



Post-harvest intervention of chicken meat effected by myopathy: potential of enzymatic marination in quality improvement

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ARTICLE INFO

Keywords:

Broiler
Genetic selection
Prevalence
Exoenzymes

ABSTRACT

Over the past decade, the incidence of myopathies in broiler chickens, such as wooden breast myopathy (WB), white striping (WS), and spaghetti meat (SM), has significantly increased in intensive poultry production systems worldwide. These conditions negatively affect the sensory, nutritional, and technological quality of meat, resulting in substantial economic losses. In Serbia, although no official prevalence data exist, empirical observations suggest a rising frequency of these defects. One promising approach to improve the quality of affected meat involves marination using natural proteolytic enzymes (bromelain, papain, ficin). This review presents current knowledge on broiler myopathies, their impact on meat quality, and use of exogenous enzymes in meat processing. Additional focus is given to marination parameter optimization, emerging technologies, and strategic recommendations for industry and research.

1. Introduction

Modern poultry production has undergone a remarkable transformation over the past 50 years, driven by advances in genetics, nutrition, and management practices. These changes have enabled broiler chickens to reach market weight in nearly half the time compared to traditional breeds, with a marked increase in breast muscle yield. While these achievements have contributed to meeting the growing global demand for affordable animal protein, they have also led to unintended consequences related to muscle tissue health (Korver, 2023).

One of the most concerning outcomes is the rising prevalence of growth-related myopathies affecting the *Pectoralis major* muscle, such as wooden breast myopathy (WB), white striping (WS), and spaghetti meat (SM) (Giampietro-Ganeco *et al.*, 2024). These conditions compromise the organoleptic and functional properties of meat, leading to consumer rejection, reduced processing yields, and significant economic losses across the supply chain. Industry estimates from North America alone suggest that losses due to downgraded chicken meat affected by myopathy may exceed US\$1 billion annually.

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Paper received September 9th 2025. Paper accepted September 11th 2025.

The paper was presented at the 63rd International Meat Industry Conference “Food for Thought: Innovations in Food and Nutrition” – Zlatibor, October 05th-08th 2025.

Published by Institute of Meat Hygiene and Technology – Belgrade, Serbia.

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From a sustainability perspective, the inability to efficiently utilize affected meat exacerbates food waste and undermines production efficiency. Given the increasing pressure to create circular and resource-efficient food systems, the poultry sector must develop innovative, science-based strategies to mitigate quality defects while preserving product value (Barbut, 2024). The integration of biotechnological methods, such as enzymatic marination, can provide a viable and scalable solution. Enzymes offer the potential to improve texture and sensory attributes of downgraded meat, thus enabling its reintegration into high-quality processed products.

2. Prevalence and contributing factors of broiler breast myopathies

The three most prevalent breast myopathies in broilers are WS, WB, and SM. Studies show that WS can affect up to 96% of birds in moderate forms, and WB in more than 10%, especially in birds with high live weights. Histologically, these conditions are characterized by degeneration and necrosis of muscle fibers, infiltration of fat and connective tissue, and inflammatory responses. Globally, the prevalence of these myopathies varies notably depending on region, production system, and genetic lines used. In North America (the USA and Canada), the prevalence of WS is 40–96% and WB 10–25%. In Europe, WS occurs in 20–60% of cases and WB in 5–15%. In Asia (China, Thailand), the prevalence of WS is 20–70%, and WB is lower. In South America and Oceania, it is similar to that in North America (Che *et al.*, 2022). For Serbia, there are no published data on WB myopathy.

Factors that influence the development of the myopathies are: Intensive genetic selection for fast growth and breast muscle yield, pushing the physiological limits of muscle development; Metabolic and oxidative stress resulting from rapid muscle growth that outpaces vascularization and repair capacity; Nutritional imbalances that may exacerbate muscle damage; Management practices, including housing conditions and handling during transport and slaughter (Barbut *et al.*, 2024). For instance, a landmark study synthesizing data from multiple countries suggested that incidences of WB and WS are negligible or absent in slower-growing breeds or traditional production systems, but can reach epidemic proportions in commercial fast-growing lines bred for maximum breast yield (Petracci *et al.*, 2019).

3. Use of exogenous enzymes in meat processing

Exogenous enzymes such as bromelain (from pineapple), papain (from papaya), and ficin (from fig latex) have been widely studied for their ability to improve meat tenderness by hydrolyzing muscle proteins and connective tissue. In addition to the aforementioned enzymes, bacterial enzymes are also used as exoenzymes, namely *Bacillus* proteases (*B. subtilis*) and aspartic proteases (*Aspergillus oryzae*). These five exoenzymes are generally recognized as safe (GRAS) for use in the meat industry (Baltić *et al.*, 2021). Optimization is critical to balancing enzymatic tenderization without compromising structural integrity. Literature suggests using bromelain concentrations in marinade between 0.1–0.3%, papain at 0.05–0.2%, and ficin at similar ranges, with marination times from 30 minutes to 2 hours at 4–8°C.

Bromelain and papain are highly water-soluble (Boonkerd and Wantha, 2024). Bromelain's stability and proteolytic activity are significantly influenced by both pH and temperature conditions. The optimal pH for bromelain is generally between 3 and 7, with some sources citing a slightly narrower range of 5.5–8.0, or even 6.5–7.5. Beyond this range, its activity declines, although both acidic and basic forms preserve substantial enzymatic activity, down to pH 4.0. Stem bromelain operates within a pH range of 4–10, although its activity decreases above pH 10 (optimal values of 4–8), whereas fruit bromelain functions properly at pH values of 3–8. Its stability is also pH-dependent; it remains stable at pH 5.0 over extended time periods and across all temperatures, likely due to the proximity to the pH of fresh pineapple fruit. However, its activity and specific activity continue to rise up to pH 7.0 (Varila *et al.*, 2021). Papain's stability and activity are significantly influenced by both temperature and pH, with optimal conditions varying based on the context. Generally, papain exhibits stability over a broad temperature range, with some studies indicating activity from 10 to 90°C, while others suggest an optimal temperature between 40 and 80°C. However, the optimal temperature for papain activity is often reported at around 60°C, with the enzyme retaining its activity at higher temperatures, but experiencing decreased stability over time. Papain retains activity at higher temperatures, but its stability decreases over time (Baralola *et al.*, 2023). For example, at 80°C, the enzyme functions properly, but may rapidly lose its activity within an hour at

90°C. Papain's activity is also highly pH-dependent, with optimal performance often reported between pH 6 and 7, although a range from 3.0 to 9.0 is also cited, depending on the substrate (Nascomento et al., 2024). Some studies specify optimal pH ranges around 5.0–5.5, while others report it to be around 7.5 (Kusumasari et al., 2024). Papain's stability significantly decreases under strong acidic conditions and extreme pH levels, with better stability at neutral pH values. Immobilized papain can retain more activity at pH 12 than free papain, while pH levels of 5.5–6.5 enhance enzyme activity while suppressing bacterial growth. Overexposure can lead to overly soft texture, requiring strict process control, especially in industrial applications. Ficin is a cysteine protease (occurring in four forms) extracted from the trees *Ficus carica* and *Ficus glabrata*, and it is used in the food industry. In dairy processing, it is used to hydrolyze milk proteins intended for infant and elderly nutrition in order to reduce the incidence of allergic reactions. It is also used in cheese production, particularly when protein content is high, as in camel milk. It is additionally used for meat tenderization. According to recent studies, it is frequently used in human medicine as an oral antiseptic for chronic inflammations of the oral cavity and for the prevention of dental caries. It is also used as a cosmetic agent for skin diseases. It has antitumor activity. The optimal pH for the proteolytic activity of ficin is between 5 and 8, and the optimal temperature is 37–55°C (Aider, 2021).

4. Optimization of marination parameters

In chicken breasts affected by myopathies, enzyme-assisted marination can significantly reduce meat toughness, improve water retention, and enhance sensory properties such as succulence and palatability. For instance, Tasoniero et al. (2016) observed a significant reduction in Warner-Bratzler shear force from 22.8 N to 17.2 N after bromelain treatment, with parallel improvements in color and juiciness. Enzymatic marination also led to reduced cooking loss and improved consumer acceptability of WB-affected meat. Proteolytic enzymes belong to the class of hydrolases and can be classified in a variety of ways. Considering their chemical mechanism of catalysis, proteolytic enzymes are divided into serine proteinases, aspartic proteinases, threonine peptidases, metalloproteinases, and cysteine proteinases—the latter category includes bromelain, ficin, and papain (Obaha and Novinec, 2023). Further-

more, proteolytic enzymes are categorized by their mode of action into exopeptidases and endopeptidases (proteinases). Combining these two categories, an enzyme can be, for example, a serine endopeptidase or a serine exopeptidase, etc. (Kieliszek et al., 2021). Bromelain, ficin, and papain are cysteine endopeptidases. More specifically, bromelain cleaves peptide bonds between aromatic, basic, and hydrophobic residues (Kumar et al., 2023), ficin cleaves bonds in proteins like casein or gelatin, and papain has a broad specificity (Vatic et al., 2020). Lately, applying exogenous enzymes from plant sources, such as papain, bromelain, and ficin, has been suggested for meat cuts, through the methods of injection, marination, or infusion in order to reduce meat hardness (Maqsood et al., 2018). Bromelain and papain are cysteine proteases with a broad, often indiscriminate, proteolytic activity and can degrade both myofibrillar and connective tissue proteins (Fernández-Lucas et al. 2017). Furthermore, the microstructural changes that result from exposure to these proteases can also result in the release of cell content/endogenous enzymes, which would likely enhance proteolysis (Barekat & Soltanizadeh, 2017). Thus, the observed changes in myofibrillar spacing and increased fragmentation are probably associated with decreased muscle tenderness and improved texture quality. Valorizing WB myopathy with enzyme-based interventions can enhance texture and yield, reduce food waste, and create new product lines such as marinated fillets, shredded poultry, or nuggets. Studies have reported up to 20% improvements in water-holding capacity and consumer acceptance. From an economic perspective, converting downgraded meat into high-value products supports profitability and sustainability goals.

Future research is needed on enzyme formulations and delivery systems (e.g., encapsulation). Long-term strategies include genetic selection for robustness and pre-harvest interventions to reduce myopathy incidence.

5. Conclusion

Myopathies in broiler chickens challenge meat quality and economic viability. Enzymatic marination offers a practical and science-based solution for valorizing affected meat. Future directions include optimizing process conditions, developing safety protocols, and scaling up industrial trials. National classification of myopathic meat could aid regulatory clarity and support quality-based processing

incentives. Together, these efforts can support sustainable poultry production in Serbia and beyond. Novel enzyme formulations and delivery means

should be investigated. Genetic selection of poultry for robustness and pre-harvest interventions could lower myopathy incidence.

Disclosure Statement: No potential conflict of interest was reported by the authors.

Funding: This work was supported by the Ministry of Science, Technological Development and Innovations of the Republic of Serbia, based on the Contract on the implementation and financing of scientific research work in 2025. (Contract No. 451-03-136/2025-03/200050, dated 04.02.2025.)

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