JOURNAL OF FOOD AND BIOTECHNOLOGY

Review Article | ISSN 3041- 6299 | Open Access

Understanding Protein Aggregation during Thermal and Freeze Processing of Animal–Based Foods

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Citation: Fahmida Khatoon (2023). Understanding Protein Aggregation during Thermal and Freeze Processing of Animal-Based Foods. *Journal of Food and Biotechnology.* **14 to 20. DOI: https://doi.org/10.51470/FAB.2023.4.1.14**

17 January 2023: Received | 19 February 2023: Revised | 11 March 2023: Accepted | 20 April 2023: Available Online

Abstract

Protein aggregation is a pivotal phenomenon that occurs during the processing and preservation of animal-based foods, directly influencing their nutritional value, functionality, and consumer appeal. Thermal treatments such as cooking, pasteurization, and sterilization are essential for food safety but often disrupt protein structure, leading to unfolding, exposure of hydrophobic regions, and aggregation through hydrophobic and disulfide interactions. Similarly, freeze processing—widely employed to extend shelf life—subjects' proteins to ice crystal damage, cry concentration, and repeated phase transitions, which destabilize native conformations and promote aggregation. These structural modifications affect food quality on multiple levels: they alter texture, juiciness, and water-holding capacity; reduce solubility and digestibility; and influence sensory acceptance. Importantly, the extent and nature of aggregation vary with protein type, processing intensity, and storage conditions, making meat, fish, milk, and egg proteins particularly susceptible. Recent advances, including microencapsulation, cryoprotectants, and alternative processing technologies such as high-pressure or ultrasound, offer strategies to mitigate detrimental aggregation while harnessing beneficial structural changes for desired texture formation. Understanding the molecular basis and practical consequences of protein aggregation is crucial for developing optimized processing methods that preserve quality, enhance nutritional retention, and meet consumer expectations, consolidates current knowledge on aggregation mechanisms, consequences, and control strategies, with emphasis on animal-derived food systems.

Keywords: *protein aggregation, thermal processing, freeze processing, meat proteins, milk proteins, food quality, animal-based foods.*

1. Introduction

Proteins are among the most versatile biomolecules in animal-derived foods, serving not only as primary sources of essential amino acids but also as structural and functional components that shape food quality. In foods such as meat, fish, milk, and eggs, proteins contribute to solubility, water-holding capacity, gelation, foaming, and emulsification—functional properties that underpin textural integrity, sensory appeal, and consumer acceptance. From a nutritional standpoint, proteins supply indispensable amino acids required for growth, repair, and metabolic functions. However, proteins are inherently sensitive to environmental conditions [1]. Their native conformation is stabilized by a delicate balance of covalent and non-covalent forces, including hydrogen bonding, electrostatic interactions, hydrophobic associations, and disulfide linkages. Processing and storage often disrupt this balance, causing conformational changes that may be reversible (denaturation) or irreversible (aggregation).

Protein aggregation, in particular, is a critical structural modification with profound implications for food quality. It can be defined as the self-association of unfolded or partially unfolded proteins into higher-order complexes via intermolecular forces. Aggregation may be beneficial or detrimental depending on the food matrix and processing goals. For example, controlled aggregation is desirable in gelation (egg custards, surimi gels, yogurt), where it contributes to structure and mouthfeel. Conversely, uncontrolled aggregation can lead to undesirable toughness, dryness, phase separation, or reduced digestibility, thereby undermining both sensory attributes and nutritional value [2], the diverse food processing methods, thermal and freeze processing are two of the most prevalent in the animal-based food industry. Both play indispensable roles in ensuring safety, extending shelf life, and facilitating product distribution across global markets. Yet, both impose stresses that profoundly affect protein stability and aggregation behavior.

Thermal processing and its effects

Heat treatments such as cooking, pasteurization, blanching, and sterilization are applied widely to eliminate pathogenic microorganisms and inactivate spoilage enzymes. However, heat also acts as a major driver of protein denaturation and aggregation. Elevated temperatures disrupt hydrogen bonds and destabilize secondary and tertiary structures, exposing hydrophobic groups that are normally buried within the protein core. These exposed regions can interact with other unfolded proteins, initiating aggregation through hydrophobic clustering, hydrogen bonding, and disulfide exchange reactions [3]. The extent of heat-induced aggregation depends on multiple variables, including the type of protein, heating temperature and duration, pH, and ionic strength. For instance, whey proteins in milk begin to aggregate above 70 °C through disulfide cross-linking,

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which can alter milk stability and functionality in cheese production. In meat, myofibrillar proteins such as myosin undergo denaturation and aggregation during cooking, influencing tenderness, water retention, and juiciness. While moderate heat treatments can generate desirable textures, excessive heating often results in tough, dry products with reduced nutritional quality due to impaired digestibility of aggregated proteins.

Freeze processing and its effects

Freezing is another essential preservation technique, widely valued for extending shelf life and maintaining nutritional quality. However, freeze processing is not without drawbacks. The formation of ice crystals during freezing leads to physical disruption of muscle fibers in meat and fish, destabilization of protein–water interactions, and alterations in pH and ionic strength in the unfrozen fraction. These changes destabilize native protein conformations, favoring aggregation [4]. The rate of freezing plays a pivotal role: slow freezing produces larger ice crystals that cause more extensive damage, while rapid freezing generally preserves protein integrity more effectively. Furthermore, repeated freeze–thaw cycles exacerbate structural destabilization, leading to cumulative aggregation and quality loss. In fish, freeze-induced myosin aggregation results in reduced elasticity and water-holding capacity, producing a fibrous and less appealing texture. In dairy products, freezing can destabilize casein micelles, causing protein precipitation and syneresis upon thawing.

Aggregation as a double-edged sword

Protein aggregation is thus a double-edged sword in food processing. On one hand, aggregation is necessary for the development of many desirable textures and structures—such as gels, foams, and emulsions—that define consumer expectations of specific products. On the other hand, uncontrolled or excessive aggregation undermines sensory attributes, reduces protein solubility, and diminishes digestibility, thereby affecting both nutritional quality and consumer acceptance [5]. Importantly, aggregation also influences product stability by interacting with lipids and other biomolecules, potentially accelerating oxidative deterioration. Understanding protein aggregation during thermal and freeze processing is vital for food scientists, technologists, and industry stakeholders aiming to balance safety, quality, and nutritional outcomes. This review critically examines the mechanisms underlying protein aggregation in animal-based foods, highlighting how different processing stresses influence structural transitions. Case-specific insights are provided for major categories including meat, fish, milk, and eggs, as these foods represent the most protein-rich animal-derived commodities consumed worldwide. Furthermore, the review discusses the consequences of aggregation for functional and nutritional properties and outlines technological strategies—such as encapsulation, use of cryoprotectants, and emerging processing technologies—that can mitigate negative effects while enhancing positive ones [6]. An integrating molecular-level insights with practical applications, this review aims to deepen understanding of protein aggregation phenomena, bridge the gap between theory and practice, and inform the development of optimized processing methods.

Such advancements are essential not only for ensuring the quality and nutritional value of animal-derived foods but also for addressing growing consumer demand for safe, minimally processed, and high-quality protein products in a competitive global market.

2. Mechanisms of Protein Aggregation

2.1 General Principles

Proteins in their native state exist in a finely balanced three-dimensional structure, stabilized by a combination of non-covalent interactions—such as hydrogen bonds, hydrophobic associations, and electrostatic forces—and covalent disulfide linkages. This intricate structural equilibrium allows proteins to maintain solubility, functionality, and biological activity. Aggregation is initiated when this balance is disrupted, leading to the partial or complete unfolding of the protein molecule. Once unfolded, normally buried residues such as hydrophobic side chains or sulfhydryl groups become exposed, rendering the protein prone to intermolecular interactions [7]. Aggregation can result in diverse structural outcomes depending on the extent of denaturation, environmental conditions, and processing stresses. Proteins may form amorphous aggregates characterized by random associations, or ordered aggregates such as amyloid-like fibrils, in which intermolecular β-sheets predominate. While amorphous aggregates are common in food systems and often manifest as insoluble precipitates, fibrillar aggregates are more typical in long-term storage or under extreme denaturing conditions. Importantly, aggregation is often irreversible, leading to permanent changes in functional properties such as solubility, gelation, and digestibility, aggregation is influenced by intrinsic protein properties (molecular weight, amino acid composition, isoelectric point) and extrinsic factors including temperature, pH, ionic strength, water activity, and interactions with lipids or carbohydrates [8]. Thermal and freeze processing, two of the most common preservation methods, exert distinct but overlapping stresses that drive protein aggregation through different molecular pathways.

2.2 Thermal-Induced Aggregation

Heat is one of the strongest destabilizing forces for protein structures. As temperature rises, the kinetic energy of protein molecules increases, disrupting hydrogen bonds and weakening van der Waals interactions that stabilize secondary and tertiary conformations. This leads to unfolding, exposure of hydrophobic domains, and increased molecular mobility, all of which favor intermolecular association.

The pathways of heat-induced aggregation are governed by three main mechanisms:

- **Hydrophobic interactions**: Upon unfolding, buried hydrophobic residues become solvent-exposed. These nonpolar regions spontaneously cluster together to minimize contact with water, forming the initial nucleus of an aggregate.
- **Disulfide bond formation**: Heating accelerates thiol–disulfide exchange reactions. Cysteine residues, once exposed, undergo oxidation to form disulfide bridges, stabilizing aggregates in a covalent manner.
- **Electrostatic rearrangement**: At certain pH values close to a protein's isoelectric point, reduced electrostatic repulsion allows proteins to approach more closely and interact, thereby facilitating aggregation.

The extent and nature of thermal aggregation depend strongly on processing conditions. Mild heating may produce reversible aggregates that can dissociate upon cooling, while severe heating typically results in irreversible aggregates and precipitation. The impact also varies across protein systems: myofibrillar proteins in meat form gels upon heating, contributing to desirable textural characteristics, whereas whey proteins in milk aggregate extensively above 70 °C, affecting solubility and digestibility [9], protein concentration, ionic strength, and the presence of sugars or polyphenols can modulate aggregation. For example, salts may shield electrostatic repulsion and accelerate aggregation, while certain carbohydrates exert protective effects by stabilizing protein structure during heating.

2.3 Freeze-Induced Aggregation

Unlike thermal processing, freezing subjects proteins to physical and chemical stresses associated with ice formation and phase separation. As water crystallizes, solutes—including proteins, salts, and sugars—become concentrated in the unfrozen phase, a process known as cryoconcentration. This localized increase in ionic strength and pH changes destabilizes native protein conformations, leading to unfolding and aggregation.

Several important mechanisms underpin freeze-induced aggregation:

- **Ice crystal damage**: Large ice crystals formed during slow freezing can physically disrupt cellular and protein structures, leading to denaturation and aggregation.
- **Cryoconcentration and pH shifts**: As ice excludes solutes, the remaining unfrozen fraction becomes more concentrated, often lowering protein solubility and stability.
- **Interfacial denaturation**: Proteins adsorb at the ice–water interface, where partial unfolding occurs. Repeated freeze–thaw cycles exacerbate this effect, causing cumulative aggregation and precipitation.

The rate of freezing is critical in determining the extent of aggregation. Rapid freezing promotes the formation of smaller ice crystals, which exert less mechanical damage and minimize cryo-concentration, and preserving protein integrity. Conversely, slow freezing favors larger crystals that damage cellular matrices and increase protein destabilization [10]. Repeated freeze–thaw cycles are particularly detrimental in meat and fish products, leading to progressive myosin aggregation, reduced water-holding capacity, and textural deterioration. In dairy systems, freezing destabilizes casein micelles and whey protein interactions, which may manifest as syneresis in frozen–thawed products such as yogurt and ice cream. Both thermal and freeze processing disrupt protein stability, though via different pathways. Heat primarily causes chemical and structural denaturation leading to hydrophobic clustering and disulfide cross-linking, while freezing destabilizes proteins through ice-induced mechanical stress, solute concentration, and interfacial denaturation. In both cases, aggregation alters the solubility, texture, and digestibility of animal-derived foods. Understanding these mechanisms provides a foundation for developing targeted strategies to mitigate detrimental effects while harnessing beneficial aggregation in functional food system

Table 1. Major Proteins in Animal-Based Foods and Their Aggregation Behavior under Thermal and Freeze Processing

Food Source	Major Proteins	Effect of Thermal Processing	Effect of Freeze Processing	Functional Consequences
Meat	Myosin, Actin, Collagen	Denaturation and aggregation → gelation, reduced water-holding	Ice crystal damage, myosin aggregation, lipid oxidation	Tenderness loss, drip loss, toughness
Fish	Myosin, Actin	Low thermal stability; gelation in surimi; excessive heating reduces digestibility	Myosin aggregation, gaping, reduced elasticity	Texture defects, reduced water retention
Milk	Caseins, β-lactoglobulin, α-lactalbumin	Whey protein aggregation, disulfide bonding with κ-casein	Casein micelle destabilization, whey protein aggregation	Syneresis, sedimentation, reduced solubility
Eggs	Ovalbumin, Ovotransferrin, Lysozyme	Heat-induced gelation and foaming; excessive aggregation reduces digestibility	Gelation and water loss in thawed eggs	Rubberiness, reduced foaming capacity

Table 2. Consequences of Protein Aggregation in Animal-Based Foods

Category	Positive Effects	Negative Effects
Texture	Gelation in dairy, meat gels, surimi elasticity	Toughness, dryness, reduced juiciness, gaping in fish
Nutrition	Release of bioactive peptides in controlled denaturation	Reduced digestibility, decreased amino acid bioavailability
Sensory	Desirable firmness and creaminess (yogurt, cheese)	Off-flavors, sulfur notes in eggs, syneresis in dairy
Shelf-Life	Structural stability in some gels	Accelerated lipid oxidation, sedimentation, drip loss

Table 3. Strategies to Control Protein Aggregation during Processing

Strategy	Application	Mechanism of Action	Examples
Processing Optimization	Moderate heat, rapid freezing	Minimizes protein unfolding and ice crystal damage	Sous-vide meats, blast freezing
Cryoprotectants & Stabilizers	Meat, fish, dairy, eggs	Sugars, polyols, proteins replace water, reduce aggregation	Trehalose in fish, sucrose in frozen eggs
Encapsulation & Coatings	Meat, fish, dairy	Biopolymer coatings shield proteins, limit oxidation	Alginate coatings on fish fillets
Novel Technologies	All food systems	Reduce structural stress compared to conventional methods	High-pressure processing, ultrasound, PEF

3. Protein Aggregation in Animal-Based Foods

Protein aggregation in animal-derived foods is strongly influenced by the type of protein present, its structural features, and the processing environment. Different food matrices exhibit distinct aggregation behaviors during thermal and freeze processing, leading to diverse outcomes in terms of texture, digestibility, stability, and sensory perception [11]. The following subsections highlight the aggregation characteristics of meat, fish, milk, and egg proteins, which represent the most widely consumed animal-based foods globally.

3.1 Meat Proteins

Meat proteins are generally categorized into myofibrillar proteins (e.g., myosin, actin, tropomyosin), sarcoplasmic proteins (enzymes and myoglobin), and stromal proteins (collagen and elastin). Among these, myofibrillar proteins are the most functionally significant in terms of gelation, emulsification, and water-holding capacity [12]. During thermal processing, myosin and actin undergo sequential denaturation, typically beginning at 40–50 °C for myosin and around 70 °C for actin. Heat-induced unfolding exposes hydrophobic residues and sulfhydryl groups, leading to aggregation and gel formation. This aggregation is responsible for the firmness and cohesiveness of cooked meat products. However, excessive aggregation reduces water-holding capacity, resulting in dry, tough textures. Heat also affects sarcoplasmic proteins, which may precipitate and contribute to flavor and color changes [13]. In freeze processing, the formation of ice crystals disrupts myofibrillar protein integrity and promotes oxidative modifications. Myosin, in particular, is prone to aggregation during frozen storage due to cryoconcentration of salts and enhanced lipid–protein interactions. These changes reduce solubility and impair functional properties, manifesting as reduced juiciness, increased drip loss, and decreased tenderness upon thawing. Lipid oxidation often accelerates protein cross-linking, further aggravating textural deterioration. Rapid freezing and the use of cryoprotectants (e.g., polyphosphates, sugars) can help mitigate aggregation and preserve meat quality.

3.2 Fish Proteins

Fish proteins share many structural similarities with mammalian proteins but are more thermolabile, reflecting adaptation to aquatic environments with lower body temperatures. Myofibrillar proteins such as myosin and actin are particularly vulnerable to denaturation and aggregation. During freezing, fish proteins are prone to significant quality loss. Myosin aggregation is common, leading to decreased solubility, toughness, and the phenomenon known as "gaping," where muscle fibers separate visibly in thawed fillets. The extent of aggregation depends on species, fat content, and storage conditions. Lean fish such as cod are more sensitive to freeze-induced aggregation compared to fatty fish such as salmon, although lipid oxidation can exacerbate protein damage in the latter. Repeated freeze–thaw cycles worsen these effects, resulting in reduced elasticity and water-holding capacity [14]. In thermal processing, fish proteins denature at relatively lower temperatures compared to mammalian proteins. Controlled thermal aggregation is beneficial in surimi production, where myosin gelation creates the characteristic elastic texture of fish-based gels. However, excessive heating reduces digestibility and leads to textural hardening. The challenge lies in balancing microbial safety and textural optimization while avoiding undesirable aggregation.

3.3 Milk Proteins

Milk contains two main protein classes: caseins (αs-, β-, and κ-casein) and whey proteins (β-lactoglobulin, α-lactalbumin, serum albumin). These proteins exhibit distinct aggregation behaviors during processing. Thermal aggregation primarily involves whey proteins. β-lactoglobulin, which contains a reactive thiol group, begins to denature and aggregate above 70 °C. The aggregation process often involves disulfide bond formation with κ-casein, altering micellar stability. This mechanism has both positive and negative consequences: while aggregation contributes to yogurt and cheese structure, excessive aggregation in fluid milk can cause sedimentation, reduced solubility, and impaired digestibility [15]. During freezing, milk proteins undergo destabilization due to cryoconcentration and ice crystal formation. Casein micelles may destabilize, leading to phase separation and syneresis, particularly in frozen dairy desserts such as ice cream and frozen yogurt. Whey proteins also aggregate at ice–water interfaces, contributing to texture defects and graininess upon thawing. The addition of stabilizers such as polysaccharides or cryoprotectants like lactose and sucrose is a common strategy to minimize freeze-induced aggregation in dairy systems.

3.4 Egg Proteins

Egg proteins, found in both yolk and white, serve as versatile functional ingredients in culinary and industrial applications. Major egg white proteins include ovalbumin, ovotransferrin, ovomucoid, and lysozyme, while yolk proteins are dominated by lipoproteins such as livetin and phosvitin. Thermal aggregation plays a central role in the culinary functionality of eggs. Ovalbumin and ovotransferrin denature and aggregate upon heating, forming the characteristic gels in boiled or scrambled eggs and providing foaming capacity in baked goods. These processes are desirable for texture and structure development. However, excessive or uncontrolled aggregation reduces solubility and may impair digestibility by limiting enzymatic access to peptide bonds. Additionally, overcooking can lead to sulfur-containing off-flavors due to hydrogen sulfide release from aggregated proteins [16]. Freeze-induced aggregation presents significant challenges in egg storage. Freezing destabilizes protein–water interactions, leading to gelation and water loss in thawed eggs. This is especially problematic for egg whites, which may become rubbery and exhibit reduced foaming capacity after thawing. Cryoprotectants such as sucrose, glycerol, or salts are often used in frozen egg products to inhibit aggregation and preserve functional properties. In industrial applications, spray-drying is sometimes favored over freezing to circumvent these quality losses, though it introduces its own thermal aggregation concerns.

4. Consequences of Protein Aggregation

Protein aggregation in animal-based foods has wide-ranging consequences that extend beyond simple structural alterations.

The changes manifest at the molecular, functional, nutritional, and sensory levels, ultimately shaping consumer perception and market value of the final product [17]. Understanding these consequences is crucial for distinguishing between desirable aggregation, which contributes to functionality, and detrimental aggregation, which undermines quality.

4.1 Texture Modification

One of the most visible consequences of protein aggregation is modification of texture. Proteins play a central role in gelation, emulsification, and water-holding capacity, all of which determine the structural and mechanical properties of foods. Controlled aggregation, such as that observed in yogurt, cheese, or surimi, provides firmness, elasticity, and structural integrity. However, uncontrolled aggregation often compromises these properties [18]. In meat products, heat-induced aggregation of myofibrillar proteins can enhance gel formation, contributing to desirable firmness in products like sausages. Yet excessive aggregation reduces water-holding capacity, leading to dry, tough textures and increased cooking loss. In frozen fish, aggregation of myosin and actin manifests as reduced elasticity and gaping, negatively affecting consumer perception. In dairy systems, whey protein aggregation during sterilization can create undesirable sediment, while in eggs, over-aggregation of egg white proteins produces rubbery gels.

4.2 Nutritional Implications

Aggregation also affects the nutritional quality of animal-based foods. Native proteins are generally more accessible to digestive enzymes, while aggregated proteins may limit enzyme penetration, reducing hydrolysis efficiency. Excessive aggregation therefore decreases protein digestibility and bioavailability of essential amino acids. For example, β-lactoglobulin aggregates formed during high-heat milk processing resist enzymatic digestion, reducing nutritional utilization. Similarly, heavily aggregated fish proteins formed during prolonged frozen storage demonstrate reduced digestibility [19]. aggregation may alter the release and availability of bioactive peptides. Controlled denaturation can enhance peptide release with antihypertensive or antioxidant properties, but over-aggregation traps these peptides within insoluble complexes, diminishing their bioactivity. Thus, the balance between beneficial and detrimental effects is delicate and highly dependent on processing intensity.

4.3 Sensory Attributes

Sensory perception of animal-based foods is strongly influenced by aggregation. Aggregated proteins modify texture, appearance, and mouthfeel, which directly affect consumer acceptance. In meats, excessive aggregation manifests as hardness and dryness, while in dairy systems it may cause syneresis (whey separation) that consumers perceive as spoilage. Frozen–thawed fish often displays gaping and loss of translucency, both consequences of aggregation-related structural changes [14]/ Flavor perception can also be indirectly influenced. Aggregated proteins may sequester volatile compounds, reducing flavor release, or interact with lipids, leading to oxidative byproducts that produce rancid flavors. In eggs, overcooked or aggregated proteins release hydrogen sulfide, creating sulfurous off-flavors. These negative sensory impacts underscore the importance of carefully managing aggregation during processing.

4.4 Shelf-Life and Stability

Protein aggregation affects not only immediate quality but also long-term stability and shelf life. Aggregated proteins may interact with lipids, accelerating lipid oxidation through radical transfer mechanisms. This is particularly problematic in high-fat systems such as meat and fish, where protein–lipid interactions lead to rancidity, discoloration, and nutrient loss during storage [6-9]. Freeze-induced aggregation also contributes to drip loss in thawed products, reducing yield and economic value. In dairy, destabilized protein aggregates can lead to sedimentation, phase separation, and reduced shelf stability in sterilized or frozen products. Consequently, controlling aggregation is essential not only for maintaining immediate sensory and nutritional quality but also for preserving product stability throughout distribution and storage.

5. Strategies to Control Protein Aggregation

Given the significant consequences of protein aggregation, food technologists have developed a range of strategies to mitigate its detrimental effects while preserving or enhancing beneficial functionality. These strategies span from conventional processing optimization to the adoption of novel technologies designed to apply less structural stress to proteins.

5.1 Processing Optimization

One of the simplest approaches to minimizing aggregation is optimizing processing conditions. In thermal treatments, moderate heat application (e.g., pasteurization instead of sterilization) can reduce protein denaturation while still ensuring microbial safety. In meat cooking, adopting sous-vide methods at controlled low temperatures minimizes protein aggregation and preserves tenderness [7-11[. In freeze processing, the rate of freezing is critical. Rapid freezing produces smaller ice crystals, reducing mechanical damage and cryoconcentration effects that promote aggregation. Blast freezing or cryogenic freezing with liquid nitrogen are effective techniques for maintaining protein integrity. Additionally, minimizing freeze–thaw cycles during storage and distribution helps reduce cumulative aggregation damage.

5.2 Cryoprotectants and Stabilizers

Cryoprotectants are widely used to protect proteins during freezing. Sugars (sucrose, trehalose), polyols (glycerol, sorbitol), and certain proteins (gelatin, caseinates) stabilize protein structures by replacing water molecules and preventing ice-induced denaturation. In frozen fish and meat, cryoprotectants maintain myosin solubility and water-holding capacity, preserving texture and juiciness [12-14]. Stabilizers such as hydrocolloids (e.g., guar gum, carrageenan) are often used in dairy and egg products to prevent syneresis and phase separation. These compounds provide a protective matrix around proteins, reducing the likelihood of aggregation during both heating and freezing.

5.3 Encapsulation and Coatings

Encapsulation of proteins or protein-rich systems in biopolymer matrices offers another strategy to mitigate aggregation. Encapsulation using alginate, chitosan, or lipid-based carriers shields proteins from thermal and freeze stresses, reducing structural damage. In meat systems, edible coatings with antioxidants and cryoprotectants slow aggregation by preventing oxidative cross-linking. Similarly, coating fish fillets with polysaccharides or proteins before freezing reduces ice crystal damage and aggregation of myofibrillar proteins.

5.4 Novel Processing Technologies

Emerging non-thermal or minimally thermal technologies show promise in controlling aggregation by applying less severe stresses compared to conventional methods.

- **High-pressure processing (HPP):** Applies hydrostatic pressure to inactivate microbes while preserving protein structure. Although some denaturation occurs, pressure-induced aggregation is often reversible and less detrimental than thermal aggregation.
- **Ultrasound processing:** Enhances heat and mass transfer, allowing reduced thermal load. At controlled intensities, ultrasound can improve protein functionality while minimizing irreversible aggregation.
- **Pulsed electric fields (PEF):** Induces microbial inactivation with minimal heating, preserving protein integrity in dairy and liquid egg systems.
- **Microwave-assisted heating:** Provides rapid, uniform heating that reduces localized overheating and aggregation compared to conventional thermal processing.

Protein aggregation represents a critical challenge in the processing of animal-based foods, with consequences ranging from textural changes to nutritional loss and reduced shelf life. However, through strategic control—whether via processing optimization, use of cryoprotectants, encapsulation, or adoption of novel technologies—food scientists can mitigate detrimental aggregation while leveraging its functional benefits. Moving forward, integrating these approaches with molecular-level understanding of aggregation mechanisms will be essential for tailoring strategies to specific food systems and advancing the development of high-quality animal-derived products [12-14].

6. Conclusion and Future Perspectives

Protein aggregation is an inevitable yet complex phenomenon in the thermal and freeze processing of animal-based foods. On one hand, controlled aggregation underpins desirable functional properties such as gelation in meat and fish, foam stability in eggs, and textural structuring in dairy. On the other hand, uncontrolled or excessive aggregation compromises digestibility, reduces amino acid bioavailability, induces syneresis, and contributes to undesirable hardness or dryness, all of which undermine consumer acceptance and nutritional value. The extent of aggregation is highly dependent on protein type, matrix composition, and processing intensity, emphasizing the need for system-specific strategies. Technological advancements are beginning to offer practical solutions. Optimized thermal profiles, rapid freezing techniques, and the use of cryoprotectants have shown significant success in limiting structural damage. More innovative approaches—including encapsulation systems, edible coatings, and non-thermal technologies such as high-pressure processing, ultrasound, and pulsed electric fields—hold promise for minimizing detrimental aggregation while preserving microbial safety and sensory quality, future research should move toward a deeper molecular-level understanding of protein–protein and protein–matrix interactions under processing stresses. The integration of omics approaches and advanced analytical techniques may facilitate the development of predictive models to anticipate aggregation behavior. Additionally, tailored interventions—ranging from protein engineering for greater stability to the design of novel protective additives—will be essential to adapt strategies to different food systems. Such efforts will not only enhance the safety and nutritional quality of animal-derived foods but also align with consumer demand for minimally processed, functional, and high-quality products.

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