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Review

Innovative Techniques to Enhance Muscle Protein Digestibility

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Abstract

Meat is an important supply of proteins for our diet. Proteins are unique among nutrients in that they can refold and reaggregate into different structures throughout processing and digestion, which may affect how digestible they are as a whole. Consumers' increased desire for food products high in protein has led to a lot of meat-based food products being produced. But it won't help if these protein-rich foods can't be broken down into absorbable amino acids and peptides that our bodies can use. Maximizing the use of the available protein sources is crucial as worries about farming's sustainability grow (meat foods). Attempts have been made to improve the bioavailability and digestibility of protein-rich meat products through advancements in processing technologies like pulse electric field, hydrodynamic shock wave processing high-pressure processing, and ultrasound processing.

Keywords: Electric Field, Hydrodynamic Shock Wave, Pulse Ultrasound Wave, Protein Digestibility

Introduction

Meat is an excellent source of proteins, vitamins, minerals and fat, all of which have been shown to improve human health. Demand for high-quality meat, particularly red meat, is expected to rise (Mullen *et al*., 2017), with eating quality being the most significant consideration in consumer meat selection. Flavor, juiciness, and tenderness are the three key attributes that determine meat sensory satisfaction (Aaslyng and Meinert, 2017). Tenderness, defined as "the ease with which meat structure is disorganized during mastication as perceived by the consumer" (Lepetit and Culioli, 1994), has been shown to play the most important role in consumer purchasing decisions (Morton *et al*., 2018), particularly repeat buying (Miller *et al*., 2001). Meat quality has become a more important concern for consumers in terms of health and sensory features. The quality of a protein depends not only on the composition of essential amino acids but also on its digestibility i. e. the proportion of food nitrogen that becomes available for absorption after ingestion. The essential amino acid content and the extent to which these amino acids are available within the body determine the quality of proteins. High-quality proteins contain all essential amino acids in amounts greater than their reference levels detailed in FAO/WHO/UNU (2001) and have a digestibility comparable to or better than those of egg-white or milk proteins.

Studies have been conducted on almost all aspects of digestion including the effects of traditional food processing and cooking methods on the digestibility, bioavailability, and absorption of proteins. However, studies that examine the effects of novel processing technologies, such as high-pressure, pulsed electric field, shock wave, and ultrasound, on the digestion kinetics of food proteins, particularly muscle proteins, are still limited and require scientific attention as there are several unanswered questions. Technological advances over time led to the creation of a number of unique techniques for enhancing tenderness that is envisioned as quick, affordable, nonthermal, environmentally friendly, and energy-efficient technologies. To comprehend the viability and advantages of the novel technologies, it is vital to compare these cutting-edge technical procedures with the currently used industrial methods.

Protein Digestion

Food researchers are interested in protein digestion because there is a strong link between protein consumption and health, especially in older populations. Protein digestion is a complicated process that involves many physical, physiological, and biochemical factors (Lonnie *et al*., 2018). Protein digestion involves two simultaneous processes. First, the mouth and stomach undergo mechanical changes. This modification breaks up muscular meal clumps. This makes food simpler for enzymes to digest in the stomach and intestines. Proteases break down large protein molecules into smaller ones (free amino acids and short peptides) that may be absorbed into the bloodstream (Alminger *et al*., 2014).

Need for Improving Muscle Protein Digestibility

Enhancing the digestion of muscle protein is necessary. Every year, 83 million people are added to the world's population, which has an average growth rate of 1.10 percent and puts further pressure on the food supply (United Nations, 2017). Protein digestibility may be favorably or adversely influenced by the denaturation patterns of processed proteins, which depend on the processing circumstances and the source of the protein. To order to enable processing conditions suitable for various meat cuts and from various species, extensive research is needed to understand the optimal processing conditions of these emerging technologies towards the favorable effects on protein digestibility as affected by protein structure and muscle configuration. In particular, "in vitro" techniques for studying meat digestion need to be improved and standardized. In a diet of poor quality, protein is the limiting macronutrient; yet, the low digesting efficiency and digestibility of some proteins often result in their restricted bioavailability. Aging impacts all aspects of gastrointestinal digestion (digestion, absorption, and excretion) of nutrients, which is another strong reason for the development of muscle foods with improved digestibility (Baker and Blakely, 2017).

Emerging Methods of Processing for Improving the Digestibility of Muscle Proteins

Several thermal and non-thermal emerging methods may improve muscle proteins' susceptibility to gastrointestinal proteases, improving protein digestibility (Bhat, *et al*., 2019b). Emerging technologies may denature proteins and muscle microstructure by changing their structural and functional characteristics and affect the unfolding and refolding of proteins, influencing the diffusion of gastrointestinal proteases deep into the protein matrix and the accessibility of their cleavage sites. These technologies are exclusive to the meat industry and need more research before they can be used commercially to produce meat products with enhanced digestibility. Other methods, such as heat processing, the use of enzymes, and aging, are well-established in the food industry and might be enhanced to produce unique products with increased protein digestibility, but at the expense of higher resource and energy costs or a loss of sensory attributes.

Table 1: Novel processing techniques have an impact on meat proteins digestibility

Processing technologies	Power and time	Type of muscle	Muscles digestibility	References
Pulsed electric field	10 kV, 20 Hz, 20 µs	Cold-boned bovine Bicep femoris	Improved	Bhat et al. (2018b)
	10 kV, 90 Hz	Cold-boned deer Longissimus dorsi	Improved	Bhat et al. (2019)
	10 kV, 20 Hz, 20 µs	Bovine Semimembranosus	Improved	Bhat et al. (2019)
	0.7 and 1.5 KV/cm, specific energy of 90 to 100 kJ/kg	Beef brisket	Not improved	Alahakoon et al.(2019)
High-pressure processing	600 MPa	Bovine longissimus dorsi muscle	Improved	Kaur et al. (2016)
Ultrasound processing	20 kHz, 600 W for 15 min	Infant meat puree	Improved	Luo et al. (2021)
	20 kHz, 464 ± 9 W) for 5 min	pāua meat	Improved	Bagarinao et al. (2020)
Hydrodynamic shockwave processing	11 kJ/pulse	Beef brisket steaks	Improved	Chain et al. (2018)
	83, 104 and 124 MPa	Bovine Biceps femoris steaks	Not improved	Schilling et al. (2002)

Pulsed Electric Field (PEF)

According to reports, PEF treatment of meat improves mass transfer during drying and transport of the product. Due to its ability for cell membrane permeabilization with better micro diffusion. PEF may alter various qualitative characteristics of meat, such as texture, colour and improvement in water-retention capacity, and increase mass transfer processes such as curing and brining (McDonnell *et al*., 2014). Additionally, the mild ohmic heating (Lindgren *et al*., 2002) that caused a moderate temperature rise (5–30°C) during PEF processing could have an impact on the meat's quality (O'Dowd *et al*., 2013), and the two together might have a thermo-electric effect on the membranes of the muscle cells (Ortega-Rivas, 2011) and thus on meat quality attributes. Meaty, roast beef, juicy, browned, fatty and salty are examples of "positive" attributes (Ma *et al*., 2016)

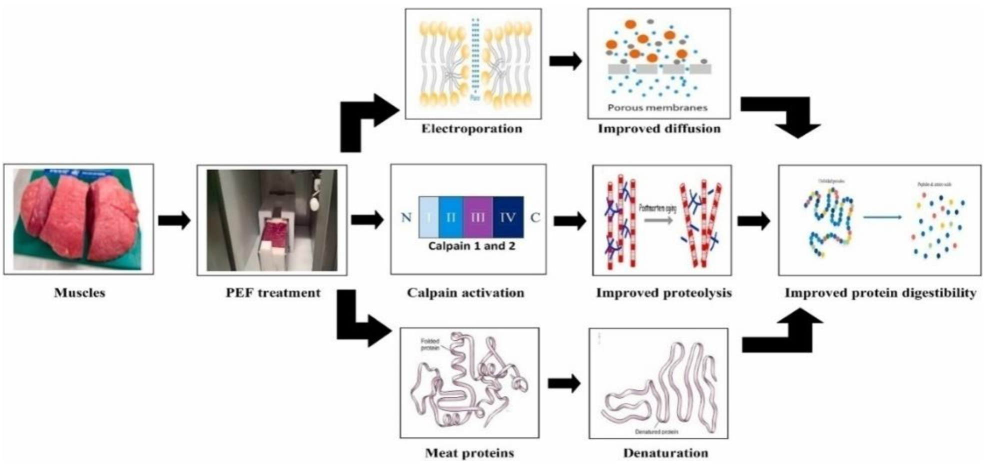

Fig. 1: Mechanisms of pulsed electric field for improving the digestibility of muscle proteins (Bhat *et al*., 2021)

Pulsed electric field processing, as a non-thermal method, diminishes nutritional and sensory aspects of foods less than conventional thermal processing technologies (Buckow *et al*., 2013). In comparison to conventional processing methods, it offers several benefits, including a lower treatment temperature, a shorter processing time, and the possibility of continuous flow (Walkling-Ribeiro *et al*., 2011). Through the early activation of calpains by the release of calcium ions from cell organelles as a result of increased membrane permeability, pre-aging muscle therapy with PEF may aid in the tenderization process. This likely mechanism is further reinforced by research demonstrating that a post-mortem ageing time is necessary to provide the tenderization advantage of PEF through enhanced proteolysis (Warner *et al*., 2017). However, other variables, such as the release of cathepsins from lysosomes, the acceleration of glycolysis owing to the release of calcium (pre-rigour muscles), and the physical disturbance of muscles may also play a role. PEF treatment reduces protein-bound mineral availability in meat (Khan 2017) and increases muscle temperature during processing (Bekhit *et al*., 2014), suggesting probable denaturation at high treatment intensities. The processing of PEF does not result in adverse effects such as significant structural and oxidative alterations or the production of off-flavours. Additionally, there is no evidence of its toxicity or environmental dangers (Pal, 2017).

High-Pressure Processing

High-pressure processing (HPP) is a non-thermal technology that has several applications in meat processing such as tenderization, salt reduction, and strategy to mitigate the quality defects of low-fat meat products, and quite recently has been used to improve digestibility (Bhat *et al*., 2018b). For commercial uses in meat and muscle products, HPP has often been utilized in the MPa range of 200–600, with processing times typically being no longer than 20 minutes for financial considerations. It can modify the functional and nutritional properties of foods, affecting protein quality and digestibility with positive or negative effects. These modifications in protein structures induced by HPP can influence the digestive behavior of proteins because the digestibility of proteins from several sources has been reported to change as a function of their secondary structure (α-helix, β-sheets, β-turns, and random coil) (Deng et *al*., 2015). As a result, pressure-induced changes in protein structure can alter how easily proteins are absorbed during gastrointestinal digestion. Studies have shown that HPP has a good effect on the protein digestibility of a variety of diets, including foods that come from muscles (Cepero-Betancourt, 2020).

Ultrasound

Ultrasound is also a non-thermal processing approach that employs sound waves with a frequency exceeding 20 kHz, which surpasses the hearing limit of the human ear and primarily functions by creating bubble cavitation in the biological matrix. According to reports, ultrasound is economically viable, uses less energy, has a greater mass transfer rate without affecting the essential qualities of foods, and satisfies all process criteria, including scalability (Chemat, 2011). This enhanced digestibility was attributable to the ultrasound-induced structural changes in proteins, which exposed enzyme cleavage sites (Wang, *et al*., 2016). The samples generated earlier had a wide range of smaller peptides and free amino acids throughout the intestinal and stomach phases of digestion utilizing pepsin and pancreatin, respectively. This improvement in digestibility was linked to several microstructural and ultrastructural modifications brought about by ultrasonic therapy, including protein unfolding, which has been reported to accelerate gastrointestinal protease's hydrolysis of proteins. Actomyosin particle size was affected by ultrasound; this impact was ascribed to the protein dissociation during cavitation, which also resulted in the breakdown of aggregates and agglomerates. Studies have shown how sonication affects the size of various protein particles (Zhao *et al*., 2014). All of the modifications to tropomyosin's structure and characteristics brought on by ultrasound were intended to make it more vulnerable to digestive enzymes.

Hydrodynamic Shockwave

Hydrodynamic shockwave is a new approach that uses mechanical high-pressure pulses caused by a high-voltage electrical discharge between two electrodes (electrohydraulic) underwater and may generate up to 1 GPa of underwater pressure (Bolumar *et al*., 2013). Production of underwater shockwaves through continuous electrical discharge has been characterized as an inexpensive, safe, and reproducible means of propagating mechanical pressure pulses into flesh to generate a "rupture effect" that splits the muscle tissue (Bolumar and Toepfl, 2016). Therefore, it may increase the quality and digestibility of meat proteins and trigger structural changes in meat. Shockwave processing research has primarily concentrated on meat tenderization, particularly for red meats (Bolumar *et al*., 2013). Depending on the processing parameters and muscle types, shockwave processing of bovine Longissimus dorsi, Semimembranosus, and Biceps femoris muscles reduced Warner-Bratzler (WB) shear force by 10 to 70% (Bolumar *et al*., 2014). Shear resistance of beef loin decreased by 10 to 50% following explosive SW treatment. Aside from red meat, both explosive and electrical SW treatment improved the tenderness of poultry pectoralis by 12 to 42 percent (Bowker *et al*., 2010).

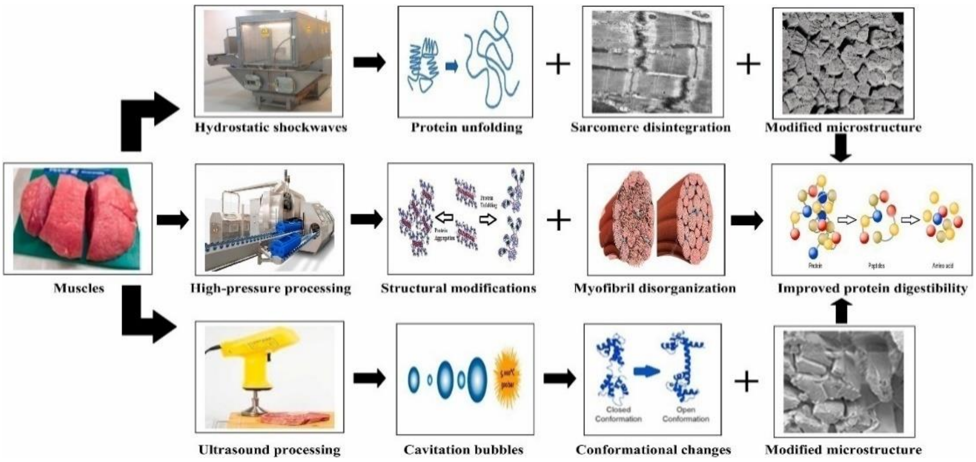

Fig. 2: Mechanisms of hydrostatic shockwaves, high-pressure and ultrasound for affecting the digestibility of muscle proteins (Bhat *et al*., 2021)

Conclusion

Protein digestion is a complicated process that is significantly affected by heat and non-thermal mechanisms that may create structural changes in proteins. The digestibility and bioavailability of muscle proteins are altered by processing them under various circumstances or with a combination of procedures; thus, these processes must be adjusted. While some of these techniques, such as high-pressure and ultrasound, are currently in use in the meat industry and might be refined to generate innovative muscle meals with higher digestibility, others, such as PEF and shockwaves, need more research before they can become commercially viable. Meat proteins are susceptible to aggregation during high-temperature thermal processing, which causes the unwinding of tightly coiled polypeptide chains and the production of intermolecular cross-links, resulting in the development of massive aggregates. Consequently, the use of non-thermal processing methods, such as the pulsed electric field, for meat processing is of interest. PEF technology offers ecologically beneficial and energy-efficient choices for a variety of foods, including meat. PEF may find commercial use in the future if additional study optimizes technical inputs for various muscles, cuts, and species.

Contribution by authors

All the authors contributed equally to writing the manuscript. The final manuscript was read by all others and consented to publication.

Conflict of Interests

There is no conflict of interest.

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