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FORTIFICATION OF TRADITIONAL BALINESE *ORET* SAUSAGE WITH CHICKEN FEET SKIN COLLAGEN: EFFECTS ON PHYSICOCHEMICAL PROPERTIES, TEXTURE, COLOR, AND SENSORY ACCEPTANCE

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ABSTRACT

Oret sausage, a traditional Balinese product made from chicken offal and blood, is limited by its nutritional value and less favorable sensory characteristics. This study evaluated the effects of chicken feet skin fortification on the physicochemical properties, amino acid profile, texture, color, and sensory acceptance of *oret* sausage. A completely randomized design (CRD) was employed with four levels of chicken feet skin addition (0%, 5%, 10%, and 15%), each with four replications. Analyses included proximate composition (moisture, protein, fat, ash), antioxidant capacity, texture profile (hardness, cohesiveness, chewiness), color (CIE *L**, *a**, *b**), amino acid composition (via HPLC), and sensory evaluation using a hedonic scale by 20 semi-trained panelists. The results showed that fortification with chicken feet skin significantly increased protein content from 21.10% (control) to 23.38% at the 15% addition level (P<0.05), and also improved pH and antioxidant capacity. However, higher levels of chicken feet skin resulted in a significant reduction in hardness (from 2548.66 g to 1080.92 g, P<0.05), indicating a softer texture, while lightness increased and redness decreased. Sensory evaluation indicated that up to 5% addition, there were no significant differences in color, texture, or overall acceptance compared to the control (P>0.05), whereas at 15% addition, aroma and flavor were significantly enhanced (P<0.05). In conclusion, fortification of *oret* sausage with up to 15% chicken feet skin can improve its nutritional quality and certain sensory attributes, particularly protein content, antioxidant capacity, aroma, and flavor, although changes in texture and color should be considered in future product development.

Keywords: *oret* sausage, chicken feet skin, physicochemical properties, texture, sensory

INTRODUCTION

Processed meat products continuously face challenges related to nutritional quality, texture stability, and sensory appeal **(Kim *et al*., 2020)**. *Oret* sausage, a traditional Balinese product, is prepared primarily from chicken offal—including liver, heart, kidneys, and blood—yet its high fat content, unstable texture, and pronounced off-flavors often undermine consumer acceptance **(Choe & Kim, 2019)**. Additionally, the elevated levels of lipids and blood components in offal-based sausages accelerate lipid oxidation and compromise emulsion stability, resulting in a shortened shelf life **(Han & Bertram, 2017)**.

Chicken feet skin, an abundant by-product of the poultry industry, is rich in collagen—a protein renowned for its gel-forming capacity and its ability to enhance emulsion stability, texture, and water-holding properties in meat products **(Mokrejs *et al*., 2017; Kim *et al*., 2017)**. Previous research has demonstrated that incorporating poultry-derived collagen can increase protein content, bolster antioxidant capacity, and improve sensory attributes in products such as nuggets and sausages **(Yoo & Kim, 2017; Araujo *et al*., 2019)**. However, the application of chicken feet skin collagen in offal-based formulations, specifically *oret* sausage, remains underexplored.

This study investigates the impact of fortifying *oret* sausage with chicken feet skin on its physicochemical characteristics, textural profile, amino acid composition, and sensory acceptance. We hypothesize that incorporating chicken feet skin will enhance protein content, increase antioxidant capacity, and improve textural stability without adversely affecting sensory acceptance. The findings are expected to elucidate the potential of chicken feet skin as a functional, value-added ingredient in offal-based meat products, contributing to the sustainable utilization of poultry by-products.

MATERIAL AND METHODS

Materials

The formulations for each treatment comprised 950 g of chicken offal and 640 g of chicken blood, to which chicken feet skin was incorporated at specified percentages of the combined offal and blood. Each batch also contained one whole egg and 238.5 g of rice flour (equivalent to 15% of the total offal and blood weight). All primary ingredients were obtained from traditional markets in Denpasar, Bali, Indonesia. The seasoning blend, which was applied uniformly across all treatments, included 70 g of Balinese spice mix, 10 g of fried chili-shrimp paste, 130 mL of coconut milk, 5 g of ground pepper, 5 g of salt, and 25 g of flavor enhancer. This study was conducted using four treatment levels of chicken feet skin inclusion, namely 0% (control), 5%, 10%, and 15%. Each treatment group was replicated four times, yielding a total of 16 experimental samples.

Experimental Design

This study employed a Completely Randomized Design (CRD) with four treatment levels representing varying proportions of chicken feet skin incorporated into the oret sausage formulation: 0% (control), 5%, 10%, and 15%. Each treatment was repeated in quadruplicate, yielding a total of 16 experimental units. The specific treatments were as follows:

- O_0 (Control): No chicken feet skin addition (1,590 g of offal and blood)
- O_1: 5% chicken feet skin addition (79.5 g from 1,590 g of offal and blood)
- O_2: 10% chicken feet skin addition (159 g from 1,590 g of offal and blood)
- O_3: 15% chicken feet skin addition (238.5 g from 1,590 g of offal and blood)

Preparation of Chicken Feet Skin

The preparation of chicken feet skin followed the method described by **Susanto *et al*. (2018)**. The chicken feet were aged for 8 hours at 16 °C, then sorted and cleaned to remove nails, the outer skin layer, and any attached impurities, ensuring a clean raw material. The skin was preheated at 75 – 80 ºC in a pressure cooker for 5 minutes to facilitate further processing. Subsequently, the chicken feet skin was ground into a fine paste and incorporated into the sausage mixture.

Production of *Oret* Sausage with Chicken Feet Skin Fortification

The chicken offal, blood, and chicken feet skin were ground using a meat grinder with an 8 mm plate, according to the designated formulation for each treatment. The mixture of offal, blood, chicken feet skin, and supplementary ingredients was blended using a mixer for 7 minutes to achieve a homogeneous emulsion. The resulting *oret* sausage emulsion was then stored at a temperature below 10 °C for

5 minutes before being stuffed into collagen casings. The raw sausages were subsequently cooked in a water bath at 70–75 °C for 30 minutes, cooled at room temperature for 15 minutes, and then stored in a freezer prior to further analysis. The entire production process was conducted in four replicates for each treatment.

Measurement of Variables

Texture Profile Analysis

The texture of oret sausage was evaluated using Texture Profile Analysis (TPA) conducted with a Texture Analyzer TXT 32 **(TA.XT express, 2008)**. Samples were prepared by cutting them into cubes measuring 3 × 3 × 3 cm and subjected to a double compression test using a 6 mm diameter probe. The analysis was performed at a probe speed of 5 mm/s, compressing the samples to 30% of their original height. Key texture parameters such as hardness, springiness, cohesiveness, gumminess, chewiness, and resilience were calculated using the instrument's built-in macro software.

Quantitative Determination of Color and Organoleptic Properties

Color evaluation was conducted objectively using a Colorimeter PCE-CSM 1. The color of *oret* sausage was reported according to the CIE color system, including L^* (lightness), a^* (redness and greenness), and b^* (yellowness and blueness). The L^* value ranges from 0 to 100, indicating black to white, with higher L^* values signifying increased whiteness. The a^* and b^* values represent chromatic coordinates, where a^* indicates the red-green spectrum ($a+$ from 0 to +127 for red intensity, $a-$ from 0 to -127 for green intensity), and b^* represents the yellow-blue spectrum ($b+$ from 0 to +127 for yellow intensity, $b-$ from 0 to -127 for blue intensity) **(Lee & Kim, 2020).**

Consumer preferences were assessed via a hedonic test with 20 semi-trained panelists **(Yusof *et al*., 2010)**. Panelists evaluated color, aroma, texture, taste, and overall acceptance on a five-point Likert scale, where 5 = like very much, 4 = like, 3 = neutral, 2 = dislike, and 1 = dislike very much.

Measurement of pH

The pH of each sample was measured using a calibrated digital pH meter (Laqua-PH210). To prepare for measurement, 5 grams of sausage were homogenized with 20 mL of distilled water, and the pH value was recorded once the reading stabilized **(Peripolli *et al*., 2025).**

Cooking Loss Analysis

Cooking loss was measured according to the procedure described by **Wang *et al.* (2025)**. Fresh sausage samples were weighed prior to cooking, then heated in a water bath maintained at 75 °C for 30 minutes. After cooking, the samples were cooled to room temperature and weighed again. The percentage of cooking loss was calculated based on the reduction in weight relative to the initial weight.

Proximate Analysis

Proximate composition was analyzed following the standardized **AOAC (2005)** procedures. Moisture content was determined by measuring the weight loss after drying the samples at 105 °C for 12 hours. Fat content was quantified using Soxhlet extraction with a solvent extraction system, while protein levels were assessed through the Kjeldahl method. Ash content was measured by incinerating the samples in a muffle furnace in accordance with AOAC guidelines. Additionally, antioxidant capacity was assessed using the DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging assay as outlined by **Molyneux (2004)**.

Scanning Electron Microscopy (SEM) Analysis

The surface morphology and emulsion distribution of oret sausage enriched with chicken feet skin were analyzed using Scanning Electron Microscopy (SEM). Prior to imaging, each sample was affixed to SEM stubs with double-sided adhesive tape and coated with a thin, 10 nm layer of gold. The SEM observations were carried out at 500× magnification and operated at an accelerating voltage of 10 kV.

Amino Acid Profile Analysis

Amino acid profiling was carried out following the **AOAC (2005)** protocol. Initially, proteins were extracted using the Kjeldahl method, after which the amino acids bound to proteins were released by hydrolyzing the samples with 6 N hydrochloric acid at 110 °C for 24 hours. The hydrolysate was then neutralized and diluted with a sodium citrate buffer at pH 2.2, followed by filtration through a 0.45 µm membrane. Individual amino acids were separated and quantified using high-performance liquid chromatography (HPLC) with pre-column derivatization, and detection was performed at 254 nm.

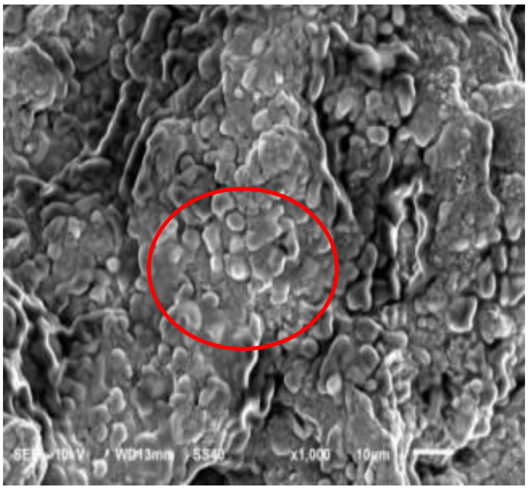
Statistical Analysis

Data were analyzed by one-way analysis of variance (ANOVA) to identify significant differences among treatments, followed by Duncan's Multiple Range Test at a 5% significance level ($P<0.05$) using SPSS version 25 software 2017. Instrument validation and calibration were performed prior to measurement: the Texture Analyzer and pH meter were calibrated according to factory standards, and the spectrophotometer was calibrated with a DPPH standard solution. Proximate analyses included quality-control checks using AOAC reference samples to ensure accuracy.

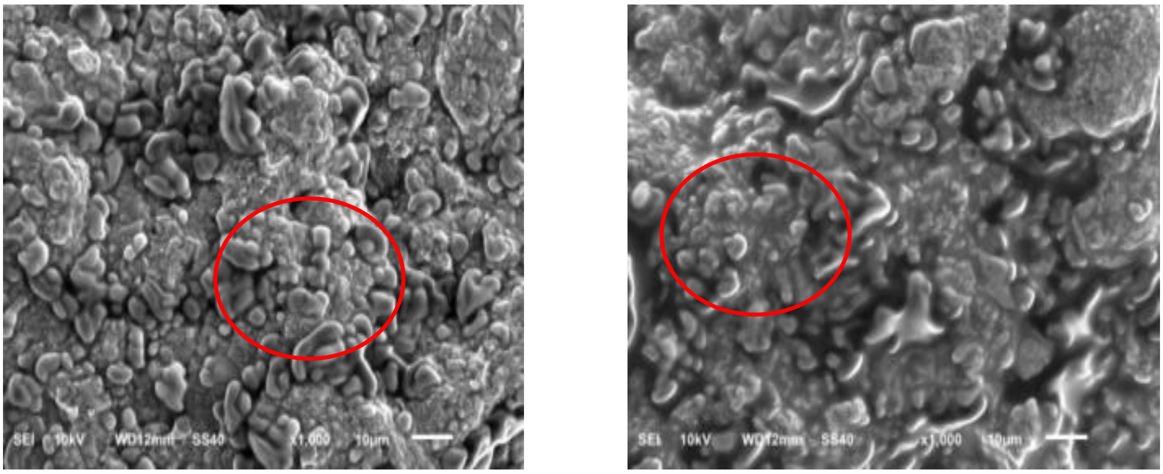
RESULTS AND DISCUSSION

Microscopic examination of *oret* sausage fortified with chicken feet skin revealed a consistently porous protein–gel matrix integrating fat globules across all treatments, with no marked differences in globule distribution. However, as chicken feet skin levels increased, fat and long-term sensory quality.

SEM Analysis of Treatment O_0



SEM Analysis of Treatment O_1



SEM Analysis of Treatment O_2 SEM Analysis of Treatment O_3

Figure 1 Microstructural Observation of *Oret* Sausage with Chicken Feet Skin Fortification at 1000 × Fat globules (◯)

Incorporating up to 15% chicken feet skin into oret sausage did not significantly affect the moisture, fat, or ash content but resulted in a significant increase in protein content (P<0.05). This increase is attributed to the high collagen content present in chicken feet skin. Collagen, as a major structural protein of connective tissues, has been reported to enhance protein levels and reinforce the protein matrix within processed meat products. Additionally, the pH level rose significantly (P<0.05) with collagen addition, which is likely linked to improved water-holding capacity and emulsion stability caused by interactions among collagen, myofibrillar proteins, and lipids. Elevated pH in collagen-enriched meats has been associated with enhanced elasticity and chewiness, key determinants of sensory quality **(Kang *et al.*, 2014)**. Antioxidant capacity also improved significantly (P<0.05) following chicken feet skin addition. This enhancement is attributed to bioactive peptides released during collagen hydrolysis, particularly those rich in lysine and glycine, which serve as effective electron donors in free-radical scavenging **(Siow & Gan, 2013; Liu *et al.*, 2023)**.

Table 1 Mean Proximate Analysis Results of *Oret* Sausage with Chicken Feet Skin Fortification

Variables	Chicken Feet Skin Addition			
	O_0	O_1	O_2	O_3
Moisture Content (%)	64.81±0.84	65.10±1.13	66.70±2.94	65.97±1.75
Protein Content (%)	21.10±0.96^a	21.82±0.85^a	22.58±1.24^a	23.38±1.26^b
Ash Content (%)	2.09±0.26	2.23±0.19	1.94±0.26	2.05±0.25
Fat Content (%)	8.31±0.18	8.20±0.20	8.30±0.18	8.10±0.08
pH Value	5.38±0.79^a	5.95±0.03^b	6.68±0.30^d	6.20±0.12^c
Antioxidant Capacity (mg/L GAEAC)	7.24±0.24^a	7.84±0.25^b	7.89±0.37^b	7.71±0.10^b
Cooking Loss (%)	1.05±0.14^a	2.00±0.72^b	1.64±0.39^b	1.89±0.09^b

Effect of Chicken Feet Skin Addition on Cooking Loss

Statistical analysis revealed that chicken feet skin incorporation significantly affected cooking loss in *oret* sausage (P<0.05). Cooking loss—which reflects the product's ability to retain water and fat during heating—increased at the 5% fortification level compared to the control but did not differ significantly at 10% and 15% levels. The elevated cooking loss at 5% may result from the high fat content of chicken feet skin interacting with meat proteins during heating, thereby releasing water from the emulsion matrix **(Kim *et al.*, 2016)**. Previous research has similarly shown that poultry skin–enriched meat products exhibit greater cooking loss due to fat melting, which destabilizes the emulsion and promotes moisture release **(Kim *et al.*, 2016; Han & Bertram, 2017)**. These findings suggest that optimizing *oret* sausage formulations to include water-binding agents or stabilizers may be necessary to mitigate cooking loss when chicken feet skin is added.

Effect of Chicken Feet Skin Addition on the Texture of *Oret* Sausage

Texture Profile Analysis indicated that chicken feet skin addition significantly reduced sausage hardness (P<0.05), likely due to collagen–myofibrillar protein interactions that produce a softer, more elastic gel network **(Okuskhanova *et al.*, 2017)**. The gel-forming properties of collagen and its association with fat from chicken feet skin enhance water binding and matrix flexibility, thereby decreasing hardness while increasing adhesiveness and gel strength **(Araujo *et al.*, 2019)**. Although springiness remained unchanged, chewiness declined significantly (P<0.05), reflecting a weaker protein emulsion structure and increased fat-induced tenderness. This aligns with observations in emulsified meat products—such as duck ham fortified with poultry skin—where added skin reduced emulsion capacity and protein solubility, yielding a softer, less compact texture **(Kang *et al.*, 2014; Kim *et al.*, 2017)**.

Table 2 Mean Texture Profile and Color Quantification of *Oret* Sausage with Chicken Feet Skin Fortification

Variables	Chicken Feet Skin Addition			
	O_0	O_1	O_2	O_3
Hardness (N)	2548.66±649.05^a	1737.11±378.37^b	1116.19±213.56^c	1080.92±319.04^c
Springiness	0.47±0.03^c	0.40±0.05^{ab}	0.45±0.05^{bc}	0.38±0.06^a
Cohesiveness	0.29±0.02^a	0.28±0.03^{ab}	0.27±0.01^{ab}	0.25±0.02^b
Gumminess (N)	734.78±151.62^a	475.12±75.61^b	304.95±53.09^c	267.62±72.60^c
Chewiness (N)	344.44±70.95^a	191.84±42.11^b	136.76±35.03^c	100.55±22.82^c
*L Value**	12.33±0.31^{ab}	12.10±0.27^a	12.68±0.47^b	12.54±0.61^{ab}
*a Value**	17.38±1.14^a	18.14±0.47^a	20.19±0.75^b	19.72±1.01^b
b Value *	6.57±0.07	6.53±0.35	6.36±0.65	6.50±0.28

Effect of Chicken Feet Skin Addition on the Color of *Oret* Sausage

Color characteristics of *oret* sausage were significantly altered by the incorporation of chicken feet skin, as evidenced by CIE L*ab** measurements (P<0.05). At 15% fortification (O_3), lightness (L*) increased markedly relative to other treatments, reflecting the superior light-reflective properties of collagen-rich skin **(Kim *et al.*, 2017)**. Redness (a*) declined progressively with higher inclusion levels, likely due to fat-mediated dispersion of myoglobin pigments within the emulsion matrix **(Kang *et al.*, 2014; Kim *et al.*, 2016)**. Yellowness (b*) did not differ significantly from the control at up to 5% addition (O_1; P>0.05), indicating stable pigment–protein interactions at low inclusion levels, but exhibited a perceptible increase at higher fortification levels, suggestive of combined fat and collagen effects on pigment distribution.

Amino Acid Profile of *Oret* Sausage with Chicken Feet Skin Addition

Oret sausage fortified with chicken feet skin contained 18 amino acids, with total content ranging from 14.17% to 15.12%. Dominant amino acids included

glutamate (2.71–3.08%), aspartic acid (1.17–1.24%), leucine (1.25–1.36%), proline (0.76–0.94%), glycine (0.64–0.92%), alanine (0.95–1.09%), valine (0.80–0.87%), phenylalanine (0.72–0.80%), lysine (0.88–0.92%), and arginine (0.75–0.83%).
Collagen degradation during cooking releases glycine, proline, and hydroxyproline—peptides that enhance viscosity and functional properties in meat emulsions **(Liu *et al.*, 2023)**. The amino acid composition of processed meat products is influenced by factors such as the formulation ingredients and processing conditions, including the levels of fat and types of protein used **(Bar *et al.*, 2020)**.
From a sensory perspective, glutamate and aspartic acid impart umami and freshness; glycine and alanine contribute sweetness; arginine, leucine, valine, and phenylalanine introduce bitterness; and lysine exhibits dual sweet-bitter notes, collectively shaping taste perception **(Berisha *et al.*, 2023)**.

Table 3 Mean Amino Acid Profile of *Oret* Sausage with Chicken Feet Skin Fortification (%)

Variables	Chicken Feet Skin Addition			
	O_0	O_1	O_2	O_3
Aspartic Acid	1.24±0.63	1.17±0.01	1.20±0.02	1.21±0.02
Threonine	0.64±0.08	0.67±0.26	0.61±0.20	0.65±0.10
Serine	0.61±0.10	0.61±0.20	0.56±0.10	0.59±0.30
Glutamic Acid	2.71±0.18[a]	3.08±0.04[b]	2.82±0.10[a]	2.82±0.10[a]
Proline	0.76±0.20	0.92±0.01	0.90±0.10	0.94±0.27
Glycine	0.64±0.24	0.89±0.10	0.89±0.27	0.92±0.11
Alanine	0.95±0.26	1.02±1.02	1.03±1.03	1.09±1.09
Cystine	0.21±0.13	0.21±0.02	0.21±0.10	0.21±0.02
Valine	0.86±0.18	0.87±0.02	0.80±0.05	0.86±0.03
Methionine	0.20±0.09	0.13±0.03	0.19±0.10	0.13±0.10
Isoleucine	0.62±0.26	0.62±0.01	0.57±0.20	0.61±0.01
Leucine	1.33±0.30	1.36±0.02	1.25±0.10	1.31±0.03
Tyrosine	0.38±0.06	0.39±0.04	0.38±0.10	0.39±0.10
Phenylalanine	0.77±0.10	0.80±0.10	0.72±0.10	0.76±0.10
Variables	**Chicken Feet Skin Addition**			
	O_0	O_1	O_2	O_3
Histidine	0.45±0.20	0.46±0.03	0.40±0.02	0.49±0.10
Lysine	0.91±0.07	0.91±0.02	0.88±0.05	0.92±0.18
Arginine	0.75±0.20	0.81±0.04	0.79±0.04	0.83±0.27
Tryptophan	0.14±0.02[a]	0.20±0.04[b]	0.12±0.01[a]	0.10±0.01[a]
Total Amino Acids	14.17±2.65	15.12±0.02	14.31±0.15	14.84±0.10

Effect of Chicken Feet Skin Addition on the Sensory Acceptance of *Oret* Sausage

Sensory evaluation revealed that adding up to 5% chicken feet skin to *oret* sausage did not significantly affect color preference, texture, or overall acceptance compared to the control (P>0.05). However, at 15% fortification, panelists rated aroma and flavor significantly higher (P<0.05), indicating that elevated levels of chicken feet skin can enhance the sensory profile of offal-based products by mitigating undesirable odors and intensifying desirable flavors.
This improvement likely stems from collagen-derived bioactive peptides generated during heating and hydrolysis, which have been shown to possess flavor-enhancing properties **(Lin *et al.*, 2010)**. Moreover, collagen's abundance of hydrophobic amino acids—particularly glycine and proline—facilitates the formation of volatile compounds that enrich meat aroma **(Lee *et al.*, 2012)**.
Prior studies corroborate these findings: **Kim *et al.* (2017)** observed improved flavor and aroma in duck ham with added duck skin without compromising overall acceptance, and **Yoo & Kim (2017)** reported that poultry skin–derived gelatin enhanced juiciness and palatability in processed meats. Additionally, hydrophobic collagen peptides can act as antioxidants, donating hydrogen ions to neutralize free radicals and thus preventing off-flavor development from lipid oxidation during processing and storage **(Siow & Gan, 2013)**.
Therefore, incorporating chicken feet skin into *oret* sausage not only improves texture and protein content but also offers the potential to enhance and stabilize sensory quality over shelf life.

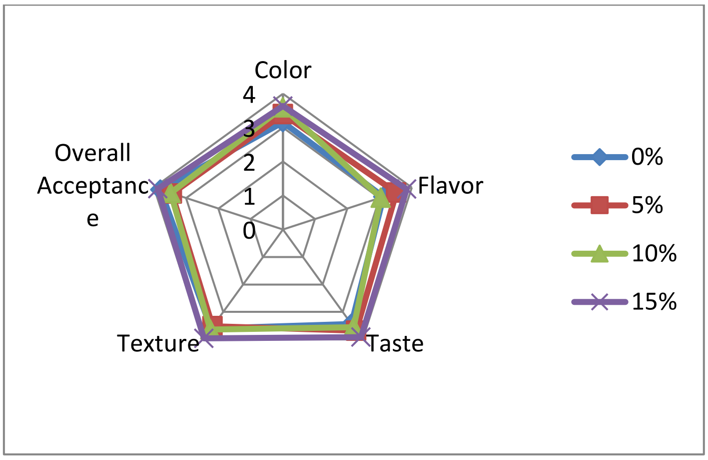


Figure 2 Mean Hedonic Scores of *Oret* Sausage with Chicken Feet Skin Addition
Twenty panelists evaluated sensory attributes in quadruplicate, using *oret* sausage without chicken feet skin as the reference standard.

CONCLUSION

This study demonstrates that fortifying *oret* sausage with up to 15% chicken feet skin significantly enhances its physicochemical properties and sensory characteristics. The addition of chicken feet skin led to a notable increase (P<0.05) in protein content, antioxidant capacity, and emulsion stability, without adversely affecting moisture or fat content. Sensory evaluation indicated that up to 5% fortification maintained color, texture, and overall acceptance comparable to the control, while 15% addition resulted in significant improvements in aroma and flavor (P<0.05). These results highlight the dual benefits of chicken feet skin as a nutritional enhancer and flavor improver, particularly addressing sensory limitations commonly associated with offal-based products. Scientifically, the findings confirm that chicken feet skin is a valuable source of collagen, functioning as both a texturizing agent and emulsion stabilizer in processed meat formulations. Collagen's role in improving water retention, reducing moisture loss during heating, and generating antioxidant bioactive peptides contributes to enhanced product stability and quality. From an industrial perspective, these results offer promising opportunities for the valorization of chicken feet skin as a high-value functional ingredient, supporting both product innovation and sustainability in the meat processing sector. Future research should explore optimization of formulation, long-term storage stability, and broader consumer acceptance to fully realize the commercial potential of collagen-enriched meat products.

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AUTHOR CONTRIBUTIONS

Conceptualization and Data Curation– Research conceptualization and data curation: INSM
Formal Analysis: INSS
Manuscript Editing: IWW
Physicochemical Analysis: INTA
Texture Profiling and Manuscript Editing: AS
Manuscript Writing – managing submission, revising and editing (grammar and clarity): RL

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