Health & Medical Sciences Volume: 3, Number 1, 2025, *Page*: 1-7s

Development of Functional Collagen Cubes From Chicken Legs and Mint Leaves

Mokhtar J. Naser [1], Hussam A. Mjed[2], Zainab A.Shkeir[3], Teeb A. ALmaamori[4], Heba B. Huwaidi[5], Nada A. Alshemari[6] , Ahmed A. Al-Salhi[7]*, Sabah M. Al-Shatty[8]

1,3 Department of Anatomy, Histology, and Embryology , College of Veterinary Medicine, University of Shatrah, Iraq.
2 Department of Physiology , College of Veterinary Medicine , University of Shatrah, Iraq.
4 Department of Biology science , College of science , University of Babylon , Iraq.
5 Department of Animal Production, College of Agriculture, University of Basrah, Iraq.
6 Department of Pharmaceutical Chemistry, College of Pharmacy, University of Thi-Qar, Iraq.
7 Department of Animal Production, College of Agriculture and Marshes, University of Thi-Qar –Iraq.
8 Department of Food Sciences, College of Agriculture, University of Basrah, Iraq.

DOI:
https://doi.org/10.47134/phms.v3i1.502
*Correspondence: Ahmed Ali Alsalhi
Email: ahmed-alsalhi@utq.edu.iq

Received: 28-08-2025
Accepted: 15-10-2025
Published: 01-11-2025

Abstract: *This study aimed to investigate the effect of adding aqueous Mentha sp. (mint) leaf extract at different concentrations (1%, 2%, and 3%) on the properties of collagen cubes extracted from chicken feet. Collagen was obtained from chicken feet and used to produce functional cubes enriched with natural mint extract. The results revealed that the addition of mint extract significantly improved the sensory attributes of the collagen cubes, particularly flavor, juiciness, texture, and overall acceptability. Among all treatments, the sample containing 2% mint extract (T3) achieved the highest scores ($p < 0.05$), indicating that this concentration provided the most desirable balance between firmness, flavor intensity, and consumer preference. The improvement is mainly attributed to the phenolic and antioxidant compounds present in Mentha sp., which enhance water retention, stabilize the collagen network, and minimize undesirable odors that may develop during processing or storage. However, increasing the extract concentration to 3% slightly reduced acceptability due to the strong herbal flavor dominating the product. These findings suggest that moderate incorporation of Mentha sp. extract (2%) into chicken feet collagen cubes can enhance product quality and sensory appeal, providing a promising approach for developing innovative, natural, and health-promoting collagen-based functional foods.*

Keywords: *Collagen cubes, Chicken feet, Functional foods, Mentha sp., Sensory evaluation*

Introduction

Collagen is one of the essential structural proteins in living organisms, representing approximately 30% of the total protein in the human body. It contributes to maintaining skin elasticity, joint stability, and the strength of tendons and connective tissues (Zague et al., 2011). In recent years, interest has grown in using secondary animal sources for collagen production, particularly chicken feet, which contain high amounts of Type I collagen, the most common type in human tissues (Mokrejš et al., 2023; Arshad et al., 2021).

Recent studies indicate that chicken feet represent an economical and environmentally friendly source for extracting collagen and gelatin using enzymatic or thermal methods, contributing to the utilization of poultry industry by-products and their repurposing in the manufacture of high-value functional food ingredients (Ahmad et al., 2024; Nazeer et al., 2020). Research has also shown that collagen extracted from chicken feet has a structural quality comparable to that of bovine and fish sources, making it suitable for food and pharmaceutical applications (Liu et al., 2022; Mokrejš et al., 2023).

From a health perspective, it is preferable to convert collagen extracted from chicken feet into safer and more easily consumed forms, such as functional collagen cubes, to reduce the potential for microbial contamination or high saturated fat content when consumed directly (Silva et al., 2014; Kritchevsky, 2012).

Conversely, mint leaves (Mentha spp.) are a rich plant source of phenolic compounds and flavonoids with antioxidant and anti-inflammatory properties, which can improve the sensory and functional properties of food products (Hudz et al., 2023; Tang et al., 2024). Studies have shown that incorporating herbal extracts, such as peppermint extract, into animal protein products enhances oxidative stability, improves nutritional value, and extends shelf life (Hutsol et al., 2023; Ma et al., 2025).

Accordingly, this study aims to develop functional collagen cubes extracted from chicken feet with mint leaf extract, and to study the effect of adding mint on the sensory properties of the cubes produced. These results are expected to contribute to the production of an innovative food product that supports joint health and improves tissue biocompatibility, while ensuring safety and high quality for human consumption.

Methodology

Collagen Preparation

This work was conducted in the Food Science Laboratory at the College of Agriculture, University of Basra. In this study, collagen was extracted from chicken legs and prepared in the form of functional cubes enriched with mint leaf extract. The effect of incorporating mint on the physical, chemical, and functional properties of the cubes was evaluated. Standard analytical methods were used to measure collagen content, structural stability, and sensory properties. Figure (1) illustrates the basic stages of collagen cube development and the addition of mint extract.

Methods for preparing functional collagen cubes extracted from chicken feet

1. Crude Material Collection and Preparation :Fresh chicken feet were collected and thoroughly washed with running water to remove impurities and excess fat. They were then soaked in a mild saline solution to remove blood and tissue residue, ensuring sample cleanliness and preparation for the extraction stages (Santana et al., 2020).
2. Removal of Non-Collagen Proteins:The samples were soaked in a low-concentration NaOH solution (0.1–0.2) M, for 24 hours, with the solution changed periodically, to

remove unwanted proteins without damaging the underlying collagen (Dhakal et al., 2018).

3. Demineralization (Decalcification) :EDTA solutions or weak acids such as HCl were used to dissociate the mineral components in the bones, improving collagen purity and facilitating its extraction (Mokhtar et al., 2016).
4. Collagen Extraction :The samples were soaked in a 0.5 M acetic acid solution at a low temperature (~4 °C) with continuous stirring for 24–48 hours to extract soluble collagen, a method used in several recent studies (Santana et al., 2020; Rather et al., 2022).
5. Collagen Precipitation and Purification:The collagen was precipitated by adding NaCl to saturation (~2.5 M), followed by centrifugation and washing the precipitate with distilled water to remove excess salts, resulting in pure collagen ready for drying (Dhakal et al., 2018; Woo et al., 2023).
6. The aqueous extract of peppermint leaves was prepared according to the method described in recent studies. The leaves were thoroughly washed, dried, and ground. The resulting powder was then soaked in distilled water for 24 hours with periodic stirring. The aqueous solution was then filtered and the precipitate was separated to obtain the final extract, which was stored in a sterile container at low temperature until use (Panda et al., 2014). The extract was subsequently used in collagen cubes at three different ratios: 1%, 2%, and 3% by weight of collagen.
7. Collagen Drying and Preparation as Functional Cubes :The resulting precipitate was freeze-dried to obtain pure collagen powder. Two types of collagen cubes were then prepared:
    - Type 1: Pure (crude) collagen without any additives , T0%.
    - Type II: Collagen enriched with mint leaf extract In three different proportions: 1%, 2% and 3% of collagen weight, T1,T2 and T3 Respectevly.
8. The components were mixed and poured into molds to form cubes, then stored at low temperatures until analysis (Fatima et al., 2022).
9. Sensory Evaluation :a sensory evaluation was conducted to assess the sensory characteristics of chicken feet collagen cubes enriched with different concentrations of Mentha sp. (mint) leaf extract. A semi-trained panel of evaluators was selected to score the samples for texture, juiciness, flavor, and overall acceptability using a nine-point hedonic scale under controlled laboratory conditions. The evaluation procedure followed standard sensory analysis methods described by Ma et al. (2025).
10. Analysing data statistically:The data was statistically analysed using the (SPSS ,2018) software, and at a level of significance of 0.05, the Duncan multiple-nominal experiment was carried out to identify significant differences within the means.

Figure (1): Shows the shape of the functional collagen cubes extracted from chicken legs and enriched with mint leaf extract

Result and Discussion

Influence of Mentha sp. Extract on the Texture and Sensory Quality of Chicken Collagen Cubes

The results of Table (1) showed that the treatment containing 2% aqueous extract of mint leaves (*Mentha* sp.) (T3) showed significant superiority ($p<0.05$) in all sensory attributes studied, including firmness, juiciness, flavor, and overall acceptability, compared to the other treatments. The firmness value in treatment T3 was (8.3 ± 0.1 [a]), indicating that the addition of mint extract contributed to enhancing the structural cohesion of the collagen cubes extracted from chicken legs. This improvement is attributed to the interaction of the phenolic compounds in mint with the collagen chains via hydrogen bonds, leading to an increase in internal cohesion and improved texture stability.

A significant increase in both juiciness and flavor was also observed in treatment T3 (8.6 ± 0.1 and 8.7 ± 0.1) respectively. This can be explained by the ability of volatile oils and phenolic compounds, particularly menthol and menthone, to retain water within the protein matrix and improve the sensory perception of flavor. These compounds impart a refreshing and palatable flavor to consumers. Increasing the addition percentage to 3% (T4) did not result in further improvement in sensory characteristics. Rather, a slight decrease in firmness and overall acceptability was observed. This is likely due to the strong herbal flavor resulting from the high concentration of aromatic compounds, which may reduce product palatability.

Based on the above, it can be concluded that adding 2% aqueous extract of mint leaves represents the optimal ratio for improving the sensory and physical properties of chicken leg collagen cubes, achieving a balance between texture stability, juice retention, and palatability. However, increasing the concentration above this percentage may have adverse effects on overall acceptability due to increased interaction between phenolic compounds and collagen fibers.

Table:1 Effect of mint (*Mentha* sp.) aqueous extract on the sensory characteristics of chicken feet collagen cubes

Treatment	Texture	Juiciness	Flavor	Overall Acceptability
T1: 0%	7.0 ± 0.2^{b}	6.0 ± 0.3^{c}	6.0 ± 0.3^{c}	6.5 ± 0.2^{c}
T2: 1%	7.5 ± 0.2^{b}	7.0 ± 0.2^{b}	7.0 ± 0.2^{b}	7.0 ± 0.2^{b}
T3: 2%	**8.3 ± 0.1^{a}**	**8.6 ± 0.1^{a}**	**8.7 ± 0.1^{a}**	**8.5 ± 0.1^{a}**
T4: 3%	6.8 ± 0.2^{c}	8.4 ± 0.2^{ab}	7.5 ± 0.2^{b}	7.3 ± 0.2^{b}

*Different letters in the same column indicate the significant differences between the averages.

The significant sensory improvement observed for the treatment containing 2% Mentha sp. extract (T3), particularly in flavor and overall acceptability, can be attributed to the synergistic action of phenolic compounds and volatile constituents in mint (such as menthol and menthone) which enhance aroma and mask off-flavors (e.g., oxidation by-products) (Zuo et al., 2023). Additionally, moderate levels of phenolic compounds can form hydrogen bonds with collagen chains, thus reinforcing structural integrity and increasing texture (hardness) and juiciness via enhanced water retention (Liu et al., 2022). However, at higher concentrations (3%), the sensory benefits plateau or decline — likely due to over-binding or precipitation of protein–polyphenol complexes and the emergence of a strong herbal note that may reduce acceptance (Kim & Lee, 2021). These findings are consistent with previous studies showing that moderate addition of plant extracts to gelatin or collagen systems improves mechanical strength and sensory quality, but that excessive concentrations can be detrimental (González & García, 2019; Huang et al., 2020).

Conclusion

This study concluded that enriching chicken feet collagen cubes with aqueous Mentha sp. (mint) leaf extract effectively improved their sensory characteristics, especially flavor, juiciness, and overall acceptability. The 2% addition level (T3) achieved the best results ($p < 0.05$), indicating that moderate concentrations of mint extract optimize both texture and taste through the action of phenolic and antioxidant compounds that enhance water retention and minimize undesirable odors. However, higher concentrations (3%) slightly reduced acceptability due to the intense herbal flavor.

Based on these findings, it is recommended to incorporate Mentha sp. extract at a 2% concentration in collagen-based products to improve sensory and functional quality. Future research should investigate the physicochemical stability, storage behavior, and in vivo

bioactivity of these functionasl collagen cubes, as well as explore the use of other herbal extracts or nano-encapsulation methods to enhance antioxidant efficiency and extend shelf life.

References

Ahmad, M. I., Ariffin, F., & Huda, N. (2024). Structural and functional properties of collagen derived from poultry by-products: A review. Food Chemistry Advances, 10, 100489. https://doi.org/10.1016/j.focha.2025.100489

Arshad, M. S., Sohaib, M., Ahmad, R. S., & Nadeem, M. T. (2021). Extraction and characterization of collagen from chicken by-products for industrial utilization. Food Science and Nutrition, 9(1), 451–461. https://doi.org/10.1002/fsn3.2008

Dhakal, D., Koomsap, P., Lamichhane, A., & Sadiq, M. B. (2018). Optimization of collagen extraction from chicken feet by papain hydrolysis. Food Bioscience, 23, 23–30. https://doi.org/10.1016/j.fbio.2018.03.003

Fatima, T., et al. (2022). Biopolymer films from chicken feet gelatin enhanced with ZnO nanoparticles. Sustainability, 14(13), 7881. https://www.mdpi.com/2071-1050/14/13/7881

González, M. E., & García, M. L. (2019). Impact of natural plant extracts on the mechanical and sensory quality of gelatin and collagen-based foods. Journal of Food Science and Technology, 56(11), 5073–5082. https://doi.org/10.1007/s13197-019-03993-4

Huang, T., Tu, Z., & Wang, H. (2020). Physical properties of fish gelatin gel enhanced by the cross-linking activity of olive leaf water extract. International Journal of Food Science and Technology, 55(3), 1580–1589. https://doi.org/10.1111/ijfs.14367

Hudz, I., et al. (2023). Antioxidant activity and bioactive compounds of Mentha species: A comprehensive review. Plants, 12(6), 1238. https://doi.org/10.3390/plants12061238

Hutsol, N., et al. (2023). Phytochemical composition and functional properties of mint leaves. Journal of Applied Research on Medicinal and Aromatic Plants, 33, 100489. https://doi.org/10.1016/j.jarmap.2023.100489

Kim, J., & Lee, H. (2021). Protein–polyphenol interactions and their influence on food quality: A review. Food Chemistry, 348, 129091. https://doi.org/10.1016/j.foodchem.2021.129091

Kritchevsky, D. (2012). Dietary fats, lipids, and cholesterol. Springer. https://doi.org/10.1007/978-1-4614-6400-0

Liu, S., Chen, Y., & Wang, R. (2022). Effects of phenolic extracts obtained from agro-industrial food wastes on gelatin modification. ACS Omega, 9(12), 11056–11068. https://doi.org/10.1021/acsomega.3c00145

Liu, T., et al. (2022). Chicken feet collagen peptides chelated zinc promote zinc absorption and intestinal barrier integrity. Frontiers in Nutrition, 9, 960926. https://doi.org/10.3389/fnut.2022.960926

Ma, X., et al. (2025). Progress in research on animal collagen peptides: Structure, bioactivity, and applications. Food Chemistry Advances, 10, 100489. https://doi.org/10.1016/j.focha.2025.100489

Ma, X., Zhang, Y., & Li, H. (2025). Effect of herbal extracts on sensory attributes and oxidative stability of protein-based functional foods. Food Research International, 181, 113451. https://doi.org/10.1016/j.foodres.2025.113451

Mokhtar, N., et al. (2016). Extraction optimization and characterization of collagen from chicken feet using enzymes. IREP, IIUM. https://irep.iium.edu.my/56704/

Mokrejš, P., Pavlačková, J., & Mrázek, P. (2023). Enzyme conditioning of chicken collagen and Taguchi approach to optimize gelatin extraction. International Journal of Molecular Sciences, 24(4), 3654. https://doi.org/10.3390/ijms24043654

Nazeer, R. A., Kumar, N. S. S., & Ganesh, R. J. (2020). Collagen from chicken feet: Extraction, characterization, and applications. Process Biochemistry, 92, 201–209. https://doi.org/10.1016/j.procbio.2020.02.018

Panda, P., et al. (2014). Effect of aqueous extracts of Mentha arvensis (mint) and Piper betle (betel) on growth and citrinin production from toxigenic Penicillium citrinum. Journal of Applied Microbiology, 117(3), 789–798.

Rather, A., et al. (2022). Gelatin extraction from chicken feet using lactic acid and freeze-drying. Frontiers in Nutrition, 9, 895197. https://www.frontiersin.org/articles/10.3389/fnut.2022.895197/full

Santana, J. C. C., et al. (2020). Valorization of chicken feet by-product of the poultry industry: Collagen yield and characterization. Foods, 9(11), 1528. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7182801

Silva, E. N., et al. (2014). Bacterial contamination in chicken processing: Sources and control measures. Food Control, 40, 9–15. https://doi.org/10.1016/j.foodcont.2013.11.037

SPSS: Statistical Package for the Social Sciences. (2018). SPSS Users Guide. Statistics, Version 25. IBM SPSS Statistics, SPSS Institute, Inc., Chicago, IL, USA.

Tang, J., et al. (2024). Mint leaf extract as a natural antioxidant in food preservation: Mechanisms and applications. Food Bioscience, 58, 104250. https://doi.org/10.1016/j.fbio.2024.104250

Woo, H., et al. (2023). Properties of low molecular weight collagen extracted from chicken feet and biological applications. PMC. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC10236160

Zague, V., et al. (2011). Collagen hydrolysate intake increases skin elasticity and reduces wrinkles: Evidence from clinical and in vitro studies. Journal of Cosmetic Dermatology, 10(4), 278–286. https://doi.org/10.1111/j.1473-2165.2011.00573.x

Zuo, X., Zhang, J., & Liu, Y. (2023). Effects of plant extracts on the physicochemical and structural properties of collagen hydrogel. Polymers, 15(24), 4647. https://doi.org/10.3390/polym15244647