provided by the bioactive compounds in sweet orange. Previous studies have shown that vitamin C supplementation reduces heat stress-induced hyperthermia in poultry by enhancing the bird's ability to dissipate heat (Akbarian et al., 2016). Additionally, vitamin C has been reported to enhance peripheral blood flow, which aids in heat dissipation by increasing heat loss through the skin (Alabi et al., 2021). Sweet orange extract, rich in vitamin C, likely reduced the metabolic and oxidative stress associated with heat, allowing the birds to maintain more normal respiratory function. The reduction in panting behavior in the OE groups could also be attributed to better hydration and electrolyte

balance, which are often disrupted under heat stress (Sahin et al., 2002). Vitamin C has been shown to reduce respiratory alkalosis, a common consequence of excessive panting in heat-stressed birds, by buffering blood pH (Minka and Ayo 2011, Ayoola et al., 2023).

Biochemical markers of stress, such as aspartate aminotransferase (AST), alanine aminotransferase (ALT), glucose, and creatinine, were also measured in this study to assess the effects of heat stress and the protective role of OE. Elevated levels of AST and ALT are indicative of liver stress and damage, while high creatinine levels suggest renal impairment, both of which are common under heat stress conditions (Sahin et al., 2002). The AST and ALT levels were significantly higher in the control group as compared to OE and synthetic vitamin, suggesting that OE helped to protect the liver from heat-induced damage. The protective effect of OE on liver function may be due to the antioxidant properties of vitamin C, which reduces oxidative stress and prevents liver cell damage (Majekodunmi et al., 2014). Studies have shown that antioxidants like vitamin C enhance the liver's capacity to neutralize free radicals, thereby preventing lipid peroxidation and maintaining normal liver function (Sahin et al., 2002).

Heat stress often leads to hyperglycemia as a result of increased corticosterone levels, which stimulate gluconeogenesis to provide additional energy for coping with stress (Yahav, 2009). The glucose levels in the control group were significantly higher (195.1 mg/dL) than those in the OE-supplemented and synthetic vitamin groups, indicating that the OE helped to regulate glucose metabolism and prevent excessive glucose release. The lower glucose levels in the treated groups suggest that the birds were under less stress, likely due to the antioxidant and anti-inflammatory effects of the supplements (Majekodunmi et al., 2014). Similarly, creatinine levels, which indicate renal function, were highest in the control group but significantly lower in the treatment groups, suggesting that vitamin supplements and OE also helped to protect kidney function under heat stress. Heat stress can lead to dehydration and reduced kidney function, but the presence of bioactive compounds in sweet orange likely improved the birds' hydration status and electrolyte balance, preserving renal function (Goto et al., 2022).

Glycogen storage is crucial for energy supply, especially under stressful conditions where energy demand is heightened (Minka and Ayo 2011, Oguntunji et al., 2019). The study found that the treated groups with vitamin supplement and OE had significantly higher glycogen reserves in both the liver and muscle tissues compared to the control group. This suggests that OE compared well with synthetic vitamin supplement, and helped to enhance energy storage, possibly by improving glucose utilization and reducing the energy cost of coping with heat stress. The antioxidant properties of vitamin C may have also contributed to preserving glycogen stores by reducing the oxidative breakdown of glycogen (Attia et al., 2011). The external thermal gradient (ETG) and internal thermal gradient (ITG) are measures of the bird's ability to regulate body temperature in response to environmental heat. The results showed that the T₁ had the highest ETG and ITG, indicating poor thermal regulation and greater susceptibility to heat stress. In contrast, T₄ group had the lowest ETG and ITG, demonstrating improved thermoregulation. This is consistent with previous research, which suggests that vitamin C enhances heat dissipation by improving blood circulation and reducing oxidative stress (Mujahid et al., 2009). According to Mormede et al. (2017), chickens generally recover from acute handling stress within 24 hours, therefore, the consistent results of measured parameters of experimental birds in this study can be attributed to the induced heat stress.

CONCLUSION

The findings of this study provide strong evidence that sweet orange extract, particularly at a concentration of 50%, is an effective organic alternative to synthetic vitamin C for mitigating heat stress in broilers. The OE-supplemented groups exhibited significantly better growth performance, lower physiological stress indicators, improved biochemical parameters, and greater energy storage compared to the control and synthetic vitamin C groups. The antioxidant, anti-inflammatory, and heat-mitigating properties of sweet orange likely contributed to these improvements. This study supports the use of organic supplements like sweet orange extract in sustainable poultry production, particularly in tropical climates where heat stress is a major challenge.

DECLARATION

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Author's contribution

Ayoola Mathew O. contributed to data collection and article write up, Aderemi Foluke A and Lawal Tunde .E. contributed to writing of manuscript, Alabi Olufemi M and Adeleye Bobola. E contributed to research design and data collection, Oguntunji Abel O. contributed to statistical analysis and result interpretation, Oladejo Opeyemi O contributed to data collection and laboratory analysis.

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Competing interest

The authors declare that there is no competing interest during the data collection or writing up of this article.

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EFFECT OF PROBIOTIC SUPPLEMENT ON GROWING LAMBS OF THE LACON BREED DURING THE SUCKLING PERIOD

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Supporting Information

ABSTRACT: Dairy lambs are susceptible to many pathogens that can affect their subsequent performance. The use of probiotics in the rearing of Lacon lambs during the suckling period has been identified as a means of maintaining intestinal microbial balance. Studies have shown that probiotics can be used as an alternative therapy that prevents the use of antibiotics and, thus, can reduce the emergence and spread of antibioticresistant bacteria as well as antibiotic residues in dairy products, meat and milk. In addition, it increases the growth rate of the animal and its stress resistance under various conditions. In this regard, we conducted a comprehensive assessment of the impact using the therapeutic and probiotic supplement Immunobacterin-D on the growth and development of Lacon dairy lambs and their clinical parameters during the suckling period. It was found that the use of the probiotic supplement Immunobacterin-D during the suckling period of Lacon dairy lamb provided an increase in average daily gains by 16.1%, an increase in the content of total protein by 15.8% and an increase in the level of globulin proteins by 35.4%, which indicates a high resistance of the lambs' organism. Thus, the use of the probiotic Immunobacterin-D in feeding lambs had a positive effect on their growth rate, development, improved their health and adaptive capacity, as they showed greater resistance to elevated ambient temperatures compared to those that did not receive the supplement. The use of the probiotic increases the stress resistance of lambs, which can be used as one of the effective approaches in the breeding system of Lacon sheep.



Keywords: Breeding system, Gastrointestinal microbiota, Intestinal microbial balance, Sheep, Stress resistance.

INTRODUCTION

Probiotics are increasingly being used worldwide to improve animal welfare, health and productivity. The probiotics market has been growing steadily for several years and is expected to reach \$7.3 billion, growing at a CAGR of 8.8% between 2020 and 2030. Europe is one of the most important regions in the probiotics market for animal feed with a market share of 35% (Size, 2019).

The mechanism of action of probiotics is that they prevent the development of pathogenic microflora, and can also synthesize biologically active substances such as vitamins, amino acids, and enzymes, modulate the immune system, while increasing digestibility and utilization of nutrients. The addition of Lactobacillus acidophilus, Lactobacillus salivarius and Lactobacillus plantarum at a concentration of 107-108 CFU/g (colony forming units) reduces the incidence of diarrhea in young calves (Signorini et al., 2012). Probiotic microorganisms create a physical barrier between the intestinal epithelial cells and their contents. Some intestinal probiotics (such as Lactobacillus, Bifidobacteria, several strains of Escherichia coli, and new-generation probiotics including Bacteroides thetaiotaomicron and Akkermansia muciniphila) can support intestinal epithelial homeostasis and promote health (Liu et al., 2020). In addition, probiotic bacteria produce short-chain fatty acids, which lead to a decrease in pH (Maldonado Galdeano et al., 2019). In the intestine, short-chain fatty acids are mainly produced by bacterial fermentation of cellulose. Therefore (Liao et al., 2024) suggest that safe and effective cellulolytic bacteria may become new probiotics. Recently, microbiome studies have identified a number of putative psychobiotic strains, including SCFAs. SCFA-producing bacteria have attracted particular attention from neuroscientists. Recent studies have shown that SCFA-producing bacteria, such as Lactobacillus, Bifidobacterium, and Clostridium, have very specific functions in various psychiatric disorders, suggesting that these bacteria may be potential new psychobiotics (Cheng et al., 2022). Probiotics increase the digestibility of the feed consumed by animals; For example, a study in buffalo calves showed that probiotic feed containing Lactobacillus acidophilus can provide higher dry matter absorption and nutrient digestibility compared to a group of animals whose diets lacked probiotic (Sharma et al., 2018). In another study (Boyd et al., 2011), the use of a mixture of *Lactobacillus acidophilus* NP51 and *Propionibacterium freudenreichii* NP24 as a feed additive for dairy cows improved the digestibility of fiber and crude protein by 7.6%. Similar results were obtained in poultry with regard to increased digestibility of nutrients, essential amino acids and calcium (Khalid et al., 2021; Maas et al., 2021). Recently, probiotics have also been used in sheep farming in numerous studies; Khalid et al., (2021) reported that adding probiotics to the lamb diet improves animal growth during the suckling period. In the study Shoukry et al., (2023), adding probiotics to the main diet also improved lamb growth. Studies conducted by Indian scientists in the Nellore province indicate that adding probiotics to the ram diet improves feed utilization and average daily live weight gain of animals (Khalid et al., 2021). Feeding probiotics to Barki lambs has been shown to have a positive effect on growth, nutrient absorption, and overall economic efficiency (Darwish, 2025).

An important feature of probiotics is their immunostimulatory properties (Bilal et al., 2021; Kong et al., 2020). Recently, the mechanisms of action of probiotics on the immune system have been actively studied by various scientists (Anee et al., 2021; Kober et al., 2022). It has been proven that probiotics can affect immune functions in different ways. Thus, a number of studies have been conducted on the effect of different probiotic strains on the immunological profile of the animal body (Kulkarni et al., 2022). Probiotic supplements can promote growth and development of immune systems in ruminants by regulating the structure, abundance, and fermentation levels of the gastrointestinal microbiota (Ban and Guan, 2021; Mao et al., 2023).

Based on the reports Escobedo-Gallegos et al., (2023) there is evidence of the positive effect of probiotics on relieving stress in animals, particularly heat stress. Thus, Kotsampasi et al. (2023) indicate that the use of probiotics (direct-feeding microorganisms) by ruminants in stressful environmental conditions alleviated heat stress and increased animal productivity. As indicated by Shah et al. (2025) the addition of a probiotic preparation to the diet of bulls contributed to the avoidance of heat stress and increased nutrient digestibility, which was positively reflected in the average daily weight gains. The mechanism of how probiotics migrate heat stress in animals can be explained by a decrease in the concentration of cortisol in the blood, which was recorded in the study by Binuni Rebez et al., (2025), where in lambs that were exposed to heat stress and did not receive a probiotic, a tendency toward lower cortisol concentration was observed.

The most promising probiotics are preparations with spore-forming bacteria of the genus *Bacillus*, as they are prominent representatives of exogenous microflora. A wide range of strains of this genus have been studied as therapeutic agent for the prevention and treatment of intestinal infections: *B. cereus*, *B. polymyxa*, *B. coagulans*, *B. brevis*, *B. megaterium*, *B. pumilus*, *B. laterosporus* etc. However, the most fully and comprehensively studied species *B. subtilis* and *B. licheniformis*. Aerobic spore-forming bacteria of the genus *Bacillus* are widely distributed in nature. Despite the fact that Bacillus was described more than 100 years ago, the taxonomy of the genus is still developing and improving (Bilal et al., 2021).

One of the most promising strains in the production of probiotics is *Bacillus subtilis*. The alternative name for *B. subtilis* is *hay bacillus*, since for the first time the cumulative culture of this organism was obtained from hay extract. In 1835, Christian Gottfried Ehrenberg first described this strain, but in his interpretation this microorganism was called *Vibrio subtilis*, later in 1872 *B. Subtilis* received its modern name (Bilal et al., 2021). Another promising culture is *Bacillus Licheniformis* or *B. Licheniformis*. It is a gram-positive, mesophilic bacterium. The principle for the action of *Bacillus Licheniformis* is its ability to regulate bacterial colonies, contributing to the formation of normal microflora in the body (Makowski et al., 2021). Thus, Onubi et al., (2015) probiotics containing *Bacillus Licheniformis* have been shown to improve the absorption of certain nutrients (calcium, zinc, and vitamin B₁₂) and reduce the incidence of anemia, preventing infections and micronutrient deficiencies.

The new domestic probiotic Immunobacterin-D (the supplement used in this study) contains bacteria of the genus *Bacillus subtilis* and *Bacillus licheniformis*, indicating excellent potential properties in terms of impact on the animal body. Thus, according to Sidashova et al. (2022), the use of the probiotic Immunobacterin-D on bees had a positive effect, increasing the live weight of worker bees by the end of the beekeeping season by 9.15% compared to the control. This can increase the viability of bees during wintering. Also, Pogranichniy et al. (2023) reported that probiotic supplement increased average daily milk yield and milk fat content, improved cow health, and enhanced stress resistance—evidenced by a normalized AST/ALT ratio, which supports liver cell renewal. In addition, cows receiving Immunobacterin-D had a significant increase in the content of phosphorus and total bilirubin in the blood serum. Rybachuk et al. (2020) found that adding Immunobacterin-D to German Shepherd dogs' diets normalized serum calcium levels—significantly increasing them after 14 days—and led to a marked decrease in ALT activity, indicating improved hepatocyte function likely due to reduced liver antigenic and toxic load from the symbiotic action of *B. subtilis* and *B. licheniformis*

However, based on the available literatures, no similar studies on the use of the probiotic supplement Immunobacterin-D conducted on sheep, in particular lambs during the suckling period, have not been conducted in Ukraine. Therefore, the purpose of this study was to study the impact of the use of the therapeutic and prophylactic probiotic Immunobacterin-D on the growth and development of dairy lambs of the Lacon breed and clinical indicators of their body during the suckling period.

MATERIALS AND METHODS

Ethical regulation

Rules for handling animals in experiments fully comply with European legislation (Directive of the Council of the European Union No. 98/58/EC, 1998; Nalon and Stevenson, 2019). The research protocol for biochemical parameters blood of sheep was approved by the local commission on bioethics of "Ascania Nova" Institute of Animal Breeding in the Steppe Regions named after M. F. Ivanov - National Scientific Selection-Genetics Centre for Sheep Breeding, Ukraine, in accordance with Good Clinical Practice (GCP) for the protection and humane treatment of experimental animals.

Materials

A scientific experiment was conducted on dairy lambs of the Lacon breed under the conditions of the production and commercial company (PCC) "Piligrim" of the Khmelnytskyi region to assess lamb growth resistance during the suckling period using the probiotic "Immunobacterin-D" as an immunomodulator. Two groups of ewes with lambs were formed using the group-analogue method (10 ewes and 10 lambs in each).

Components of probiotic supplement

1 kg of the preparation contains active ingredients: bacteria of the genus *Bacillus subtilis*, *Bacillus licheniformis*, not less than 6×1012 CFU/kg (6×109 CFU in 1 g), xylanase 300,000 units/kg, protease 5,000 units/kg, amylase 1,000 units/kg.

Methods

Newborn lambs of the experimental group, starting at 7-10 days of age, were given the prophylactic drug Immunobacterin-D at a dose of 50 g/100 kg mixed with concentrated feed. The mixture (Immunobacterin-D + concentrated feed) was prepared as follows: 25 grams of Immunobacterin-D was thoroughly mixed with 100 g of concentrate. Then, 1 kg of concentrate was added to the mixture and mixed well. Next, concentratewas added to reach 10 kg and mixed thoroughly again. Finally, more concentrate was added to bring the total mass to 100 kg, with all components thoroughly blended. The control group did not receive the probiotic. The experiment continued until the experimental lambs reached 2 months of age.

Live weight of lambs was determined by individual weighing at the start and end of the experiment. Body structure was studied at 2 months of age during weaning. Seven main measurements were taken: height at the withers; height at the sacrum; oblique body length; chest width; chest depth; chest circumference behind the shoulder blades; and pastern circumference. For a more detailed description of the animals and their development stage, the following indices were calculated from these measurements: massiveness; compactness; chest development; stretch; boniness; long-leggedness; and deep-chestedness (Zonabend König et al., 2017). Blood for analysis was collected from the jugular vein of three lambs group before morning feeding. The following indicators were determined: AsAT, AIAT, GHT, Alkaline phosphatase, Bilirubin, Creatinine, Urea, Total protein, Albumin, Glucose, Calcium, Potassium (Polizopoulou, 2010).

Lamb adaptability was assessed by measuring body temperature and breathing rate, recorded alongside weather conditions (air temperature and relative humidity). Body temperature was measured rectally with an electronic thermometer. Breathing rate was the count of chest movements per minute (inhalation acts) while the animal was calm. Pulse rate was measured by counting heartbeats per minute on an artery near the heart. Clinical and climatic parameters were recorded over two consecutive days at 6:00 a.m. and 2:00 p.m. (Mykytyuk et al., 2021).

The coefficient of thermal sensitivity, coefficient of thermal vulnerability and heat resistance index (HRI) in young sheep were calculated as follows. The coefficient of thermal sensitivity of the body was calculated using the formula M. V. Benezra (Mykytyuk et al., 2021):

 $I = \frac{T_2}{39,5} + \frac{RR}{65}$ where, T₂: body temperature at OC during thermal stress; RR: respiratory rate per minute under

thermal stress; 39,5 i 65: average values of body temperature and frequency of respiratory movements of sheep in optimal conditions.

The coefficient of thermal vulnerability of the animal body was measured using A. F. Dmitriev's method (Mykytyuk et al, 2021):

 $C_{TV} = \frac{T_D}{T_M} + \frac{R_D}{R_M}$ where, C_{TV} is the coefficient of thermal vulnerability; T_D: daytime body temperature of animals;

 T_{M} – animal body temperature in the morning; R_{D} – respiratory rate per minute during the day; R_{M} – respiratory rate per minute during morning.

The heat resistance index was calculated using Y. O. Rauschenbach's method (Mykytyuk et al, 2021):

 $ITC = 2 \times (0.5 \times t_2 - 10 \times dt + 30)$

where, ITC: heat resistance index; t₂: temperature of the medium under thermal stress; dt: the difference in body temperature during the day at high ambient temperatures and in the morning in the thermoneutral zone.

Statistical analysis

Biometric data were processed using MS Excel software and statistical functions according to S. S Kramarenko algorithms (Kramarenko et al., 2019) with the determination of the arithmetic mean, its error, as well as the statistical reliability of the studies conducted.

RESULTS AND DISCUSSION

Studies on the effectiveness of the probiotic Immunobacterin-D were conducted on lambs of the Lacon breed. This breed belongs to the dairy production. And is considered the best dairy sheep breed in France. The live weight of breeding rams is 80-100 kg and ewes are 50-70 kg. The animals are characterized by high precocity (up to 60% of ewes can be mated at 7-10 months of age). A feature of these sheep is early weaning of lambs; afterweaning, the ewes begin milking. The average yield of marketable milk over 150 days of lactation is 155-160 (Kudryk et al., 2024).

The therapeutic and prophylactic preparation Immunobacterin-D is a white powder that dissolves well in water. The probiotic consists of *Bacillus subtilis* and *Bacillus licheniformis* at a concentration of not less than 6×10^{12} CFU/kg (6×10^9 CFU/g). The mechanism of action is that *Bacillus* bacteria inhibit the growth of opportunistic microorganisms, provide partial destruction of mycotoxins, contribute to restoration of microflora, and enhance feed digestion, thereby improving feed conversion, productivity, and animal health. It should be noted that the farm where the studies were conducted was epizootically safe regarding infectious gastrointestinal diseases.

As the results showed, use of Immunobacterin-D by the experimental group had a significant effect on live weight (Table 1). At birth, lamb live weight was 3.5 ± 0.11 kg in the control group and 3.4 ± 0.10 kg in the experimental group. At 2 months of age, control lambs weighed 16.6 ± 0.17 kg, while experimental lambs weighed 18.1 ± 0.15 kg, with average daily gains of 218 ± 2.1 g and 253 ± 4.5 g, respectively (P ≤ 0.001).

Table 1 - Growth indicators of Lacon lambs during the suckling period.							
	Experimenta	Compared to					
Indicator	Control	Experimental	gro	group			
	Control	Experimental	+/-	%			
Live weight at birth, kg	3.5±0.11	3.4±0.10	-0.1	-2.9	0.71		
Live weight at 60 days of age, kg	16.6±0.17	18.6±0.15***	+2.0	+12.0	8.85		
Absolute gain, kg	13.1±0.12	15.2±0.14***	+2.1	+16.0	11.41		
Average daily gain, g	218±2.1	253±4.5***	+35.0	+16.1	7.05		
± to control, %	14,9	16.1	1,2	8,1	0,32		
Relative gain	374±11.73	447±15.59**	+73.0	+19.5	3.74		
Marke: * P<0.05: ** P<0.01 ***P<0.001							

Thus, the advantage of the experimental group using Immunobacterin-D was 16.1%. Since absolute and average daily increases in body weight per unit of time do not always objectively characterize growth rate, the relative growth rate is used, expressed as a percentage. It was found that relative growth of lambs in the control group was 374%, while in the experimental group it was 447%, an increase of 73 percentage points. During the experimental period, isolated gastrointestinal disorders were noted in the control group, whereas lambs in the experimental group exhibited no such disorders. Probiotic bacteria in Immunobacterin-D colonized the intestines of newborn experimental lambs with beneficial microflora, creating a barrier against opportunistic pathogens. In addition to changes in body weight, basic body measurements were taken at 2 months of age to objectively assess animal growth (Table 2).

It was found that lambs in the experimental group exceeded their peers in the control group by 0.7 cm, or 1.32% in height at the withers; by 1.1 cm, or 2.04% in height at the sacrum; by 0.8 cm, or 1.42% in oblique body length; by 1.0 cm, or 5.95% in chest width; by 1.2 cm, or 5.0% in chest depth; by 1.6 cm, or 2.09% in chest circumference behind the shoulder blades. Based on the obtained mean ANOVA or, body structure indices were calculated in the control and experimental groups (Table 3). Thus, lambs in the experimental group exceeded those in the control group in the indices ofmassiveness by 1.1%; compactness by 0.9%; chest development by 0.6% and deep chest by 1.7% ($P \le 0.05$). Body composition indices indicate that the animals developed proportionally in accordance with ontogenetic changes in body mass during growth.

It is known that the use of probiotics can improves blood parameters and the, antioxidant and immune systems in animals (Anee et al., 2021). Therefore, during the experiment, the aim was to perform a biochemical analysis of the blood of the experimental animals, as reflected in Table 4. It was found that lambs in the control group had a total serum protein content of 53.35 g/l, lambs in the experimental group had 61.80 g/L, a 15.8% increase. This suggests a sufficient
amount of structural material to support weight gain. Globulins are of particular interest. This is a significant group of proteins of various structures with important biological functions. The level of globulin proteins influences young animal's future productivity and immune defenses. Thus, the total serum globulin concentration in the control group was 21.31 g/L, while in lambs receiving the probiotic Immunobacterin-D it reached 28.86 g/L, (a) 35.4 % increase, demonstrating improved resistance ($P \le 0.05$). Domestic sheep exhibit high adaptive capacity to diverse climates and feeds, enabling widespread farming across Ukraine, yet even they can suffer productivity losses under heat stress, particularly with global warming. Markers of adaptive capacity in Lacon lambs treated with Immunobacterin-D, measuring body temperature, respiratory rate, and pulse under thermoneutral and heat-stress conditions (Table 5).

Table 2 - Body measurements of Lacon lambs.								
	Experimenta	l animal groups	Compare					
Indicator	Control	Evnerimental	control group		td			
	Control	Experimental	+/-	%				
Height at the withers	53.2±0.58	53.9±0.68	+07	+1.32	0.80			
Height at the sacrum	53.8±0.66	54.9±0.49	+1.1	+2.04	1.34			
Oblique body length	56.2±0.58	57.0±0.55	+0.8	+1.42	1.01			
Broadness of the chest	16.8±0.37	17.8±0.66	+1.0	+5.95	1.33			
Depth of the chest	24.0±0.32	25.2±0.58	+1.2	+5.00	1.82			
Circumference of the chest behind the shoulder blades	76.4±1.03	78.0±0.95	+1.6	+2.09	1.14			
Height at the withers	8.4±0.24	8.4±0.24	0	0	0			
* P≤0.05; ** P≤0.01 ***P≤0.001								

Table 3. Indexes of body structure of lambs of the Lacon breed.

	Experimenta	l animal groups	Compare		
Indicator	Control	Experimental	contro	td	
	Control	Experimental	+/-	%	
Massiveness	143.6±0.57	144.7±0.49	1.1	+0.77	1.47
Compressedness	135.9±0.59	136.8±0.54	0.9	+0.66	1.13
Thoracic	70.0±0.90	70.6±1.25	0.6	+0.86	0.39
Stretchiness	105.6±0.05	105.8±0.65	0.2	+0.19	0.31
Boneness	15.8±0.31	15.6±0.30	0.2	-1.3	0.47
Long-leggedness	54.9±0.41	53.2±0.68	1.7	-3.1	2.15
Deep-chest	45.1±0.37	46.8±0.49*	1.7	+3.77	2.78
* P≤0.05: ** P≤0.01: ***P≤0.001					

Table 4 - Biochemical analysis of blood of Lacon lambs at 2 months of age.

	Experiment	al animal groups	Compare		
Indicator	Control	Evnorimental	control	td	
	Control	Experimental	+/- %		
AST, IU/L	174.5±15.09	166.4±17.38	-8.1	-4.6	0.35
ALT, IU/L	58.43±4.77	47.9±12.89	-10.53	-18.1	0.77
GGT, IU/L	66.68±6.53	81.33±4.50	+14.65	+21.9	1.85
Alkaline phosphatase, IU/L	777.0±31.98	790.4±18.64	+13.4	+1.7	0.36
Bilirubin, µmol/L	10.54±0.38	9.3±0.27*	-1.24	-11.8	2.69
Creatine, µmol/L	129.2±11.64	119.6±7.66	-9.6	-7.4	0.69
Urea, µmol/L	10,0±1.7	8.9±0.77	-1.1	-11.0	0.83
Total protein, g/L	53.35±2,14	61.8±2.59	+8.45	+15.8	2.52
Albumin, g/L	31.85±1.01	32.9±2.50	+1.05	+3.3	0.39
Glucose, g/L	5.41±0.52	5.5±0.27	+0.09	+1.7	0.16
Calcium, µmol/L	2.92±0.15	3.2±0.07	+0.28	+9.6	1.64
Potassium, µmol/L	6.29±0.38	5.8±0.54	-0.49	-7.8	0.74
Globulins, g/L	21.31±2.70	28.86±0.32*	+7.55	+35.4	2.78
* P<0.05· ** P<0.01· ***P<0.001					

Indicator		Experimenta	l animal groups	Compared to the control group		
		Control	Experimental			td
		Control	Lycimenta	+/-	%	
6.00	Body temperature, OC	38.9±0.09	39.0±0.09	+0.1	+0.26	0.83
(21.00)	Respiration rate, movement/min	64.0±2.31ª	65.3±1.33	+1.3	+2.03	0.49
(21 °0)	Pulse rate, beats/min	73.3±2.40ª	74.7±1.33	+1.4	+1.91	0.51
14.00	Body temperature, OC	39.7±0.33	39.7±0.09	+0.8	-	-
(37 0C)	Respiration rate, movement/min	81.3±1.33	84.0±2.31***	+20.0	+28.6	5.06
(31 -0)	Pulse rate, beats/min	97.3±1.33	101.3±3.53***	+28.0	+38.2	10.46
* P≤0.05; **	[•] P≤0.01; ***P≤0.001					

Table 5 - Clinical indicators of physiological functions of lambs of the Lacon breed.

It was found that in the morning (21 °C) lambs of the control and experimental groups had body temperature of 38.9 °C and 39.0 °C, respectively. And body temperature increased: control lambs by 0.8 °C and experimental lambs by 0.7 °C. Respiration is the main vital process providing gas exchange between a living organism and the environment. In our study, an increase in air temperature to 37 °C was accompanied by an increase in respiratory rate in lambs of the control and experimental groups by 27.0% and 28.6% (P \leq 0.001), compared to their morning rates at lower temperatures. Lambs that consumed the probiotic adapted better to elevated temperatures as their respiratory rates were higher than those of control lambs. Based on physiological data, indices and coefficients characterizing physiological adaptation were calculated (Table 6).

The heat resistance index, along with the heat sensitivity coefficient, is an objective indicators of animal responses elevated temperatures. Lambs in the experimental group receiving the probiotic Immunobacterin-D outperformed control lambs by 2.47%, which indicates their enhanced heat resistance. Animals that suffer less from heat stress have a higher growth rate. Regarding the heat sensitivity coefficient and the heat vulnerability coefficient no significant differences were observed between groups. These results warrant further study during both the suckling and intensive fattening phases. This will allow us to more accurately determine the role of probiotics in reducing heat stress in sheep. In addition, the economic efficiency of using the probiotic "Immunobacterin-D" in two-month-old lambs was evaluated (Table 7).

It was established that the use of the therapeutic and prophylactic drug "Immunobacterin-D" on Lacon lambs during the suckling period (60 days) had a positive effect on their growth dynamics They achieved a 9.0 % increase in live weight and a 16.0 % increase in absolute gain, corresponding to an additional \$6.88 per kilogram of gain.

Table 6 - Indicators of the adaptive ability of Lacon lambs.							
	Experimenta	l animal groups	Compare				
Indicator	Control	Experimental	control group		td		
			+/-	%			
Heat resistance index	81.0±2.00	83.0±0.67	+2.0	+2.47	1.28		
Coefficient of thermal sensitivity	2.26±0.02	2.30±0.04	+0.04	+1.77	0.91		
Coefficient of thermal vulnerability	2.29±0.05	2.31±0.02	+0.02	+0.87	0.38		
* P≤0.05; ** P≤0.01; ***P≤0.001							

Table 7 - Economic efficiency of using the probiotic "Immunobacterin-D" during the suckling period.

	Experimental animal groups		
Indicator	Experimental animal Broupe	Control	Experimental
Live weight at birth, kg		3.5±0.11	3.4±0.10
Live weight at 75 days of age, kg		16.6±0.17	18.1±0.15 (+9.0%)
Absolute gain, kg		13.1±0.12	15.2±0.14 (+16.0%)
Consumption rate of "Immunobacterin-D" accord recommendations	ing to the manufacturer's	-	50 g per 100 kg of compound feed
Compound feed used in 60 days		10.5	10.5
Price of 1 kg of "Immunobacterin-D"		-	700
Use of probiotic "Immunobacterin-D":		-	6.0
P. Additional products were obtained compared to t	the control	-	0.1
Live weight, kg Cost (1 kg of live weight / 3.8 \$), \$.		-	2.1 6.95
Profit from the use of "Immunobacterin-D", \$, pe	r kg of gain		6.88

DISCUSSION

Prolonged antibiotics use leads to resistant bacteria and antibiotic residues in livestock and fish. Consequently, finding effective alternatives has become urgent. Currently, the most suitable alternative to antibiotics is probiotics. Probiotics are live microorganisms that, when consumed in optimal amounts, provide health benefits. They consist primarily of beneficial bacteria and yeasts that inhibit pathogens, enhance immunity, and restore gut microbial balance. Probiotics eliminate pathogens through multiple molecular mechanisms and modulate the host's immune response promote animal well-being (Anee et al., 2021).

Thus, Maldonado Galdeano et al. (2019) indicate that probiotics improve health by inhibiting harmful bacteria. For example, Lactobacillus rhamnosus and Lactobacillus plantarum can prevent the adhesion of Escherichia coli in the intestinal tract. Kawai et al. (2004) approved, that bacterias such as Lactobacillus, Leuconostoc, Pediococcus, Lactococcus, Enterococcus, Streptococcus, Bifidobacteria can produce proteins or bacteriocins that minimize the development of pathogenic microflora. In other words, these probiotics reduce the number of harmful microorganisms in the gastrointestinal tract of animals.

In addition, probiotics enhance host immunity by modulating the immune system. Consumed probiotics stimulate the mucosal immune system (MIS) and induce signaling networks. Antigen-presenting cells identify bacterial components and trigger, primary immune responses leading to T- and B-cell development. They activate immunity via cytokines secretion that stimulates T-cell activation. Signals from dendritic cells determine T-helper polarization or T-regulatory responses, which in turn shape B-cell responses against pathogens (Van Zyl et al., 2020). Probiotics also increase digestion rates by enhancing gastrointestinal enzyme activity and improving feed digestibility (Sharma et al., 2018).

Probiotics play a significant role in ruminants. Ruminantsingest large amounts of protein and carbohydrates, which rumen microbes break down. Ruminant probiotics containing *Saccharomyces cerevisiae*, *Lactobacillus*, *Aspergillus oryzae*, *Bacillus*, and *Enterococcus* confer substantial health benefits (Elghandour et al., 2020). They also boost dairy cattle milk yield: *Bacillus subtilis*, *S. cerevisiae*, and *Enterococcus faecalis* increase secretion, and *Bifidobacterium bifidum* suppresses milk allergies (Ma et al., 2020; Jing et al., 2021).

A significant positive effect of probiotics in sheep farming has also been established. Saleem, et al. (2016) indicate that probiotics improve the health of sheep and lambs. According to Kritas et al. (2006), probiotic feed components can improve the quality of sheep milk. Feed additives, including *Bacillus subtilits* and *Bacillus licheniformis*, help reduce mortality, increase the protein content in milk and enhance overall milk production in sheep.

The effect of probiotic substances on reducing stress in sheep, particularly heat stress, has also been established. As the climate in Ukraine has changed significantly in recent decades, researchers are increasingly recording abnormally high temperatures (Vozhegova, 2021). Therefore, according to Estrada-Angulo et al., (2021), the use of probiotics in lambs is recommended to mitigate heat stress in hot environmental conditions. Heat stress can lead to imbalances in the rumen and its function, an increased risk of acidosis, greater energy expenditure to maintain vital body functions, and reduced immunity (Marai et al., 2007). When exposed to high environmental temperatures, sheep that consume probiotics activate a series of physiological compensatory mechanisms such as increased rectal temperature, respiratory rate, and heart rate which help the body adapt to extreme conditions (Mc Manus, 2020).

CONCLUSION

It was found that the use of the probiotic Immunobacterin-D in the lactation period of Lacon dairy lambs provided an increase in average daily gains of 16.1%, an increase in total protein content of 15.8% and an increase in globulin proteins of 35.4%, indicating enhanced lamb resistance. Immunobacterin-D in lamb diets positively affected growth rate and development, improved health and adaptive capacity, and conferred greater heat resilience compared to unsupplemented lambs.

DECLARATIONS

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Authors' contribution

A.Tsvihun and Viktor Yakovchuk participated in the design of the study, performed the experiments, and wrote the original manuscript. L.Ponko assisted in data organization and calculations. O.Yulevich coordinated the research methodology. L.Ponko and O.Karatieieva edited the manuscript. O.Karatieieva and P.Ponichtera made final revisions of the manuscript. All authors read and approved the published version of the manuscript.

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Data availability

The datasets used and/or analysed during the current study available from the corresponding author on reasonable request.

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Competing interests

The authors did not declare any competing interests.

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DETERMINATION OF BREWERY YEAST-TREATED CROP RESIDUES AS ANIMAL FEED RESOURCE

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Supporting Information

ABSTRACT: The study evaluates the effects of brewery yeast to improve the nutritional value and in vitro digestibility of selected crop residues in Dessie Town, Amhara region, Ethiopia. The experiment was laid out in a completely randomised design (CRD) with a 3×5 factorial arrangement, i.e., three-selected crop residues (Teff straw, maize, and sorghum stover) with five levels of fermentation periods (0, 3, 6, 9, and 12 days). The mean crude protein (CP) values of brewery-spent yeast (BSY) treated Teff straw, maize stover, and sorghum stover treated crop residues were 7.85%, 14.3%, and 14.78%, respectively. Among proximate and detergent values of the interaction effect of fermentation period and crop residue type, dry matter (DM), crude protein (CP), estimated metabolizable energy (EME), acid detergent lignin (ADL), and neutral detergent fiber (NDF) content were significantly (p < 0.01) varied at different fermentation periods. While ash, organic matter (OM), and acid detergent fiber (ADF) content were highly significant (p < 0.001) variations between the interaction of crop residues and the fermentation period. The value of in vitro dry matter digestibility (IVDMD) and in vitro organic matter digestibility (IVOMD) of BSY treated crop residue was significantly (p < 0.001) varied for the interaction of crop residue type and fermentation period. The EME, IVDMD, and IVOMD of BSY treated crop residues were observed in the range of 7.54-10.64 MJ/kg, 54.44%-61.46%, and 47.14%-66.48%, respectively. Therefore, the study investigates how brewery yeast can enhance the nutritional quality and in vitro digestibility of certain crop residues in Dessie Town, Ethiopia. It followed a random design with three crop residues (Teff straw, maize stover, and sorghum stover) and six fermentation periods. Treated residues showed varying crude protein values and significant differences in in vitro digestibility and energy content. Therefore, utilizing brewery-spent yeast (BSY) presents a cost-effective and sustainable approach to enhancing the nutritional value and in vitro digestibility of crop residues. Further studies may explore the scalability of BSY treatments and their effects on livestock performance.

Keywords: Brewery spent yeast, Crop residue, In vitro digestibility, Nutritional value.

INTRODUCTION

Ethiopia is home to a huge number of livestock due to its vast and diverse agro-ecological zones and the significance of livestock in subsistence strategies. Indeed, Ethiopia has the largest livestock population in Africa, with 70 million cattle, 42 million sheep, 52 million goats, 8 million camels, and 56 million chickens (CSA, 2022). Despite the huge numbers and multiple roles of livestock, productivity remained very low in Ethiopia and unable to meet the demands of the rapidly growing population (Yigezu Wendimu, 2021). Numerous constraints are often associated with this, such as scarcity of feed, high incidence of diseases and parasites, limited genetic potential of local breeds, poor veterinary care, restricted credit availability, scarcity of land, and poor management techniques in all livestock production systems (Welay et al., 2018). Among these limits include low quality and insufficient amount of feed availability, particularly during the dry season, were identified as the major causes of low livestock productivity and it accounts about 80% of the overall production cost (Duguma and Janssens, 2021).

Ruminant diets in Ethiopia are generally based on roughage feeds especially crop residues like Teff straw, maize and sorghum stover. The intake, in vitro digestibility and nutrient absorption of the crop residues are very poor because of high cell wall content and low essential nutrients. The nutrient content of crop residue cannot even support the maintenance requirements of the animals and consequently, the performance of an animal feeding crop residue is very low. Improving the nutritive value of the poor-quality feed resources is one option for proper utilization. Intake and utilization of low-quality roughage can be improved by different methods of treatments technologies (Duguma and Janssens, 2021).

Brewer's spent yeast (also known as residual yeast or excess yeast) is a common by-product of the brewing industry that is produced when the yeast used in fermentations is no longer usable and must be discarded (Jaeger et al., 2020). In recent years, there has been growing awareness in Ethiopia regarding the importance of improving the utilization of crop residues to address feed shortages and enhance livestock productivity (Duguma and Janssens, 2021). The nutritional value and in vitro rumen fermentation features of a study conducted on yeast-fermented maize dust and cassava pulp at several ratios (40:60, 20:80, and 0:100%) and ensiled for 15 days are improved (Ratchataporn et al., 2022). In accordance with Kim et al. (2015), the liquid brewery spent yeast mixed with cassava pulp at a ratio of 30%:0% increases

the chemical composition and reduces *in vitro* gas production. Similarly, the use of brewery-spent grain (20%) into corn treated crop residue and total mixed ration boost in vitro digestibility and fermentation characteristics.

Treating crop residues through fermentation with brewery-spent yeast (BSY), can enhance their nutritional value, increase in vitro digestibility, and improve their suitability as animal feed (Terefe 2022, and 2023). This approach contributes to addressing feed shortages, particularly during the dry season, while also promoting the sustainable use of agricultural by-products. Teff straw, maize stover, and sorghum stover was likely based on their widespread availability in the study area (Dessie Town) and their significance as staple crop residues in Ethiopian livestock systems. The fermentation periods were systematically assessed; fermentation duration influences the nutritional improvement of the residues. These intervals provide insights into the dynamic changes in crude protein, in vitro digestibility, and energy content over time, helping to identify the optimal fermentation period for practical application (Terefe et al., 2022).

In Kombolcha Brewery industry, there is a huge amount of brewery-spent yeast dumping daily in the local environment. This may contaminate the local environment and cause public health hazard. However, this resource has an alternative advantage in enhancing the nutritionally poor contents of the roughage feed sources as the result of additive effect of the different inclusion rate. It is essential to create suitable methods of utilizing BSY as animal feed due to its affordability, high nutrient concentration (Jaeger et al., 2020). Hence, mixture of brewery yeast cell on poor quality forage and high grain diets can increase the nutritional value of the ration (Aubrey, 2017; Yadessa et al., 2023). Therefore, the main aim of this study was to investigate the effects of brewery yeast on the chemical composition and in vitro digestibility of selected crop residues fermented at different time.

MATERIALS AND METHODS

Description of the study area

The selected crop residue type (Teff straw, maize and sorghum stover) was collected from Dessie town, South Wollo Zone of Amhara Region. It is found in the north-central part of Ethiopia at a distance of 401km from Addis Ababa, which is the capital city of the country (Figure 1). Its topography varies from 2400-3000 meters above sea level. Geographically it lies on the intersection of 11°8′N and 39°38′E. Dessie town is subdivided into 5 sub-cities, 18 urban kebeles and 8 rural kebeles. It is located within the highland (Dega) agro-climatic zone and it has a bimodal rainfall distribution where the major annual rain fall season occurs in winter (Meher) (May to end of September) and short rainy season occur in autumn and spring (Belg) (beginning January to April). Dessie typically has mean annual rainfall is 1120 mm and mean annually temperature ranges from 15.0°C (59.0°F) to 26.78°C (80.2°F).

Dessie town is located within Dessie Zuria District and suitable for the production of different livestock and crop production. Small-scale farming is considered the most prevalent method for maintaining a subsistence economy at the household level. Farmers usually own draught animals. The crops are produced during two cropping seasons, *Meher* and *Belg*. During *Belg* season, the most important crops are barley, pea, whereas, wheat, maize, bean, and Teff are the most important one in *Meher* season. Based on the district agriculture office, the livestock population of the Dessie Zuria District has 22,831 cattle, 3,179 goats, 25,875 sheep, 1,259 horses, 578 mules, 5,656 donkeys, 1,155,661 hens and 619 bee colonies are found as documented.



Experimental materials and sample preparation

Liquid brewer spent yeast (LBSY) was obtained from BGI Kombolcha Brewery Industry, Ethiopia. This material was collected with plastic buckets (20 litters), transported in airtight Jerica, and packed until utilized via proper experimental procedure. Liquid brewery spent yeast (LBSY) was stored for approximately 12 hours and allowed to cool before diluted with water in a 1:5 ratios, respectively (Terefe et al., 2022). The crop residues such as Teff straw, maize and sorghum stover was obtained from Dessie town. Liquid brewery spent yeast (LBSY), distilled water (DW), polyethylene plastic bag (PPB) is used to ensiling the materials, electrical chopper machine (ECM) was used for cutting/chopping the crop residue at recommended sieve size. Homogenizer is used to mixed crop residues with brewery by-products; sensitive balance is used to measuring the exact amount of crop residue or brewery by-product required for experiments; cold-chain (freezer) is used to preserve samples at low temperatures before or after processing to prevent degradation or microbial growth, and crop residue types (CRT) were used.

Experimental design and treatments allocation

This research has an experimental design with two main factors and fifteen treatment combinations (3 crop residue type (CRT) × 5 fermentation period (FP)) to examine the effect of brewery yeast in combination with three crop residues and five fermentation period (0, 3, 6, 9, and 12 days) on nutritional composition, in vitro digestibility (IVDMD), DM and OM contents, and organoleptic quality. This experiment has two-factorial arrangement in a completely random design (CRD). The treatment combinations were considered a 1:1 ratio of three-selected crop residue (Teff straw, maize and sorghum stover) with BSY at five different fermentation periods (0, 3, 6, 9, and 12 days) (Terefe et al., 2022). The control group was assigned as treated crop residue without fermentation. The entire experiments were conducted with five replications, and resulted 75 total observations. The crop residue was chopped at a recommendable sieve size of 2 mm by an electrical chopper machine according to method of Terefe et al. (2022). The mixture of brewery spent yeast, and three (3) dominant crop residues treatment combinations ensiled with five different fermentation time (0, 3, 6, 9, and 12 days) with five replications and which resulted 75 total observations. Polyethylene materials were chosen for the anaerobic ensiling process due to its suitable mechanical characteristics, low costs, and availability.

Table 1 – Completely Randomized Design experimental layout and treatment combinations.								
Fermentation period/days Crop residue type (CRT)	0	3	6	9	12			
	TS01+BSY	TS31+BSY	TS61+BSY	TS91+BSY	TS121+BSY			
	TS02+BSY	TS32+BSY	TS62+BSY	TS92+BSY	TS122+BSY			
Teff straw (TS)	TS03+BSY	TS33+BSY	TS63+BSY	TS93+BSY	TS123+BSY			
	TS04+BSY	TS34+BSY	TS64+BSY	TS94+BSY	TS124+BSY			
	TS05+BSY	TS35+BSY	TS65+BSY	TS95+BSY	TS125+BSY			
	MS01+BSY	MS31+BSY	MS61+BSY	MS91+BSY	MS121+BSY			
	MS02+BSY	MS32+BSY	MS62+BSY	MS92+BSY	MS122+BSY			
Maize stover (MS)	MS03+BSY	MS33+BSY	MS63+BSY	MS93+BSY	MS123+BSY			
	MS04+BSY	MS34+BSY	MS64+BSY	MS94+BSY	MS124+BSY			
	MS05+BSY	MS35+BSY	MS65+BSY	MS95+BSY	MS125+BSY			
	SS01+BSY	SS31+BSY	SS61+BSY	SS91+BSY	SS121+BSY			
	SS02+BSY	SS32+BSY	SS62+BSY	SS92+BSY	SS122+BSY			
Sorghum stover (SS)	SS03+BSY	SS33+BSY	SS63+BSY	SS93+BSY	SS123+BSY			
	SS04+BSY	SS34+BSY	SS64+BSY	SS94+BSY	SS124+BSY			
	SS05+BSY	SS35+BSY	SS65+BSY	SS95+BSY	SS125+BSY			
Total	15	15	15	15	15			
BSY = Brewery spent yeast, CRT= crop residue type, TS = Teff straw.								

Data collection

The data was collected from experimental trails of treated crop residue with BSY. The data were collected from the brewery-spent yeast in combination with crop residue effects on in vitro digestibility (IVDMD and IVOMD), DM contents, NDF, ADF, ADL, EME, CP, OM contents, ash, and organoleptic quality.

1. In vitro dry matter digestibility (IVDMD) and in vitro organic matter digestibility (IVOMD)

- Determined using the Tilley and Terry (1963) two-stage technique.
- First stage: Incubation of samples with rumen fluid for 48 hours at 39°C.

• Second stage: Treatment with pepsin and hydrochloric acid (HCI) to simulate post-ruminal digestion.

2. Dry matter (DM) content

• Samples were dried in a forced-air oven at 105°C for 24 hours until a constant weight was achieved.

3. Neutral detergent fiber (NDF), acid detergent fiber (ADF), and acid detergent lignin (ADL)

• Determined using the Van Soest et al. (1991) detergent analysis system.

• NDF and ADF were extracted with neutral and acid detergent solutions, respectively, while ADL was measured after treating ADF with 72% sulfuric acid.

4. Estimated metabolizable energy (EME)

• Calculated using the formula: EME (MJ/kg) = 0.0157×IVDMD (%)

5. Crude protein (CP) content

• Determined using the Kjeldahl method to measure nitrogen content, followed by conversion to CP using the factor 6.25.

6. Organic matter (OM) content and ash

• Ash was determined by igniting samples in a muffle furnace at 550°C for 6 hours, and OM was calculated as: OM=DM-Ash.

Procedure for data analysis

Data preparation: The raw laboratory data were organized in a structured format, ensuring that all variables (organoleptic evaluation, fermentative characteristics, chemical composition, and in vitro digestibility) were correctly labeled. Data were checked for completeness, consistency, and possible outliers before analysis.

Experimental design and model specification

A. Completely Randomized Design (CRD) was used with a 3 × 5 factorial arrangements to assess the effects of crop residue types (Teff straw, maize stover, and sorghum stover) and fermentation periods (0, 3, 6, 9, and 12 days). Where the model was specified as follows: $yijk = \mu + \tau i + \beta j + (\tau\beta) ij + \epsilon ijk$

Where yijk is a response variable (organoleptic evaluation, fermentative characteristics, chemical compositions, invitro digestibility); μ is an overall mean; τi^{th} is the effect of crop residue types (Teff straw, maize, sorghum stover); βj^{th} is the effects of fermentation period (0, 3, 6, 9, and 12); $(\tau\beta)j^{\text{th}}$ is the effect of the interaction between i and j; ϵijk^{th} is the random error.

Statistical analysis

The laboratory data were subjected to analysis of variance using the general linear model (GLM) procedures of R software (Version 4.3.3). Mean Separation was performed using the least significant difference (LSD) at ($P \le 0.05$). The statistical model was structured to evaluate both the fermentation effects over different durations and the baseline effects of brewery spent yeast (BSY) treatment compared to untreated residues. This approach provided a comprehensive framework for comparison.

Interpretation and reporting

Results were interpreted by assessing main effects (crop residue type and fermentation period) and their interaction effects on response variables. Significant findings were summarized in tables and figures to facilitate comparisons. Statistical assumptions (normality and homogeneity of variance) were checked to validate the results.

RESULTS AND DISCUSSION

Chemical composition of treated crop residues with BSY

The chemical composition of brewery spent yeast treated CRT ensiled under different fermentation period was presented in Table 2. Accordingly, DM content (p < 0.01), ash (p < 0.001), OM content (p < 0.001), and CP (p < 0.01) was significantly varied for the interaction of crop residue type and fermentation period. The DM, ash, OM, and CP contents increased consistently with an increase in the fermentation period. At 12 days of fermentation period, the DM contents of the Teff straw, maize stover and sorghum stover were 96.04, 95.17 and 95.4%, respectively was recorded.

Brewery spent yeast treated Teff straw, maize and sorghum stover have high DM and OM content. It was found between 89% and 94%; Teff straw tends to have a slightly higher OM content compared to maize and sorghum stover. The

DM content of the Teff straw, maize, and sorghum stover were observed in the current study slightly lower than the DM content of 96.23%, reported by Taddess et al. (2016). This suggests that both the type of crop residue and the duration of fermentation period have crucial roles in determining the moisture content retained in the treated CRT. Variations in DM content can affect the storage stability and nutritional quality of the treated CRT. In this study, with high DM content, the OM percentage often remains high, leading to a dense concentration of nutrients in the feed. This is beneficial for feeding livestock as it provides more energy and nutrients per unit of feed.

The CP content of Teff straw, maize stover, and sorghum stover treated CRT was improved due to the addition of BSY; thereby CP content was increased as compared to the untreated group. The CP content of crop residue types treated with BSY was significantly (p < 0.05) difference. This is in agreement with the finding of Abebaye et al. (2020), who reported that green maize stover treated with additives was higher CP content from than the control group. In line with the findings of Bilal (2009), who reported that adding molasses and corn to grass silage and inoculating coffee husks with effective microorganism produced a higher CP content than the control.

The ash content, which indicates the total mineral content, also showed significant variation (p < 0.001) among crop residues and fermentation period. In line with the current findings Yonatan et al. (2011), reported that coffee husk ensiled with additives for varying fermentation times had an increase in ash content. In contrast, the ash content did not exhibit a significant (P > 0.05) difference change across all treatments, according to the findings published by Abebaye et al. (2020). This indicates that the mineral composition of the treated CRT is influenced by both the nature of the crop residue and the fermentation duration. Higher ash content may reflect greater mineral retention or contamination of soil during ensiling.

The current result showed that, fermentation period increases, the DM contents of crop residues were slightly increase. During fermentation, microorganisms such as bacteria and fungi break down the complex carbohydrates, proteins, and other organic compounds present in the crop residues. This breakdown can lead to the production of metabolites like organic acids, which might result in a slight reduction in the moisture content of the material. The ash and CP content of brewery-spent yeast treated CRT were increased with increasing fermentation period runs from zero to twelve days. The organic matter contents of brewery-spent yeast treated crop residues was increased as the fermentation period increased. The organic matter and dry matter contents of treated crop residues types are related each other's. Some changes in the organic matter content were also influence the total dry matter content.

Table 2 - Chemical composition of treated on twith brewery-spent yeast (Dim basis).							
Treatments	Contents	DM	Ash	ОМ	СР		
	0	90.42°	6.66 ^{fg}	92.76 ^{abc}	6.31 ^{fg}		
	3	91.97 ^d	6.68 ^{fg}	93.08 ^{ab}	8.32 ^{ef}		
Teff straw (TS)	6	93.67°	6.91 ^{fg}	93.32 ^{ab}	8.68 ^{ef}		
	9	94.32 ^b	7.25 ^{efg}	93.34 ^{ab}	9.06 ^d		
	12	96.04ª	8.27 ^{bcdef}	94.14ª	9.22 ^d		
	0	91.75 ^d	8.91 ^{bcde}	90.22 ^{fg}	8.95 ^{de}		
	3	92.91 ^d	9.34 ^{abc}	90.25 ^{fg}	11.62 °		
Maize stover (MS)	6	93.02°	9.78 ^{ab}	90.66 ^{efg}	15.98 ⁵		
	9	94.02 ^b	9.76 ^{ab}	91.09 ^{cdef}	16.36 ^b		
	12	95.17ª	10.64 ª	91.31 ^{cdef}	18.32 ª		
	0	90.84°	7.78 ^{cdef}	91.79 ^{bcdef}	9.41 ^d		
	3	92.44 ^d	7.81 ^{cdef}	91.94 ^{bcde}	11.92 °		
Sorghum stover (MS)	6	93.47°	8.06 ^{cdef}	92.19 ^{bcde}	16.28 ^b		
	9	94.67 ^b	8.20 ^{bcdef}	92.22 ^{bcde}	16.66 ^b		
	12	95.41 ª	9.09 ^{abcd}	92.50 ^{abcd}	18.62 ª		
	SEM	0.141	0.328	0.328	0.832		
	P-value	0.01	0.001	0.001	0.01		
es Means with in a column with different superscripts differ at p < 0.05. CRT= crop residue type; FP= fermentation period; DM= dry matter; OM= organic matter; CP= crude protein; SEM= standard error of the mean.							

Fiber composition of treated crop residue with BSY

The neutral detergent fiber, acid detergent fiber and acid detergent lignin of brewery-spent yeast treated crop residue types that ensiled under different fermentation period were presented in Table 3. The brewery spent yeast treated CRT of NDF content (p < 0.01), ADF (p < 0.001) and ADL (p < 0.01) were significantly varied for the interaction of crop residue type and fermentation period.

Contents		NDF	ADF	ADI			
Treatments							
	0	72.32 ª	46.20 ^{cdefg}	7.78 ^{ab}			
	3	71.02 ª	45.67 ^{fg}	7.73 ^{ab}			
Teff straw (TS)	6	70.65 ^{ab}	45.14 ^{fg}	7.60 ^{abc}			
	9	69.34 ^{ab}	45.87 ^{defg}	7.56 ^{abc}			
	12	68.86 ^{ab}	44.09 ^{fg}	6.45 ^{defg}			
	0	57.38 ^{defg}	46.52 ^{cdef}	6.42 ^{defg}			
	3	54.65 ^{efgh}	46.21 ^{cdefg}	5.94 ^{efgh}			
Maize stover (MS)	6	53.98 ^{fgh}	45.85 ^{defg}	5.90 ^{efgh}			
	9	53.49 ^{gh}	45.76 ^{efg}	5.81 ^{efgh}			
	12	51.36 ^h	43.06 ^g	5.25 ^h			
	0	62.13 ^{cd}	52.26 ^b	6.48 ^{defg}			
	3	61.75 ^{cd}	51.92 ^b	5.86 ^{efgh}			
Sorghum stover (MS)	6	61.69 ^{cd}	51.36 ^b	5.68 ^{fgh}			
0 ()	9	59.71 ^{de}	49.46 ^{bc}	5.55 ^{gh}			
	12	58.65 ^{def}	49.07 ^{bcde}	5.29 ^h			
	SEM	0.993	0.65	0.204			
	P-value	0.01	0.001	0.01			
^{a-h} Means with in a column with different superscripts differ at $p < 0.05$ CRT= crop residue type, FP= fermentation period, NDF= neutral detergent fiber ADE= Acid detergent fiber SEM= standard error of the mean							

Table 3 - Fiber composition of brewery-spent yeast treated crop residue (DM basis).

The NDF content, indicative of the cell wall components such as hemicellulose, cellulose, and lignin, showed significant variation (p < 0.01) due to the interaction of crop residue type (TS, MS, and SS) and fermentation period. The value of BSY treated CRT was lower than the control grouped. The value of neutral detergent fiber was higher for Teff straw than maize and sorghum stover. The value of NDF, ADF and ADL was declined as the fermentation period increased. Among the crop residue types, the lower value of neutral detergent fiber, acid detergent fiber and acid detergent lignin was recorded for maize stover than sorghum stover and Teff straw. The observed decline in NDF (Neutral Detergent Fiber), ADF (Acid Detergent Fiber), and ADL (Acid Detergent Lignin) values as the fermentation period increased suggests that fermentation improves the in vitro digestibility of crop residues by breaking down fiber components. This can enhance the nutritional quality of the residues for ruminants. The decrease in NDF and ADF suggests an improvement in the potential in-vitro digestibility of the crop residues over the fermentation period. In line with Abebaye et al. (2020) found that during all fermentation periods, silage treated with molasses and ensiled for six weeks had significantly lower levels of NDF (p < 0.05) value than the control. Similarly, Hosseini et al. (2019), the alfalfa, barely and corn silages that were not treated had higher levels of NDF than silages that had additive treatment. This shows that, fermentation generally reduces NDF content, improving forage nutritional value, digestibility, and intake and increase fiber breakdown. Lower values of these fiber components typically reflect better in vitro digestibility and higher nutrient availability for animals, especially in ruminant diets.

The ADF content, which includes cellulose and lignin, significantly (p < 0.001) varied with the interaction of crop residue type and fermentation period. The ADF value from the present study was lower 12th day that zero days of FP. NDF and ADF are essential for balancing the nutritional needs of ruminants, promoting rumen health, and optimizing overall feed efficiency. Proper management of these fiber components leads to better performance, enhanced energy utilization, and improved long-term health in animals (Carrillo-Díaz et al., 2022).

Acid detergent fiber founds between the ranges of 43.0-56.2%DM and the fermentation period increase the contents of ADF also decrease. The lower value was observed during the 12 days of FP with inclusion of brewery-spent yeast. Similarly, in a related study by Yadessa et al. (2023), reported that, a decreasing in ADF values with the level of BSY increases in the silage materials, it was found to be declining, with the lowest ADF value recorded at 20 and 30% of the BSY inclusion level in the fourth and sixth weeks of the fermentation period. On the other hand, Kamphayae et al. 2016) reported reducing trend of ADF values with the level of brewery spent yeast increases in the silage ingredients. This indicates lower ADF values indicate a higher proportion of digestible nutrients in the feed. This means that animals can more efficiently break down and absorb nutrients like carbohydrates and proteins, leading to better growth and productivity.

In vitro digestibility and energy estimation of treated crop residue with BSY

In vitro dry matter and organic matter digestibility of brewery spent yeast treated CRT was significant (p < 0.001 and p < 0.001) varied respectively and presented in table4. The value IVDMD and IVOMD was statistically significant for

brewery spent yeast treated CRT. The metabolized energy of brewery spent yeast treated crop residue type was significantly (p < 0.001) varied and presented in Figure 2. The value of estimated metabolized energy (EME) was increased as the fermentation period increased. The estimated metabolized energy of treated Teff straw, maize, and sorghum stover was higher than the day zero. At 12 days of fermentation period, the value of Teff straw, maize and sorghum stover were 7.9, 10.64, and 9.64 MJ/kg respectively.

Table 4 - IVDMD and IVOMD of brewery-spent yeast treated crop residue.							
Digestibility parameters (DM basis) Treatments		IVDMD	IVOMD				
	0	53.75 ^h	47.52 ^f				
	3	54.45 ^h	47.71 ^f				
Teff straw (TS)	6	55.05 ^{gh}	48.61 ^f				
	9	55.16 ^{fgh}	48.74 ^f				
	12	55.73 ^{efgh}	49.25 ^f				
	0	61.30 ^{bcd}	62.87 ^{abc}				
	3	61.77 ^{bc}	63.46 ^{abc}				
Maize stover (MS)	6	62.75 ^{ab}	63.49 ^{abc}				
	9	62.79 ^{ab}	64.03 ^{ab}				
	12	65.17 ª	66.48ª				
	0	58.31 ^{defg}	57.23 ^{de}				
	3	58.48 ^{cdef}	58.48 ^d				
Sorghum stover (SS)	6	59.74 ^{bcd}	58.52 ^d				
	9	59.00 ^{cde}	59.57 ^{cd}				
	12	61.46 ^{bcd}	60.33 ^{bcd}				
	SEM	0.66	0.77				
	P-value	0.001	0.001				
^{a+h} Means with in a column with different superscripts differ at p < 0.05, C matter digestibility, IVOMD= <i>In vitro</i> organic matter digestibility, SEM= sta	RT= crop residue typ ndard error the of me	e, FP= fermentation peri-	od, IVDMD= <i>In vitr</i> o dry				





The maize stover had a metabolizable energy of 10.0 MJ/kg at day zero. The ME initially day zero (10.06 MJ/kg), then increased progressively, reaching a peak of 10.64 MJ/kg at 12th days of fermentation period. Overall, maize stover showed the highest ME values across all fermentation periods compared to Teff straw and sorghum stover. The value of ME at day zero was 9.16 MJ/kg, but then gradually decreased with extended fermentation, reaching 9.65 MJ/kg at day 12.

The EME, IVDMD and IVOMD of brewery spent yeast treated CRT in the present study was higher at longer fermentation period than zero day (control). Comparatively slightly higher EME, IVDMD and IVOMD values in the present study reported for 12th days of fermentation period. In a similar investigation, Kamphayae et al. (2016) found that adding up to 30% more BSY to cassava pulp significantly increased the silage materials of IVDMD, IVOMD and EME within 4 weeks of the start of the ensiling period. This is crucial for assessing the energy and digestibility value of the treated CRT, which is a key factor in animal performance. Higher digestibility ensures that the animals can extract more nutrients from the feed, improving their growth, milk production, and overall health. Enhanced digestibility and energy content of feed can lead to better-feed conversion ratios, reduced feeding costs, and improved sustainability in livestock production systems. Abebaye et al. (2020), also reported that the IVOMD (*in vitro* organic matter digestibility) and ME (MJ/kg) content was higher in green maize stover silage, which was treated with molasses and fermented for six to eight weeks of the than the control group. Other study by Kitaw et al. (2018), denoted the reduction in DM digestibility and significant losses in nutritional components may be attributed because of the aerobic deterioration of treated CRT material stored at high temperatures for extended period.

CONCLUSION

Results indicated that the fermentation time significantly affected the chemical composition of the treated crop residues. The study found that as fermentation time increased, the contents of dry matter, organic matter, and crude protein rose. Conversely, the levels of neutral detergent fiber, acid detergent fiber, and acid detergent lignin decreased over time, suggesting improved digestibility. Digestibility rates for dry matter and organic matter were notably higher compared to untreated residues. The findings highlight that the use of brewery yeast can enhance the nutritional quality of crop residues, making them more suitable for livestock feed. Therefore, based on the finding of this research the following recommendations are given: A) Broad awareness should be created among smallholder farmers about the significance role of brewery spent yeast treated crop residues; B) In terms of nutritional value, organoleptic quality, and digestibility values and wisely utilization of the bulk available crop residue, has paramount significant contribution to the feed cost reduction.

DECLARATION

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Data availability

The data that support the study findings are available from the corresponding author upon request.

Author contribution

M. Getachew: Formal analysis, data curation, investigation, software, supervision, validation, visualization, writing original draft, writing reviewing and editing, methodology; T. Amare: Conceptualization, funding acquisition, project administration, supervision, validation, methodology, writing original draft; E. Yimer: Conceptualization, funding acquisition, funding acquisition, project administration, supervision, methodology, writing original draft

Consent to publish

All participants have consented to the submission of the research article to the journal.

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The experiment was approved by the Ethics Committee of the Holeta Agricultural Research Institute.

Competing interests

There are no disclosed conflicts of interest for the writers. There are no financial interests to disclose, and all coauthors have reviewed and approved the manuscript's contents.

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THE EFFECT OF NITROGEN AND SULFUR IN MAKING ONCOM OF CASSAVA PULP BY *Neurospora sitophila* AND ITS IMPACT ON *IN VITRO* DIGESTIBILITY AND FERMENTABILITY IN SHEEP DIET

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Supporting Information

ABSTRACT: This study focused on the modification of oncom making based on cassava (*Manihot esculenta*) pulp to improve the nutritional profile through fermentation by *Neurospora sitophila* and enrichment with a mixture of urea (0, 2.5, 5, 7.5, and 10% of dry matter) and a nitrogen:sulfur ratio of 15:1 (oncom cassava pulp= OCP). Hence an investigation was carried out on the *in vitro* digestibility and fermentability of OCP when included in sheep diets. *In vitro* evaluation (diet oncom cassava pulp; DOCP) was carried out on diets consisted of a mixture of 50% grass and 50% concentrate containing 2.5%OCP (which selected based on the highest analysed crude protein and crude fiber levels when compared to the nutrient contents of other enriched OCP levels, P < 0.001) with compositions of 0, 10, 20, and 30% of DOCP. Results showed that the higher use of urea and nitrogen:sulfur ratio caused a decrease in crude fiber and gross energy and an increase in nitrogen-free extract (NFE) levels (P < 0.001). Using 2.5%OCP in the diet resulted in significantly different N-ammonia (N-NH₃), volatile fatty acid (VFA), *in vitro* dry matter digestibility (IVDMD), and *in vitro* organic matter digestibility (IVOMD) (P < 0.05), except for total gas and ruminal pH values. The highest IVDMD and IVOMD were obtained in 30% DOCP with a close N-NH₃ and VFA and within the normal range. In summary, CP fermentation by *Neurospora sitophila* which is enriched with a mixture of 2.5% urea and nitrogen:sulfur at a 15:1 ratio can be used in sheep diet up to 30%.

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INTRODUCTION

Oncom is a traditional peanut meal-based ferment that has long been produced by the Indonesian people, especially in the West Java region (Kenyamu et al., 2014). Through the fermentation process, oncom acquires a distinctive aroma and taste, because the chemical structure of complex ingredients breaks down into compounds that are simpler and easier to digest (Firoh et al., 2024). Two types of oncom are known to Indonesian society, namely red oncom and black oncom (Mulyani and Wisma, 2016). Red oncom is fermented by *Neurospora sitophila*, a microscopic filamentous fungus of the Ascomycota division, which spreads rapidly in the environment through vegetative spores of orange colour formed on the lateral branches of conidiogenous hyphae (Firoh et al., 2024). Meanwhile, black oncom is made through fermentation by the mold *Rhizophus oligosporus*, with the resulting spores being black (Mulyani and Wisma, 2016). The impact of fermentation on feed processing, apart from improving the quality of oncom products as well as the development of mold, is expected to also have a positive impact on ruminants if they consume it. This is because mold can act as a probiotic (Yu et al., 2020), where when consumed early in the rumen, the mold is still alive and will consume oxygen, thereby making the rumen environment more anaerobic. This condition will enable rumen bacteria to grow better because they live in an aerobic atmosphere.

Cassava pulp (CP; *Manihot esculenta*) is a by-product of the tapioca flour manufacturing industry which comes from cassava (Norrapokea et al., 2022). However, tapioca waste, like agro-industrial waste in general, has a limiting factor in its use, namely its low protein content. This weakness can be overcome by the fermentation process. Fermentation processing is not only useful for increasing the fermentability and digestibility of feed in digestive tract, but it can also increase nutritional value and improve palatability (Roger et al., 2015). Mold as a fermentation agent requires additional supplements in the form of nitrogen and sulfur sources for growth to work optimally during fermentation (Kampen, 2014). Nitrogen and sulfur are macronutrients essential for mold growth (Perner et al., 2011). Beyond this, the addition is expected to enrich the quality of the substrate.

Previous research on the use of *Neurospora sitophila* supplemented with urea and minerals has not been widely reported. Research on *Neurospora sitophila* has been carried out regarding the activity of cellulase, protease, amylase, gluco amylase, and phytase (Li et al., 2013; Liu et al., 2016; Syed et al., 2016; Kanti and Sudiana, 2016; Kanti, 2017). In

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another study, Neurospora *sitophila* was used to ferment tapioca for laying hen feed (Nuraini et al., 2015) and banana stems as a solid substrate for commercial cellulase production (Asad et al., 2006).

The benefit of this research is as a technology to improve the quality of CP as ruminant feed. This technology can increase the nutrient content of CP as a feed ingredient for ruminants and can replace other feed ingredients, which compete for use with other types of livestock (poultry).

MATERIALS AND METHODS

Making oncom from cassava pulp

Oncom is a typical Javanese fermented food product that uses peanut cake or tofu dregs as a substrate which is inoculated with red oncom mold spores (*Neurospora sitophila*) (Kenyamu et al., 2014). The research was carried out at the home industry oncom in Pasireungit Village, Legok District, Sumedang Regency, and at the Ruminant Animal Nutrition and Feed Chemistry Laboratory, Faculty of Animal Husbandry, Universitas Padjadjaran. Cassava pulp (CP) was obtained from a tapioca flour factory in Sumedang Regency, Indonesia. Urea was used as a nitrogen source while it contains is 46%. The source of sulfur was derived from the compound MgSO₄.7H₂O purchased from a chemical shop in Bandung City. This compound is easily soluble in water (Ramalingom et al., 2001). At the same time, the *Neurospora sitophila* culture was obtained from a producer in Sumedang Regency who usually supplies the mold inoculum needs.

Urea was weighed at 0%OCP, 2.5%OCP, 5%OCP 7.5%OCP, and 10%OCP of dry matter (DM) of the cassava pulp. MgSO₄.7H₂O was weighed based on a ratio of nitrogen (urea) and sulfur at a ratio of 15:1 and dissolved with water to 200 mL. This solution was then placed in a plastic bottle for 1 treatment requirement. The cassava pulp was steamed until hot (15-20 minutes), and then mixed with a solution of urea and MgSO₄.7H₂O until homogenous, according to the treatment. Then it was molded as a cube with dimensions of $11 \times 12 \times 2$ cm³ and weighed. It is stored on the fermentation rack. After it cooled, the *Neurospora sitophila* culture was added to the substrate surface and left for 3 days.

After 3 days, the cube molds were dried in the sun and continued with oven drying at 60°C. The nutrient content and gross energy were analyzed using the proximate analysis method (AOAC, 2010) and a bomb calorimeter. The best results at this stage are followed by *in vitro* tests. One aspect that needed to be considered in this research was the influence of the fermentation room, which was very dominant. The fermentation was successful because it was carried out in a place where OCP production fermentation is usually carried out (home industry). If conducted elsewhere, different results might be obtained. This is because the room is saturated with *Neurospora sitophila*.

In Vitro batch culture preparation

The experimental diets were then measured for fermentability and digestibility using in vitro techniques as described by Gosselink et al. (2004) and Despal et al. (2023). The diet consisted of a mixture of 50% grass and 50% concentrate containing 2.5% OCP with a composition of 0, 10, 20, and 30% DOCP. Based on Table 1, the 2.5% treatment produced significantly (P < 0.001) the highest crude protein and is most efficient in the use of urea levels and nitrogen:sulfur ratio. Concentrates were made from other feed ingredients which were arranged in such a way so that they were isonitrogenous and isocaloric. The complete feed samples were ground through a sieve with a size of 1 mm. The media solution was used as a medium for microbial development in carrying out fermentation activities and digesting feed. The media solution aimed to imitate the rumen fluid of ruminant livestock following the original conditions. The medium consisted of 3 local sheep rumen fluid and buffer solution or artificial saliva in a ratio of 4:1 mL and was maintained so that the pH value was around 6.5-7.0 (Hernaman et al., 2022). The media solution was mixed with the experimental feed sample (0.5 g) in a fermenter tube. During the filling process, CO₂ gas was supplied to maintain an anaerobic atmosphere. Previously, the fermenter tube had been placed in a water bath at a temperature of 39-40°C. Then part of it was incubated for 3 hours to take samples and measure the concentration of volatile fatty acids (VFA) using the Markam steam distillation method (AOAC, 2010) and N-NH₃ using the microdiffusion technique of Conway (Gosselink et al., 2004) and pH value analysis. Some samples were left for 2 × 48 hours to measure dry matter digestibility (DMD) and organic matter digestibility (OMD) as described by Despal et al. (2023). It was crucial to ensure that the rumen fluid used was teeming with live microbes and free from feed particles from the livestock before slaughter to obtain accurate results.

In vitro rumen fermentation and digestibility

The top of the Conway unit was smeared with Vaseline. A 1 mL of boric acid with the indicator was placed in the middle position of the Conway, 1 mL of rumen fluid or supernatant was fitted in the right position of the cup, and 1 mL of saturated Na₂CO₃ was fitted in the left position of the Conway. It was ensured that the components did not mix before the Conway unit was sealed. The Conway unit was then closed tightly so that no air could enter. The unit was gently moved in a figure-eight shape so that the rumen fluid and saturated Na₂CO₃ were mixed evenly and slowly, and it was then left for 24 hours at room temperature. After 24 hours, the mixture was titrated with 0.01 N H₂SO₄ until the color changed from blue to pink. The calculation formula for N-NH₃ concentration is as follows (Gosselink et al., 2004):

$N-NH_3 = (b \times N H_2SO_4 \times 1000/L) mM$

b = volume of H₂SO₄ used (mL); N= normality of H₂SO₄ solution

The steam distillation apparatus was turned on, and the sample holder was rinsed. Then, 5 mL of the supernatant was drawn up using a micropipette, placed in the sample holder, and 1 mL of 15% H₂SO₄ was added. The distillate was collected in a 250 mL Erlenmeyer flask filled with 5 mL of 0.5 N NaOH until the distillate volume reached 100 mL, followed by adding 2 drops of the phenolphthalein indicator. The distillate was then titrated with 0.5 N HCl until a color change was observed. A blank solution was prepared by titrating 5 ml of 0.5 N NaOH with 0.5 N HCl until a color change occurred. Measurement of total VFA concentration is calculated as follows (Gosselink et al., 2004):

Total VFA = $(b-s) \times N HCI \times (1000/5) mM$

b = volume of HCl used (mL); s = volume of sample titrant (mL); N= normality of HCl solution

The *in vitro* digestibility test consisted of two stages. The first stage simulated the fermentative digestion process in the rumen. At the end of the first stage, the tube lid was opened, proceeding to the second stage. In the second stage, after 48 hours of fermentation, microbial fermentation was halted. A 0.25 mL drop of HgCl₂ was introduced into the fermenter tube to stop microbial activity. Subsequently, 2 mL of 4 N HCl and 0.06 g pepsin were added to each tube, and then it was placed back in the incubator at 39°C for 48 hours without a rubber lid. The digested material was filtered using a filter, transferred to a crucible glass, and then placed in an oven at 105°C for 24 hours to determine the dry matter (DM) residues. After the second stage, the tube contents were filtered with sintered glass, and the residue was further processed to determine digestibility variables. The second stage replicated the enzymatic hydrolysis procedure digestion in the post-rumen.

Gas production was measured using a gas measuring cylinder which captured the gas produced (Blümmel et al., 1997). Observations of gas production occurred at intervals of 2, 4, 8, 12, 24, 48, and 72 hours of fermentation. Rumen fluid pH was measured using a pH meter, and the reading was recorded as the pH value.

Data analysis

A completely randomized design was employed for the research. The data collected were analyzed using the Duncan test using the SPSS IBM 21 data processing application.

RESULTS AND DISCUSSION

Nutrient of oncom from cassava pulp

After 3 days of fermentation process carried out by *Neurospora sitophila* on piles enriched with urea and sulfur, data on nutrient and energy content were produced which are presented in Table 1. Table 1 illustrates that fermentation by *Neurospora sitophila* and enrichment with a mixture of urea (0, 2.5, 5, 7.5, and 10% of dry matter) and nitrogen:sulfur at a ratio of 15:1 resulted in significant changes (P < 0.001) in the content of crude protein, crude fiber, NFE, and gross energy. In contrast, the crude fat and ash content did not show a significant difference.

Crude protein increased whereas crude fiber and gross energy decreased after CP was fermented by *Neurospora sitophila* enriched with urea and sulfur. NFE also experienced a decrease but then increased again in line with the decreasing amount of crude protein and crude fiber. The highest crude protein and gross energy were obtained in the 2.5% OCP treatment, which is fermentation enriched with 2.5% urea and sulfur at a nitrogen and sulfur ratio of 15:1.

Table 1 - Nutrient and energy content of OCP*							
Parameter	0%OCP	2.5%0CP	5%0CP	7.5%0CP	10%0CP	SEM	P value
Ash (%)	1.63	1.39	1.52	1.28	1.28	0.822	0.631
Crude protein (%)	2.91°	7.65ª	7.00 ^b	6.70 ^b	6.49 ^b	0.349	0.001
Crude fat (%)	1.51	1.91	2.02	2.17	2.12	0.438	0.137
Crude fiber (%)	14.90 ª	14.42 ª	13.84 ^{ab}	12.61 ^b	9.77°	0.090	0.001
Nitrogen-free extract (NFE) (%)	79.05 ^{ab}	74.62 ^d	75.62 ^{cd}	77.25 ^{bc}	80.35ª	0.505	0.001
Gross energy (kkal/kg)	3266.6ª	3162.2 ^b	3012.2 ℃	3014.2 ℃	3009.6°	22.65	0.001
Values are expressed as mean (n=5), different letters within rows represented significant differences (p <0.05). *) Fermentation of cassava pulp (CP) by <i>Neurospora sitophila</i> enriched with urea (0, 2.5, 5, 7.5, 10%) and a 15:1 ratio of nitrogen:sulfur.							

The increase in crude protein in the treatment with the addition of urea and sulfur occurs because these components are utilized to form protein in cell development and the growth of *Neurospora sitophila*. Microbes in the fermentation process can produce enzymes that degrade complex compounds into simpler ones and synthesize protein, which is a

protein enrichment process (Cruz-Casas et al., 2021). Based on this, it can be inferred that some non-protein nitrogen (NPN) compounds in the form of urea undergo conversion into pure protein compounds in the form of microbial protein (Norrapokea et al., 2022). The function of sulfur minerals in the physiology of microbial cells includes being a part of the protein amino acids cysteine and methionine and being part of several enzymes (CoA, Co-enzyme A carboxylase; Kabil et al., 2014), which are essential amino acids, while the enzymes formed play a major role in metabolic processes in cells. However, the treatment using more than 2.5% urea, produces lower crude protein; this is believed to occur because the fermentation process, being aerobically conducted, allows the dissolved urea to evaporate into ammonia (NH₃), characterized by the distinctive smell of ammonia, making urea less effective if used in amounts greater than 2.5%.

Crude fat does not change significantly. This is because crude fat is not an organic material needed by *Neurospora sitophila* in the fermentation process, and the enzymes commonly produced by *Neurospora sitophila* are those that digest fiber, sugar and protein. The absolute amount of this compound remains unchanged, only because other nutrient components decrease, it appears as if there is an increase. Meanwhile, the decrease in crude fiber, NFE, and gross energy content indicates that the application of urea and sulfur has provided nutrients for the growth of *Neurospora sitophila*, ensuring that fermentation runs well as the mold requires energy obtained from crude fiber and NFE. The ash component was not significantly different, because ash is an inorganic material with a small amount and is not an organic material component that undergoes breakdown during the fermentation process, therefore, its amount remains relatively constant.

The decrease in crude fiber content alongside the addition of urea and sulfur suggests that *Neurospora sitophila* utilizes crude fiber predominantly as an energy source. Research conducted by Qingxin et al. (2014) which analyzed enzymes produced by *Neurospora sitophila*, found that the enzymes peptidase (protease), endoglucanase, exoglucanase, β -glucosidase, and cellobiose dehydrogenase play roles in the breakdown of cellulose and hemicellulose. Fermentation using *Neurospora sp*. which has cellulolytic properties can break down cellulose bonds, causing the crude fiber content in the substrate to decrease. Fermentation using *Neurospora sp*. also leads to the degradation of cellulose, hemicellulose, and polymers into simpler sugars or their derivatives, increasing the nutrient content of the substrate material (Znameroski et al., 2012). Endo- β -1,4-glucanase cuts chain bonds in cellulose to produce shorter cellulose molecules. Exo-1,4-glucanase cuts the end of the cellulose chain to produce a cellobiose molecule, while β -glucosidase further breaks down the cellobiose molecule into two glucose molecules (Romero et al., 1999).

The NFE value decreased then increased gradually in the next treatment. This condition does not indicate an increase in NFE value. The NFE is also fermented as an energy source, as evidenced by the decreasing gross energy in treatment along with increasing NFE value. The fermentation process not only utilized crude fiber as an energy source but also other organic components, including NFE (Hernaman et al., 2017). The NFE value is obtained from calculation and depends on other nutrient components. So, changes in NFE values are caused by changes in the values of other nutrient components.

In vitro fermentability and digestibility of diet containing OCP

The results at stage 1 showed that 2.5% OCP treatment produced the highest protein, so it was used in diets for *in vitro* testing with the composition of diets and nutrient content presented in Table 2. Table 2 shows that the use of 2.5% OCP can replace the role of corn and rice bran, the higher the use of 2.5% OCP, the lower the utilization of these two conventional feed ingredients.

Table 2 - Composition of experimental diets (DOCP)*				
Ingredients	0%DOCP	10%DOCP	20%DOCP	30%DOCP
Napier Grass cv Taiwan (%)	50.00	50.00	50.00	50.00
Oncom of Cassava Pulp (2.5% OCP) (%)	0.00	10.00	20.00	30.00
Corn flour (%)	38.49	27.64	16.79	5.92
Soybean meal (%)	0.50	0.50	0.50	0.50
Pollard (%)	2.26	5.69	9.13	12.58
Rice bran (%)	7.76	5.17	2.58	0.00
Mineral mix (%)	1.00	1.00	1.00	1.00
Nutrient				
Dry matter (%)	77.24	77.16	77.08	77.00
Ash (%)	17.29	17.21	17.14	17.06
Crude protein/CPr (%)	12.00	12.00	12.00	12.00
Crude fat/CFt (%)	4.62	4.21	3.79	3.39
Crude fiber/CFr (%)	14.44	15.58	16.71	17.01
NFE (%)	51.65	51	50.36	50.54
TDN (%)	63.50	63.50	63.50	63.50
TDN was calculated using Sutardi equation as described in Hernan	nan et al. (2022	2): (70.6+0.259%CPr+2	L.01%CFt-0.76%CF	r+ 0.0991% NFE).

TDN was calculated using Sutardi equation as described in Hernaman et al. (2022): (70.6+0.259%CPr+1.01%CFt-0.76%CFr+ 0.0991% NFE). *) The diet consisted of a mixture of 50% grass and 50% concentrate containing 2.5%OCP with a composition of 0% in DOCP, 10% in DOCP, 20% in DOCP, and 30% in DOCP

Table 3 - In vitro fermen	tability and digestibility (of experimental diets (DOC	P)
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Parameters	0%DOCP	10%DOCP	20%DOCP	30%DOCP	SEM	P-value
N-ammonia (N-NH ₃) (mM)	4.16 ^b	4.31 ^{ab}	5.11 ª	3.69 ^b	0.17	0.015
Volatile fatty Acid (VFA) (mM)	148.57 ^{ab}	159.63ª	137.28 ^b	135.85 ^b	3.46	0.036
Total gas (mM)	132.06	126.74	136.08	128.30	3.17	0.760
Ruminal pH	6.92	6.96	6.92	6.89	0.02	0.716
In vitro dry matter digestibility (IVDMD) (%)	66.4 ^b	57.29°	67. 1 3 ^b	69.49ª	1.08	0.0001
In vitro organic matter digestibility (IVOMD) (%)	73.62 ^b	65.08°	75.36 ^{ab}	76.83ª	1.11	0.0001

Values are expressed as mean (n=5), different letters within rows represent significant differences (P < 0.05). *) The diet consisted of a mixture of 50% grass and 50% concentrate containing 2.5% OCP with a composition of 0% in DOCP, 10% in DOCP, 20% in DOCP, and 30% in DOCP.

Consequently, this suggests that 2.5% OCP could potentially substitute for corn and rice bran, which are commonly utilized in the poultry industry (Rohaeni et al., 2021). Subsequently, the experimental diet was evaluated *in vitro*, and the results are depicted in Table 3. In Table 3, the N-NH₃ content in the 20% DOCP treatment was significantly higher (P<0.05) compared to the 30% DOCP and 10% DOCP treatments yet shared the same value as the 10% DOCP treatment. Meanwhile, the highest VFA levels were found in the 10% DOCP treatment, which had a similar average value as the 0% DOCP treatment and greater than the 20% and 30% DOCP treatments, whereas between these treatments there was no significant difference. The *in vitro* dry matter digestibility (IVDMD) and organic matter digestibility (IVOMD) in the 30% DOCP treatment was superior (P < 0.05) compared to other treatments. Furthermore, pH and total gas measurements remained consistent across all treatments.

Overall, the N-NH₃ and VFA concentrations are 3.69-5.11 mM and 135.85-159.63 (Table 3). The N-NH₃ is generally lower than 5 mM that the minimum required for the microbial protein synthesis in the rumen (Dewhurst and Newbold 2022). Meanwhile, the VFA concentrations is still within the normal range for rumen fluid, as established by McDonald et al. (2002), at 70 -150 mM. The percentages for IVDMD and IVOMD were relatively high, ranging between 57.29-69.49% and 65.08-76.83%, respectively. These figures align closely with the findings of Tresia et al. (2024) which utilized rations composed of local feed ingredients, reporting values of 51.18-56.38% and 49.12-54.45%, respectively. The pH levels for all treatments spanned from 6.89 to 6.96 (Table 3), remaining within the physiological rumen range of 5.5-7.5 (Franzolin et al., 2010). The McDougall's artificial saliva, added as a buffer, effectively maintained pH levels within normal limits, even though significant variations were noted in fermentation products such as N-NH₃ and VFA (P < 0.05).

Total gas production, a by-product of the fermentation process in the rumen, remained consistent across all treatments, signifying that 2.5% OCP inclusion in the diet did not negatively impact the rumen microbial activity. Total gas production indicates a feed fermentation process by microbes in the rumen (Suassuna et al., 2022). The stable volume of total gas indicates effective feed fermentation by rumen microbes and suggests a uniform degree of feed degradation and organic material digestion.

These analytical results demonstrate that incorporating up to 30% of 2.5% OCP maintains normal fermentability and optimizes digestibility. Thus, 2.5% OCP proves to be a viable ingredient in rations, capable of substituting for other feed components. It is evidenced in Table 2 that 2.5% OCP can replace significant amounts of corn and rice bran, with increasing levels of 2.5% OCP, there is a corresponding decrease in the use of corn and rice bran. This is noteworthy because corn and rice bran are predominantly allocated to poultry feed formulations, with corn constituting more than 60% (Islam et al., 2015) and rice bran accounting for 20-30% (Rohaeni et al., 2021).

Fermented feed, having undergone a process of chemical structural transformation through the action of microbial enzymes, offers multiple benefits. Notably, it enhances the fermentability and digestibility of feed in the rumen, improves nutritional value, and increases palatability (Roger et al., 2015).

CONCLUSION AND RECOMMENDATION

Production of oncom resulting from cassava pulp (CP) fermentation by *Neurospora sitophila* enriched with 2.5% urea and sulfur at a nitrogen-to-sulfur ratio of 15:1 has been shown to produce optimal changes in nutrient composition and gross energy values. When included in sheep diets at a ratio of up to 30%, it demonstrates significant potential for nutritional enhancement. It highlights the significance of microbial fermentation, facilitated by *Neurospora sitophila*, in enhancing the nutritional profile of plant-based substrates. Such advancements not only propose an avenue for improving the efficiency of animal nutrition but also open up possibilities for addressing feed scarcity and promoting environmental sustainability. The implication of this research extends to the concept of circular economy in agriculture, where waste products are upcycled into valuable feed, contributing to waste reduction and resource optimization.

DECLARATIONS

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Data availability

The datasets used and/or analysed during the current study available from the corresponding author on reasonable request.

Authors' contribution

I.Hernaman is a leader in research who, conducts research, interprets data, and writes manuscripts. T.Dhalika, U.H.Tanuwiria, R.Hidayat are the co-authors who assisted in conducts research, interpreting data, and writing manuscripts. U.Rosani, A.Budiman, and M.Rifqi are the co-authors who assisted in preparing research, interpreting data, and writing manuscripts. B.K.Mutaqin and M.R.Nugraha are the co-authors who assisted in conducting research and collecting data. Budi Ayuningsih is the co-author who assisted in conducting research and financial administration.

Ethical approval

This research does not use live animals (sheep) and the rumen fluid used comes from slaughterhouses that meet animal welfare standards of the Indonesian National Standards for Halal Slaughter of Ruminant Animals No 99003:2018

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Competing interests

The authors declare that there are no competing interests.

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ESTIMATION OF BODY WEIGHT FROM BIOMETRIC TRAITS OF CHICKENS USING REGRESSION STATISTICAL METHOD: Δ SYSTEMATIC REVIEW

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Supporting Information

ABSTRACT: Live body weight is a vital tool when placing price on the chickens for profit and to assess decisions to be made in selection of animals and other husbandry practices. However, the chicken farmers, especially the resource limited farmers, lack weighing scales to perform these animal husbandry practices. The objective of this study was to systematically review the articles published on the estimation of live body weight from biometric traits of chickens using regression statistical methods. Databases such as Google Scholar, ScienceDirect, PubMed, and Web of Science, with the combination of the following keywords: "Body weight" or "body mass", "biometric traits" "zoometric measurements" or "Morphological traits" or "linear body measurements" or "Morphometric traits" or "body parameters" or "growth traits" or "growth performance traits", chicken or poultry or "Gallus gallus domesticus" or fowl, regression. There were limited articles that aimed to predict live body weight using regression statistical method across the world. A total of fourteen articles were published between the years 2009 and 2024. The results indicated that stepwise linear regression method was mostly used by 39% of the articles included, followed by multiple regression method and simple linear regression method by 22% of the included articles each. The results indicated that the highest coefficient of determination (R2 = 0.970) was recorded on the model of combination of body length (BL) and breast length (BRL) using multiple linear regression method on chickens. The limitation is that some articles did not include either sex or age of the animals that can make it difficult to make conclusion for different sexes and ages. This systematic review concludes that the multiple linear regression statistical method is the best in estimating live body weight in chickens with combination of BL and BRL. As a practical suggestion, it would be best for breeders to select chickens with the highest measurements of BL and BRL to improve the live body weight of the chickens.



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Keywords: Biometric traits, Body weight, Chicken, Linear body measurements, Regression,

INTRODUCTION

Live body weight is an important tool to use when pricing chickens for sale and assessing the choices to be made in animals husbandry practices (Semakula et al., 2011; Adenaike et al., 2015). Body measurements' components such as live body weight and biometric traits of an animal can be used to evaluate growth of that animal (Yakubu and Salako, 2009; Sadick et al., 2020). Farmers use body weight to price their reared chicken when selling them for profit and to generate income (Udeh et al., 2021).

However, one of the challenges that most farmers experience is that they lack access to weighing scales which make it difficult to estimate their chickens live body weight and growth rate (Semakula et al., 2011; Tyasi et al., 2024). The live body weight of animals such as chickens can be predicted from biometric traits using a variety of regression methods (Yunusa and Adeoti, 2014; Bila et al., 2021).

Several studies have been conducted on the estimation of live body weight from biometric traits of chickens (Adenaike et al., 2015; Yakubu and Ari, 2018; Bila and Tyasi, 2022). Although, according to the authors' knowledge, there has been no systematic review on the estimation of live body weight from biometric traits of chicken using regression statistical methods. Thus, the objective of this study was to systematically review the articles published on the estimation of live body weight from biometric traits of chickens using regression methods. The results from this study will help researchers to find the best biometric traits that they will guide farmers to use when estimating their chickens' live body weight to easily fulfil their farm husbandry practices.

MATERIALS AND METHODS

Eligibility criteria

Identification of Population, Exposure, and Outcomes (PEO) elements of the research questions were done for this systematic review. The "chickens" were identified as the population of this study, with "regression statistical methods" as

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exposure and "regression models for estimation of live body weight from biometric traits of chickens" as the outcomes. Before it was decided to pursue with this study, the first search of the PEO components on Google Scholar, PubMed, ScienceDirect, and Web of Science were performed.

Search strategy

The two investigators conducted a systematic review of articles in the databases such as Google Scholar, ScienceDirect, PubMed, and Web of Science, with the combination of the following keywords: "Body weight" or "body mass", "biometric traits" or "Morphological traits" or "linear body measurements" or "Morphometric traits" or "body parameters" or "morphobiometric traits" or "zoometric measurements" or "growth traits" or "growth performance traits", chicken or poultry or fowl or "Gallus gallus domesticus", regression. Only English articles were considered in this review. The keywords were combined in different combinations.

Inclusion criteria

The articles that were available from more than one database were removed prior screening for eligibility. The criteria for inclusion were articles that estimated the live body weight of chicken using regression method, articles written in English, and articles that are published using regression methods like simple linear, stepwise linear, multiple linear regression, power non-linear regression, exponential non-linear regression and polynomial non-linear regression methods were included. Articles that deal with the estimation of live body weight of chicken using biometric traits, regression models for estimation of live body weight of chicken from biometric traits were included in this review.

Exclusion criteria

Articles were removed from the included articles due to that they did not meet requirements such as duplicate records, studied different species, used different methods of estimating live body weight such as machine learning algorithms, and not written in English.

Data extraction

The data of this study was independently extracted by two investigators and a general agreement concerning all the materials was reached. The articles that have met inclusion criteria had an author, publication year, type of breed, and the model type.

Ethical considerations

Misconduct, Plagiarism, informed consent and manipulation of data were termed ethical criteria by all authors when conducting this systematic review.

RESULTS

Searched Literature

Figure 1 shows the flow chart of the identification and selection of articles for systematic review. In the initial search, a total of 452 articles were extracted. A number of 23 duplicate articles were excluded, and 429 articles remained. Articles were screened for title; 395 articles were excluded. A number of 34 articles were screened, and 18 articles were removed. About 16 articles were screened for eligibility and 2 articles were removed. A total number of 14 articles were included in this review.

Characterization of included articles

Table 1 shows the characterization of 14 articles that are included in this review. The results showed that Adenaike et al. (2015) and Akporhuarho and Omoikhoje (2017) both predicted body weight of chicken breeds' crosses. The findings showed that out of 14 articles included in this review, Yunusa and Adeoti (2014) used the highest sample size (n = 2641) of chickens. The results indicated that from the 14 articles included in this review, 2 articles (Yakubu and Salako, 2009; Yakubu et al., 2009) used the same sample size (n = 238) of chickens and 2 articles (Bila and Tyasi, 2022; Tyasi et al., 2024) used the same sample size (n = 100) of chickens. The results showed that most of the breeds that were used across the 14 articles included in this review were indigenous chicken breeds (n = 16). The results indicated that the most used production system across the 14 articles was intensive production system (n = 9).

Publication by year

Figure 2 shows the year of publications of the included 14 articles. The results indicated that out of the 14 articles included, only 10 different years that the articles were published. The results indicated that year 2021 (Ikeh and Okwesili, 2021; Bila et al 2021; Udeh et al., 2021) had the highest published articles (n = 3) and 2009 (Yakubu and Salako, 2009; Yakubu et al., 2009) and 2024 (Christophe et al., 2024; Tyasi et al., 2024) had the second highest published articles (n = 2) out of the 14 included articles. The results showed that year 2011 (Semakula et al., 2011), 2014 (Yunusa and Adeoti, 2014), 2015 (Adenaike et al., 2015), 2017 (Akporhuarho and Omoikhoje, 2017), 2018 (Yakubu and Ari, 2018), 2020 (Sadick et al., 2020) and 2022 (Bila and Tyasi, 2022) had the least number of articles published (n = 1).

Table 1 - Characterisation of included articles										
Author	Year	Country	Age (weeks)	Breed	Sample Size	Production system	Biometric traits	Regression method		
Yakubu and Salako	2009	Nigeria	-	Nigerian indigenous Autochthonous chicken	238	Extensive	CH, CL, BKL, BL, NL, SL, CC, TC	Multiple linear		
Yakubu et al.	2009	Nigeria	-	Nigerian Normal feathered, Naked neck, Frizzled chickens	238	Extensive	CH, CL, BKL, BL, NL, SL, CC, TC	Stepwise linear		
Yunusa and Adeoti	2014	Nigeria	-	Nigerian Yoruba chickens, Nigerian Fulani chickens	2641	Semi- intensive	KL, TL, WL, BL, DS, SL, CC, BRL	Multiple linear		
Akporhuarho and Omoikhoje	2017	Nigeria	22	Exotic Broilers * Nigerian local chickens (main), Nigerian local * exotic broiler chickens (reciprocal)	228	Intensive	BL, WS, WL, TL, SL, KL, BRG	Stepwise linear		
Adenaike et al.	2015	Nigeria	8	Normal-feathered, Marshal, Naked-neck, Marshal*naked-neck, Marshal*normal feathered	265	Intensive	BL, BRG, WL, WS, TL, SL, KL	Stepwise linear		
Bila and Tyasi	2022	South Africa	5	Ross 308 broiler chicken	100	Intensive	WL, BKL, SL, BG, BL, SC	Stepwise linear		
Yakubu and Ari	2018	Nigeria	6	Sasso, Kuroiler, Fulani	150	Intensive	BRG, NC, BL, WL, TL, TC, SL, SC	Stepwise linear		
lkeh and Okwesili	2021	Nigeria	7, 8	Nigerian Nsuka heavy chicken	120	Intensive	BL, CG, TC, TL, SL	Stepwise linear		
Udeh et al.	2021	Nigeria	8, 14, 20	Nigeria Nsuka heavy chicken	240	Intensive	BL, SL, CC, TC, TL	Simple linear, Multiple linear		
Tyasi et al.	2024	South Africa	40	White leghorn	100	Intensive	BL, WL, SL, SG, CG, BKL, BCL, CNH, TBL	Stepwise linear		
Semakula et al.	2011	Uganda	8, 24, 36, 80	Ugandan indigenous chicken	493	Semi- intensive	CPL, CC, FL, FC, SL, KL	Exponential non-linear, Polynomial non-linear, Power non-linear, Simple linear		
Bila et al.	2021	South Africa	5	Ross 308 broiler chicken	130	Intensive	WL, BKL, SL, BG, BL, SC	Multiple linear		
Sadick et al.	2020	Ghana	15	Cobb broiler chicken	50	Intensive	HL, BKL, BL, BL, SL, SC, CNH	Simple linear		
Christophe et al.	2024	Benin	-	Goliath chicken	342	Extensive	SL, SD, BL, DS, CC, WS, STL,	Simple linear		
_ = not indicated, CH length, WL = wing le length, TBL = toe-to-	I = comb height, ngth, BRL = brea back length, BC =	CL = comb len ast length, DS = body circumf	gth, BKL = beal = drum stick, W erence, CNH = c	k length, BL = body length, NL = neck length, CC = chest /S = wing span, BRG = breast girth, BG = body girth, SC hicken height, CPL = corpus length, FL = femur length, F	circumferen = shank circ C = femur ci	ce, TC = thigh circ umference, CG = rcumference, HL	cumference, SL = shank leng chest girth, NC = neck circu = head length, SD = shank d	gth, KL = keel length, TL = thigh mference, BCL = beak-to-comb iameter, STL = sternum length.		

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Figure 1 - Flow chart of identification and selection of studies used in the systematic review and Meta analysis



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Figure 4 - Publication by chicken breed.

R308BC = Ross 308 broiler chicken, WLC = White leghorn chicken, EB*LNC = Exotic broiler*local Nigerian chicken, LN*EBC = Local Nigerian*exotic broiler chicken, SC = Sasso chicken, NNHLC = Nigerian Nsuka heavy local chicken, NAC = Nigerian autochthonous chicken, FC = Fulani chicken, NNFC = Nigerian Normal feather chicken, NNNC = Nigerian Naked neck chicken, NFC = Nigerian Frizzled chicken, YC = Yoruba chicken, MC = Marshal chicken, M*NFC = Marshal*normal feather chicken, M*NC = Marshal*naked neck chicken, KC = Kuroiler chicken, UIC = Ugandan indigenous chicken, CBC = Cobb broiler chicken, GC = Goliath chickens.



Publication by country

Publications by country of the 14 articles included are presented in Figure 3. The results showed that across the 14 articles included only 5 countries that the articles were published. The results showed that out of the 14 articles, Nigeria had the highest published articles (n = 8), and South Africa (Bila and Tyasi, 2022; Bila et al., 2021; Tyasi et al., 2024) had the second highest published articles (n = 3).

Publication by chicken breed

Publications by breed of the 14 articles included are displayed in Figure 4. The results indicated that 19 breeds were used in this review. The results showed that the breed of Nigerian Naked neck chicken (Yakubu et al., 2009; Adenaike et al., 2015), Nigerian Normal feathered chicken (Yakubu et al., 2009; Adenaike et al., 2015), Ross 308 broiler chicken (Bila et al., 2021; Bila and Tyasi, 2022), Nigerian Nsuka heavy chicken (Ikeh and Okwesili, 2021; Udeh et al., 2021) and Nigerian Fulani chicken (Yunusa and Adeoti, 2014; Yakubu and Ari, 2018) were the mostly used chicken breeds (n = 2).

Publications by regression statistical methods

Figure 5 demonstrates the publications by regression methods. The results indicated that there were only 6 regression methods (simple linear, stepwise linear, multiple linear, power non-linear, exponential non-linear and polynomial non-linear) used in this review. The results showed that stepwise linear regression method was mostly used (n = 7) across the 14 articles included. The results showed that multiple linear regression (Yakubu and Salako, 2009; Yunusa and Adeoti, 2014; Bila et al., 2021; Udeh et al., 2021) and simple linear regression method (Semakula et al., 2011; Sadick et al., 2020; Udeh et al., 2021; Christophe et al., 2024) were second most used regression methods (n = 4) across the 14 included articles.

Estimation of live body weight using simple linear regression method

Table 2 shows the estimation of live body weight from biometric traits using simple linear regression. The results showed that only 4 articles (Semakula et al., 2011; Sadick et al., 2020; Udeh et al., 2021; Christophe et al., 2024) out of the 14 articles included predicted live body weight using simple linear regression method. The results indicated that the highest coefficient of determination ($R^2 = 0.760$) was reported by Semakula et al. (2011) on model of CC in Ugandan indigenous chickens and was followed by the model of BL with $R^2 = 0.750$ on Cobb broiler chickens. The least coefficient of determination ($R^2 = 0.074$) was reported by Chrysostome et al. (2024) on the model of WS on Goliath chickens.

Estimation of body weight using stepwise linear regression method

Estimation of live body weight from biometric traits of chicken using stepwise linear regression is shown in Table 3. The results mentioned that 7 articles out of the 14 articles included in this review were predicting live body weight of chickens using stepwise linear regression method. The results indicated that Yakubu et al. (2009) reported the highest coefficient of determination (R2 = 0.960) on the model of the combination of TC, CC, CH and BL on the Nigerian Frizzled chickens. The results indicated that the second highest coefficient of determination (R2 = 0.960) on the model of the combination of TC, CC, CH and BL on the Nigerian Frizzled chickens. The results indicated that the second highest coefficient of determination (R2 = 0.950) was recorded by Yakubu et al. (2009) on the model of combination of TC, CC, and CH on Nigerian Frizzled chickens, and by Akporhuarho and Omoikhoje (2017) on the model of combination of BL, WS, WL, and TL on reciprocal male LN*EBC crossbreed chickens. The results showed that the least coefficient of determination (R2 = 0.002) on the model of BL was reported by Akporhuarho and Omoikhoje (2017) on main female EB*LNC crossbreed chickens.

Estimation of body weight using multiple linear regression method

Table 4 indicates the estimation of live body weight from biometric traits of chickens using multiple linear regression method. The results showed that 4 articles out of the 14 articles included estimated body weight of chickens using multiple linear regression method. The results indicated that the highest coefficient of determination (R2 = 0.970) was recorded by Yunusa and Adeoti (2014) on the model of combination of BL and BRL using Nigerian Yoruba chickens. The results showed that Yakubu and Salako (2009) reported the second highest coefficient of determination (R2 = 0.952) on male Nigerian Autochthonus chickens from the combination of TC, CH and BL. The least coefficient of determination (R2 = 0.952) on male Nigerian Autochthonus chickens from the combination of TC, CH and BL. The least coefficient of determination (R2 = 0.952) on BRL, CC, and WL.

Estimation of body weight using power non-linear regression method

Estimation of live body weight from biometric traits of chickens using power non-linear regression method is shown on Table 5. The results indicated that only 1 article across the 14 articles included (Semakula et al., 2011), estimated live body weight using power non-linear regression. The results showed that model of CC had coefficient of determination of $R^2 = 0.830$ on Ugandan indigenous chickens.

Estimation of body weight using exponential non-linear regression method

Table 6 indicates the estimation of live body weight from biometric traits of chickens using exponential non-linear regression method. The results indicated that 1 article (Semakula et al., 2011) estimated live body weight using exponential non-linear regression across 14 articles included. The results showed that model of CC had coefficient of determination of $R^2 = 0.800$ on Ugandan indigenous chickens.

Estimation of body weight using polynomial non-linear regression method

Estimation of live body weight of chickens from biometric traits using polynomial non-linear regression method is shown on Table 7. The results indicated that only 1 article (Semakula et al., 2011) estimated live body weight using

polynomial non-linear regression across 14 articles included. The results showed that model of CC had coefficient of determination of $R^2 = 0.770$ on Ugandan indigenous chickens.

Table 2 - Estimation of live body weight using simple linear regression

Author and				G	oodness of	fit
year	Breed	Sex	Model		RMSE	P value
			LBW = -909.52 + 83.67BL	0.370	4.330	P < 0.01
lideb et el	Nigorian Nauka haaw laad		LBW = -1159.38 + 316.42SL	0.580	10.99	P < 0.01
(2021)	chicken	_	LBW = -904.80 + 59.68CC	0.720	1.490	P < 0.01
(2021)	CHICKEN		LBW = -317.43 + 143.38TC	0.460	6.260	P < 0.01
			LBW = -620.55 + 125.38TL	0.710	3.170	P < 0.01
			LBW = - 95.287 + 186.27SL	0.710	_	P < 0.01
			LBW = 478.55 + 386.88BKL	0.410	_	P < 0.01
Sodials at al	Cobb broiler chicken	-	LBW = -111.33 + 189.05HL	0.630	_	P < 0.01
Sadick et al.			LBW = -2.33 + 42.919BL	0.590	_	P < 0.01
(2020)			LBW = -119.29 + 59.08WL	0.660	_	P < 0.01
			LBW = -187.47 + 62.151CNH	0.720	_	P < 0.01
			LBW = -132.10 + 395.07SC	0.750	-	P < 0.01
Semakula et al. (2011)	Ugandan Indigenous chicken	-	LBW = -1.60 + 0.123CC	0.760	-	P < 0.01
Chrysostome et al. (2024)	Goliath chicken	-	LBW = 1.23 + 0.12SL	0.102	-	P < 0.01
			LBW = 0.41 + 1.52SD	0.327	_	P < 0.01
			LBW = 1.22 + 0.03WS	0.074	_	P < 0.01
			LBW = 0.829 + 0.10STL	0.223	_	P < 0.01
			LBW = 0.35 + 0.05BL	0.120	_	P < 0.01
			LBW = 1.27 + 0.09DS	0.119	_	P < 0.01

_= not indicated, P < 0.05 = significant, RSME: residual square mean of error, LBW: live body weight, BKL: beak length, BL: body length, CNH: chicken height, CC: chest circumference, TC: thigh circumference, SL: shank length, TL: thigh length, HL: head length, SC: sternum circumference, WL: wing length, SD = shank diameter, STL = sternum length.

 Table 3 - Estimation of body weight using stepwise linear regression.

Author and	Prood	For	Medel		Goodness of	fit		
year	Breed	Sex	Model	R ²	RMSE	P value		
	Nictorion normal		LBW = - 1.02 + 0.3BL	0.830	_	_		
	footborod		LBW = - 0.43 + 0.05BL + 0.06TC	0.870	_	_		
	chicken	-	LBW = - 0.26 + 0.04BL + 0.05TC + 0.04CH	0.880	-	_		
	Chicken		LBW = 0.24 + 0.03BL + 0.05TC +0.05CH + 0.01CC	0.890	-	_		
Yakubu et	Nigerian naked		LBW = - 0.77 + 0.08CC	0.830	_	-		
	neck chicken	_	LBW = - 0.47 + 0.06CC + 0.04TC	0.850	-	_		
al. (2009)	HECK CHICKEN		LBW = - 0.50+ 0.04CC + 0.03TC + 0.07SL	0.870	-	_		
			LBW = 0.28 + 0.13TC	0.910	_	_		
	Nigerian Frizzled		LBW = - 0.05 + 0.11TC + 0.02CC	0.940	-	_		
	chicken	-	LBW = 0.02+ 0.09TC + 0.02CC + 0.05CH	0.950	_	_		
			LBW = 0.24 + 0.08TC + 0.03CC + 0.08CH - 0.24BL	0.960	_	_		
	Nigerian heavy local ecotype chicken		LBW = - 454.93 + 221.60SL	0.605	_	P < 0.01		
		F	LBW = -1689.96 + 149.25SL + 75.83BL	0.702	_	P < 0.01		
Ikeh and			LBW = -1535.15 + 103.30SL + 63.73BL + 48.84TC	0.735	_	P < 0.01		
Okwesili (2021)			LBW = - 1515.55 + 85.12SL + 59.32BL + 40.00TC + 9.30CG	0.743	-	P < 0.01		
			LBW = - 1485.70 + 82.06SL + 58.34BL + 38.70TC + 8.39CG + 3.85TL	0.746	-	P < 0.01		
			LBW = 4040.9 - 59.5BL	0.036	-	_		
			LBW = 3848.2 - 60.7BL + 5.1WS	0.042	_	_		
Akporhuarho	Exotic broiler X		LBW = 7039.9 - 54.6BL + 10.1WS - 146.9WL	0.152	-	_		
and	local Nigerian	м	LBW = 4331.2 - 53.8BL + 9.9WS - 150.6WL + 151.4TL	0.156	-	_		
Omoikhoje (2017)	chicken (EB*LNC) main		LBW = 8170.7 - 42.4BL + 4.3WS - 161.1WL + 53.2TL - 196.8SL	0.223	-	-		
		((_ ,		LBW = 6447.2 - 56.04BL + 4.6WS - 162.2WL + 90.3TL - 188.7SL + 24.9	0.226	-	-

			LBW = 6447.2 - 56.0BL + 46WS - 162.2WL + 90.3TL -	0.235	_	_
			188.75L + 24.9BRG	0.002		
			IBW = 1000.3 + 12.28BL	0.381		-
			LBW = 1391.2 + 11.1BL - 25.4WS + 26.6WL	0.382		-
			LBW = 698.7 + 8.8BL - 25.2WS + 19.6WL + 63.0TL	0.384		_
		F	LBW = 2264.8 + 4.7BL - 24.8WS + 12.0WL + 23.0TL - 85.2SL	0.413		_
			LBW = 3956 + 7.4BL - 25.0WS + 13.72WL + 14.8TL - 87.0SL - 117.5KL	0.416	-	-
			BW = 3716.3 + 8.9BL - 25.9WS + 10.3WL + 18.4TL - 89.6SL - 113.2 + 6.9BRG	0.418	-	_
			LBW = 340 + 1.7BL	0.110	_	_
			LBW = 3183.9 + 1.9BL - 0.58WS	0.160	-	_
			LBW = 3168.6 - 0.2BL - 0.8WS + 4.7WL	0.180	_	-
		_	LBW = 2798.3 - 1.3BL - 3.2WS + 1.7WL + 28.3TL	0.119	_	_
		F	LBW = 343.9 - 3.9BL - 5.6WS + 2.5WL + 18.7 IL + 230SL	0.155		-
			36.1KL	0.265	-	-
	Local Nigerian chicken X exotic		LBW = 12/9.4 - 0.38L - 3.8WS - 4.4WL - 2.7TL + 201.35L + 60.7KL - 52.3BRG	0.399	-	-
	broiler (LNC*EB)		LBW = 4177.9 - 16.6BL	0.110		-
	reciprocal		LBW = 3190.4 - 39.7BL + 32.7WS	0.450	-	-
			IBW = 28791 - 45.2BL + 19.0WS + 40.0WL	0.850	-	-
			LBW = 3791.7 - 52.0BL + 32.2WS + 37.2WL + 1.7TL -	0.000	-	-
		М	98.5SL I BW = 4820.6 - 107.3BI + 72.6WS + 86.2WI + 9.9TI -	0.150	-	-
			141.5SL - 87.2KL IBW = 4238.9, 125.3 RL + 88.8WS + 95.0WL - 3.9TL +	0.315	-	-
			178.6SL - 98.9KL + 26.1BRG	0.339	-	-
	0		LBW = - 313.67 + 41.22BRG	0.876	19.920	P<0.01
	Sasso chicken	-	LBW = - 185.331 + 26.26BRG + 22.23TC	0.923	15.820	P<0.01
			LBW = -185.65 + 23.86BRG + 17.891IC + 11.98NC	0.932	15.080	P<0.01
Yakubu and	Nigerian Kuroiler		$LBW = -181.525 \pm 103.28950$	0.887	27.510	P<0.01
Ari (2018)	chicken	-	LBW = -413.09 + 73.50SC + 30.50WL + 23.48TL	0.940	18 300	P<0.01
			LBW = 29.98 + 14.959BL	0.821	17.920	P<0.01
	Nigerian Fulani		LBW = - 64.06BL+ 34.17NC	0.877	15.010	P<0.01
	chicken	-	LBW = - 70.32 + 8.65 BL + 27.92NC + 15.76TC	0.898	13.800	P<0.01
	Nigerian		LBW = - 981.656 + 117.564TL	0.762	9.180	_
	Marshal chicken	-	LBW = - 981.656 + 48.676BRG + 117.564TL	0.856	12.304	_
	Marshal x naked		LBW = - 447.878 + 63.141BRG	0.691	6.233	_
	neck chicken	-	BW = - 626.199 + 41.4716BRG + 78.505TL	0.735	28.733	_
Adenaike et	Naked neck		LBW = - 671.302 + 77.303TL	0.755	7.439	_
al. (2015)	chicken		LBW = -704.931 + 40.285BL + 46.221BRG	0.855	8.311	-
	Normal- feathered		LBW = - 565.974 + 68.558BRG	0.917	3.110	-
	chicken	-	LBW = - 609.395 + 34.963BL + 43.185BRG	0.939	8.938	-
	feathered chicken	-	IBW = -1072933 + 67933TI + 104214KI	0.027	25 411	_ P < 0.01
	.suthered effecter		$LBW = 0.09 \pm 0.04SC$	0.310	20.411	P < 0.01
			$LBW = -0.33 \pm 0.03SC \pm 0.00BL$	0.430		P < 0.01
			LBW = -1.47 + 0.03 + SC + 0.00BL + 0.00BG	0.540		P < 0.01
		IVI	LBW = - 1.82 + 0.02SC + 0.00BL + 0.00BG + 0.01SL	0.550		P < 0.01
Bila and	Ross 308 broiler		LBW = - 1.84 + 0.02SC + 0.00BL + 0.00BG + 0.01SL + 0.00WL	0.550	-	P < 0.01
Tyasi (2022)	chicken		LBW = - 0 .29 + 0.02SL	0.260	-	P < 0.01
			LBW = - 0.84 + 0.12SL + 0.01WL	0.330	_	P < 0.01
		F	LBW = -1.35 + 0.01SL + 0.01WL + 0.02SC	0.450	_	P < 0.01
			LBW = - 1.65 + 0.01SL + 0.01WL + 0.02SC + 0.03BKL LBW = - 1.82 + 0.01SL + 0.01WL + 0.02SC + 0.03BKL +	0.480	-	P < 0.01
			LBW = 0.584 + 0.061BL	0.240		P < 0.01
				0.000	-	D (0.01
Tyasi et al.	White leghorn	F	$LBW = -0.079 \pm 0.055BL \pm 0.0775L \pm 0.01400HL$	0.330		P < 0.01
(2024)	chicken	F	$LBW = -0.025 \pm 0.058BL \pm 0.0205L \pm 0.0205L \pm 0.0205L \pm 0.0205L$	0.370	-	P < 0.01
			LBW = - 1.471 + 0.056BL + 0.080SL + 0.12CNH + 0.109SC 0.102SC + 0.012CCH +	0.400	_	P < 0.01
P value =proba	bility value. P < 0.05	= signific	cant. = not stated. RSME = residual square mean of error. M	= male, F	= female, L	BW = live

P value =probability value, P < 0.05 = significant, _ = not stated, RSME = residual square mean of error, M = male, F = female, LBW = live body weight, CH = comb height, BKL = beak length, BL = body length, CC = chest circumference, TC = thigh circumference, SL = shank length, KL = keel length, TL = thigh length, WL = wing length, DS = drum stick, WS = wing span, BRG = breast girth, BG = body girth, SC = shank circumference, CG = chest girth, NC = neck circumference, CNH = chicken height.

Table 4 - Estimation of body weight using multiple linear regression
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Author and	Breed	Sev	Sex Model		Goodness of fit		
year	DICCU	Jex	Model	R ²	RMSE	P value	
Yunusa and	Nigerian Yoruba chicken	-	LBW = -1.21 + 0.82BL + 0.15BRL	0.970	-	P < 0.05	
Adeoti (2014)	Nigerian Fulani chicken	-	LBW = -0.09 + 0.30CC + 0.74BRL - 0.38WL	0.140	-	P < 0.05	
Yakubu and Salako	Nigerian	М	LBW = 0.070 + 0.084TC + 0.051CH + 0.016BL	0.952	-	P < 0.05	
(2009)	chicken	F	LBW = -0.727+ 0.043CH + 0.028BL + 0.066SL + 0.019CC + 0.031TC	0.820	-	P < 0.05	
Udeh et al. (2021)	Nigerian Nsuka heavy local chicken	-	LBW = -1076.39 -5.98BL + 84.97SL + 21.66CC + 24.09TC + 57.98TL	0.790	-	P < 0.05	
Bila et al. (2021)	Ross 308 broiler	М	LBW = -1.80 + 0.12 BL + 0.03 BKL + 0.23 SC + 0.11 SL	0.550	0.01	P < 0.05	
	chicken F	F	LBW = -0.33 + 0.04 BG + 0.04 BL + 0.22 SC	0.470	0.03	P < 0.05	

P value = probability value, P < 0.05 = significant, _ = not stated, RSME = residual square mean of error, M = male, F = female, LBW = live body weight, CH = comb height, BL = body length, CC = chest circumference, TC = thigh circumference, SL = shank length, TL = thigh length, BRL = breast length, BG = body girth, SC = shank circumference, WL = wing length.

Table 5: Estimation of body weight using power non-linear regression							
Author and Bread Sev	Sev	NY Medel		G	Goodness of fit		
year	Diccu	JUN	model		R ²	RMSE	P value
Semakula et al. (2011)	Ugandan indigenous chicken	-		LBW = 0.001CC ^{2.417}	0.830	-	P < 0.01

P value = probability, P < 0.01 = significant, _ = not stated, RSME = residual square mean of error, LBW = live body weight, CC = chest circumference.

Table 6 - Estimation of body weight using exponent	ential non-linear	regression
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Author and	Breed	Sov	Model		G	oodness o	f fit
year	Diccu	JCA	model		R ²	RMSE	P value
Semakula et al. (2011)	Ugandan indigenous chicken	-		LBW = 0.100e ^{0.105CC}	0.800	-	P < 0.01

P value = probability value, P < 0.01 = significant, _ = not stated, RSME = residual square mean of error, LBW = live body weight, CC = chest circumference.

Table 7 - Estimation of body weight using Polynomial non-linear regression						
Author and	Brood	Say	Model	G	ioodness o	of fit
year	DICCU	Jex	Model	R ²	RMSE	P value
Semakula et al. (2011)	Ugandan indigenous chicken	-	LBW = 0.002CC ² + 0.038CC - 0.6214	0.770	-	P < 0.01
P value = probabi circumference.	lity value, P < 0.01 = sign	ificant,	<pre>_ = not stated, RSME = residual square mean of erro</pre>	r, LBW = live	body weigh	nt, CC = chest

DISCUSSION

The live body weight and biometric traits are of economic value in chicken classification and enhancement (Yakubu and Ari, 2018; Sadick et al., 2020). This systematic review was conducted to evaluate the literature on prediction of live body weight of chickens from biometric traits using regression methods. The regression statistical methods enable weighing of

live body weight by identifying the traits that might be used for animal husbandry practices and to sale the chickens for profit (Semakula et al., 2011; Yunusa and Adeoti, 2014; Bila et al., 2021). A total number of fourteen articles were included in this review. The results indicated that stepwise linear regression was mostly used by thirty-nine percent of the articles included. The results showed that multiple regression (Yakubu and Salako, 2009; Yunusa and Adeoti, 2014; Bila et al., 2021; Udeh et al., 2021) and simple regression method (Semakula et al., 2011; Sadick et al., 2020; Udeh et al., 2021; Christophe et al., 2024) were second mostly used regression methods by twenty-two percent of the included articles each. The results showed that Yunusa and Adeoti (2014) reported the highest value of ninety-seven percent variation towards live body weight on the combination of BL and BRL using multiple linear regression method on Nigerian Yoruba chickens. The results showed that Akporhuarho and Omoikhoje (2017) reported the least value of 0.2 percent variation towards live body weight on BL using stepwise linear regression method on female EB*LNC crossbreed chickens.

Based on the author's knowledge, there was no systematic review that predicts live body weight from biometric traits of chickens using regression method. Thereof, there has been no comparison of the current findings with other systematic review findings. The implication of this systematic review is that multiple linear regression model can be used to optimally estimate live body weight of different chicken breeds, sexes and ages from differing countries. The power of this review was that there is no similar study that had been conducted to estimate live body weight using linear regression methods in chickens. The contribution of this systematic review in the body of knowledge is to suggest multiple linear regression to be used to estimate live body weight of chickens. The limitation of this systematic review was that 9 out of 14 articles did not include the age of the chickens which makes it difficult to draw conclusion for sexes and ages. Udeh et al. (2021) studied different linear regression methods to predict live body weight. Nevertheless, more studies need to be conducted on the estimation of live body weight using regression methods to validate these results.

CONCLUSION

It is concluded that multiple linear regression method is best in estimating live body weight of chickens followed by stepwise linear regression. Thus, researchers can include multiple linear regression method with combination of body length and breast length when estimating live body weight of chickens. Breeders can select chickens with the highest measurements of body length and breast length to improve live body weight of their chickens

DECLARATIONS

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Data availability

The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

Authors' Contribution

Initial thought of the study was by Joas Albino Tsenane and Thobela Louis Tyasi. Joas Albino Tsenane wrote the initial draft of the manuscript. Thobela Louis Tyasi and Obert Tada supervised, reviewed and proofread the manuscript.

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Competing interests

The authors declare no competing interests.

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