

FOOD CHEMISTRY AND ENZYMATIC REACTIONS: UNDERSTANDING THE ROLE OF ENZYMES IN FOOD PROCESSING: A NARRATIVE REVIEW

Narrative Review

Muhammad Usama Aslam^{1*}, Esha Aslam¹, Muhammad Shahbaz^{2*}

¹Department of Food Safety and Quality Management, Bahauddin Zakariya University, Multan 60800, Pakistan.

²Mawarid Food Company, Al Wizarat, Riyadh 12622, Saudi Arabia.

Corresponding Author: Muhammad Usama Aslam, Department of Food Safety and Quality Management, Bahauddin Zakariya University, Multan 60800, Pakistan, ua6965300@gmail.com

Muhammad Shahbaz, Mawarid Food Company, Al Wizarat, Riyadh 12622, Saudi Arabia, shahbazfoodtech@gmail.com

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ABSTRACT

Background: Enzymes are highly specific biological catalysts that play a pivotal role in food processing by influencing texture, flavor, nutritional value, and shelf life. Their ability to function under mild conditions makes them sustainable alternatives to chemical and thermal processing, aligning with global demands for cleaner-label and energy-efficient food technologies.

Objective: This review aims to synthesize current knowledge on the fundamentals, industrial applications, technological innovations, and regulatory frameworks of food enzymes, while critically analyzing their limitations and exploring opportunities for future advancement.

Main Discussion Points: Key themes include enzymatic mechanisms relevant to food systems, with emphasis on carbohydrate-active enzymes, proteases, lipases, oxidoreductases, transglutaminase, and lactase. The review explores industrial applications across bakery, dairy, beverage, meat, and plant-based sectors, highlighting their contributions to product quality and process optimization. Advances in enzyme immobilization, protein engineering, directed evolution, and the exploitation of cold-active and extremophilic enzymes are discussed. Regulatory oversight, analytical methods for enzyme activity, and consumer concerns regarding safety, allergenicity, and labeling are critically examined.

Conclusion: Food enzymes represent a versatile and sustainable toolbox for modern processing industries. While evidence supports their benefits in enhancing quality and sustainability, challenges related to cost, stability, and public acceptance remain. Future directions such as multi-enzyme cascades, AI-driven enzyme design, precision fermentation, and sustainability-focused evaluations offer pathways to expand their safe and effective use in global food systems.

Keywords: Food enzymes; Biocatalysis; Protein engineering; Immobilization; Food processing; Narrative review.

INTRODUCTION

Enzymes are biological catalysts, largely protein in nature, that accelerate specific biochemical reactions essential for life. Their application in food production dates back millennia, with fermentation, cheese making, and malting being classic examples of traditional practices that rely on enzymatic activity (1). Over time, the role of enzymes has expanded from these ancient uses into modern food processing, where they are now employed in purified or formulated forms to enhance product quality, consistency, and safety. They contribute to improved texture, flavor, clarity, nutrient availability, and shelf life, while simultaneously reducing dependence on chemical additives and harsh thermal treatments (2-4). The food enzyme sector has gained significant momentum in recent decades due to biotechnological innovations and enzyme engineering. Advances in recombinant DNA technology, directed evolution, and protein engineering have enabled the design of enzymes with superior stability, specificity, and functionality within complex food matrices (5,6). These developments have made enzymes indispensable tools in optimizing industrial food processes while meeting growing consumer demand for cleaner-label, sustainable, and health-oriented products.

In parallel, regulatory frameworks have been established to ensure the safe use of enzymes in food applications. For instance, the European Food Safety Authority (EFSA) has developed detailed guidance for the submission and evaluation of dossiers related to food enzymes, emphasizing both safety and efficacy before approval (7,8). These regulatory measures aim to safeguard public health while encouraging innovation and transparency within the sector. Despite these advances, gaps remain in understanding long-term safety implications, the potential allergenicity of novel enzymes, and the scalability of engineered biocatalysts in diverse industrial contexts. Given the rapid growth of the food enzyme industry and its critical role in shaping healthier and more sustainable food systems, there is a compelling need to synthesize current knowledge and identify persisting challenges. The objective of this review is to critically examine the applications, advancements, safety considerations, and regulatory dimensions of food enzymes, with the aim of highlighting opportunities for innovation and addressing existing knowledge gaps.

FUNDAMENTALS OF ENZYMATIC REACTIONS IN FOODS

Catalytic principles and kinetics

Enzymes act as biological catalysts by lowering the activation energy of reactions through highly specific active sites. In food systems, their mechanisms encompass hydrolysis by amylases, proteases, and lipases, oxidoreduction by polyphenol oxidase and glucose oxidase, transferase reactions by transglutaminase, isomerization through glucose isomerase, and lyase-mediated conversions. The kinetics of enzymatic reactions in foods are often complex due to heterogeneous matrices, substrate variability, and the presence of inhibitors such as phenolics and salts (1). Models such as first-order, Weibull, and biphasic kinetics have been widely applied to describe thermal inactivation, enabling optimized process design while retaining product quality (2).

Environmental factors

Environmental conditions strongly influence enzyme behavior in food matrices. Temperature, pH, water activity, ionic strength, and cofactors regulate both activity and stability. In complex food environments, macromolecules such as polysaccharides and lipids can shield enzymes or substrates, thereby modifying reaction rates and apparent kinetics (3). Adapted enzymes from extremophiles and psychrophiles expand operational ranges, allowing cold-active enzymes to support low-temperature processing where nutrient and flavor retention is critical. This trend has particular relevance in fruit juice clarification and dairy systems where thermal degradation is undesirable (4).

MAJOR ENZYMES USED IN FOOD PROCESSING (MECHANISMS & APPLICATIONS)

Carbohydrate-active enzymes

Carbohydrate-active enzymes represent one of the most commercially significant groups in food biotechnology. Amylases, including α -, β -, and glucoamylases, are pivotal in starch degradation, improving crumb softness in baking, ensuring efficient malt conversion in

brewing, and enabling syrup production (5). Other enzymes, such as cellulases, hemicellulases, and xylanases, enhance juice yield, dough handling, and beverage clarity. Pectinases remain indispensable in juice processing, increasing yield and reducing viscosity, particularly in fruit-based systems (6).

Proteolytic enzymes

Proteolytic enzymes play essential roles in meat tenderization, cheese ripening, and the generation of bioactive peptides. Plant-derived enzymes like papain and bromelain have been widely studied for their tenderizing effects, while microbial proteases offer greater process flexibility (7). Transglutaminase, in particular, has emerged as a key tool for modifying protein matrices through covalent cross-linking, thereby enhancing texture in meat, dairy, and plant-based alternatives. Its application in the growing plant-based meat sector reflects both technological potential and market-driven innovation (8).

Lipid-modifying enzymes

Lipases and phospholipases are primarily employed in flavor generation and fat modification. They are central to cheese ripening, structured lipid synthesis, and oil degumming. Advances in microbial lipase production have improved specificity, stability, and tolerance to industrial conditions, making them highly suitable for large-scale applications. Lipid-modifying enzymes are also critical in producing tailored fats, such as human milk fat substitutes, which have nutritional relevance for infant formula development (9).

Oxidoreductases and other enzymes

Oxidoreductases include polyphenol oxidase (PPO), which drives enzymatic browning—a major postharvest and processing challenge. Strategies such as thermal inactivation, antioxidant addition, and modified atmospheres remain central to controlling PPO activity (10). Meanwhile, glucose oxidase and catalase extend shelf life by reducing oxygen content, and phytases improve mineral bioavailability in cereal-based products by hydrolyzing phytate complexes (11).

INDUSTRY-SPECIFIC APPLICATIONS

Bakery and cereal products

In bakery systems, enzymes such as amylases, proteases, lipases, and xylanases improve dough handling, crumb softness, and shelf-life stability. The use of enzymes enables formulation with reduced sugar and fat without compromising texture, aligning with public health goals for healthier food production (12).

Dairy and milk processing

Enzymes are integral to dairy processing, where rennet and microbial proteases enable coagulation and cheese ripening. Lactase (β -galactosidase) facilitates the production of lactose-free dairy products, now a rapidly expanding market segment. Lipases contribute to flavor development, while transglutaminase enhances textural properties in yogurts and analogues (13,14).

Beverages and juice processing

Enzymes enhance both quality and efficiency in beverage systems. Pectinases and cellulases improve clarity and yield in juices, while amylases and proteases optimize fermentation processes in brewing. Emerging strategies such as immobilized enzymes and membrane bioreactors are increasingly being adopted for continuous processing, offering advantages in cost and sustainability (15).

Meat, seafood and plant-based proteins

Proteolytic enzymes remain crucial for tenderization in meat systems, while transglutaminase facilitates restructuring and improves the sensory qualities of both conventional and plant-based protein products. Enzyme-assisted extraction has gained attention in maximizing protein recovery from underutilized plant sources, thereby contributing to sustainable protein supply chains (16).

Functional and specialty products

Enzymes are also central to the development of functional foods and nutraceuticals. Hydrolytic processes release bioactive peptides with antihypertensive and antioxidant activities, while glycosylation and lipase-mediated modifications create tailored lipids and carbohydrates with enhanced nutritional profiles (17). Such innovations demonstrate the broader health implications of food enzyme applications.

Enzyme Technology: Immobilization, Engineering and Novel Sources

Immobilization strategies and benefits

Enzyme immobilization enhances operational stability and allows repeated use, making it attractive for industrial processes such as juice clarification and syrup production. Methods include adsorption, covalent binding, entrapment, and cross-linked aggregates. Recent developments in support matrices, including porous frameworks and metal–organic frameworks (MOFs), have improved performance and scalability (12).

Protein engineering and directed evolution

Advances in molecular biology and computational sciences have transformed enzyme development. Techniques such as site-directed mutagenesis and directed evolution have yielded enzymes with improved thermal stability, solvent tolerance, and broader substrate specificity (16). Machine learning and in silico design now accelerate discovery pipelines, enabling more rational and cost-effective development of industrial biocatalysts (17).

Cold-active and extremophilic enzymes

Enzymes from psychrophilic and extremophilic organisms have broadened the operational scope of biocatalysis. Cold-active enzymes enable low-temperature processing, preserving flavors and nutrients while reducing energy demands. Extremozymes with tolerance to extreme pH, salinity, or pressure provide solutions for niche applications in challenging food environments (14).

Analytical Methods and Process Monitoring

Analytical methods remain fundamental to enzyme evaluation and process optimization. Traditional assays such as DNS and pNP remain widely used for quantification, while chromatographic and mass spectrometric methods provide precise monitoring in complex matrices. The integration of process analytical technologies (PAT) allows real-time monitoring and control of enzyme activity. Kinetic modeling, including determination of D- and z-values, supports process optimization in both thermal and non-thermal technologies (15).

Safety, Regulatory & Consumer Considerations

Food enzymes are subject to rigorous evaluation by regulatory bodies such as the European Food Safety Authority (EFSA). Safety assessments involve detailed analysis of the source organism, manufacturing process, toxicology, and allergenic potential (13). Consumer acceptance remains a challenge, particularly regarding labeling and the perception of transglutaminase and genetically engineered enzymes. Nonetheless, market growth reflects both industrial reliance on enzyme technology and consumer demand for products with improved nutritional, functional, and sensory qualities (18).

CRITICAL ANALYSIS AND LIMITATIONS

While the literature on enzyme applications in food processing has yielded promising insights, several critical limitations temper confidence in the findings. Across many studies, small-scale laboratory investigations predominate, often lacking the statistical power and reproducibility of larger-scale trials. Sample sizes are frequently limited to single-batch or pilot applications, undermining the generalizability of reported outcomes. For example, research on enhancing enzyme stability through immobilization techniques often relies on single enzyme–support systems, without randomized comparisons or replication across diverse food matrices (19). Such designs may fail to capture variability introduced by different substrates or processing conditions. Methodological bias further constrains the reliability of conclusions. Studies may select conditions known to favor enzyme performance—for instance, optimizing pH or temperature specifically based on prior knowledge—introducing performance bias and overestimating real-world effectiveness. Blinding is seldom employed, particularly in sensory or texture assessments following enzyme treatment, heightening risk of subjective influence. Similarly, selection bias is evident when research focuses on “model” food systems that are easier to handle—such as clear fruit juices or defined protein substrates—while excluding challenging heterogeneous matrices like high-fiber or emulsified systems where enzyme performance is critical. Publication bias appears to favor positive or significant findings; negative outcomes—such as immobilization methods that fail to improve enzyme retention or that show rapid activity decline—are less often reported (20). This could create an overly optimistic portrayal of enzyme engineering success and obscure important limitations.

Measurement variability also hampers comparisons across studies. Outcomes are quantified using diverse units—e.g., residual activity percentages, reaction rates (U/mL), or sensory scores—making synthesis difficult. The lack of standardized metrics impedes meta-analysis and obscures whether improvements are practically meaningful across scales. Finally, many studies exhibit limited generalizability. Technologies optimized for specific enzymes (e.g., a particular lipase) or supports (e.g., certain MOFs or beads) may not translate effectively to other enzyme–matrix combinations. Research often neglects to test across different food products or under industrial processing conditions such as high shear, variable temperatures, or long durations (21). Regulatory and consumer-related studies are similarly constrained, frequently based on small panels with limited demographic diversity, compromising insights into allergenicity and public perception. In summary, although existing research demonstrates significant technological potential, it is often constrained by narrow scope, methodological limitations, and selective reporting. Future work must emphasize robust study design—incorporating larger, randomized trials, blinded assessments, standardized outcome metrics, and diverse food systems—to fully evaluate the feasibility and safety of enzymatic applications in food processing.

IMPLICATIONS AND FUTURE DIRECTIONS

This review underscores promising trajectories in enzymatic food processing that bear relevance to clinical nutrition, regulatory policy, and sustainable food innovation. In terms of clinical practice, advances in multi-enzyme cascades and co-immobilized systems suggest new paths for producing prebiotic glycans and tailored bioactive compounds that could enhance gastrointestinal health and metabolic regulation—as explored in recent studies on starch-to-prebiotic conversions (21). Enzymes engineered via AI-guided design approaches may soon enable precision manufacture of low-allergenicity ingredients, providing novel options for patients with food sensitivities or metabolic disorders (22,23). From a policy and guideline perspective, the integration of precision fermentation-derived enzymes and alternative-protein ingredients demands updated regulatory frameworks. Current guidance, like that from EFSA, may need revision to accommodate recombinant, AI-designed, or fermentation-derived enzyme preparations characterized by synthetic biology platforms—a shift already highlighted in the expanding application of *de novo* enzyme design tools (24,25). Nevertheless, critical questions remain unanswered. Longitudinal studies are needed to assess the safety, allergenicity, and nutritional outcomes of foods processed with AI-guided or multi-enzyme systems. The ecological benefits of cold-active enzymes and their actual energy savings across full-scale food systems require validation through robust, comparative life-cycle assessments (LCA)—currently underexplored in enzyme intervention literature (26). Further, the scalability and economic feasibility of co-immobilized or cascade systems remain undercharacterized in industrial contexts.

Future research should adopt more rigorous methodological designs. Randomized pilot studies comparing enzyme-engineered foods with conventional products could evaluate clinical endpoints—such as glycemic response or digestive tolerance—in representative human cohorts. In policy-relevant work, structured stakeholder consultations involving food technologists, regulators, and consumer advocacy groups are essential to shape adaptive guidelines reflecting emerging technologies. From a research standpoint, standardized LCA protocols should be embedded within experimental studies testing cold-active enzyme applications to deliver quantitative sustainability metrics. For enzyme engineering, hybrid workflows—combining AI prediction with iterative directed evolution and cell-free validation—can enhance both discovery speed and practical performance (25,26). Finally, integrating multi-site, cross-matrix trials with immobilized enzyme cascades across diverse food types will clarify transferability and economic viability. In summary, this review lays out a roadmap linking enzymatic innovations to health, sustainability, and regulation. It affirms the need for translational research, stakeholder-aligned policymaking, and rigorous trial designs that will solidify enzyme technologies’ real-world impact.

CONCLUSION

Enzymes have emerged as a powerful and sustainable toolkit in food processing, offering enhanced efficiency, product quality, and nutritional value while reducing reliance on chemical or energy-intensive methods. This review highlights key advances in immobilization, protein engineering, and the discovery of novel extremophilic and cold-active biocatalysts that broaden the scope of industrial applications. The existing evidence demonstrates considerable promise, though much of it remains limited to laboratory or pilot-scale contexts, underscoring the need for cautious interpretation and further validation. Strengthened by established regulatory frameworks such as EFSA guidance, enzyme technologies are generally considered safe; however, the rapid introduction of directed-evolution variants, GMO-derived enzymes, and precision fermentation products calls for updated oversight and transparent labeling to ensure consumer trust. For clinicians and researchers, the practical recommendation lies in aligning enzymatic innovations with health-

oriented product development, sustainability goals, and patient-centered nutrition strategies. At the same time, more rigorous, large-scale studies are required to evaluate long-term safety, consumer acceptance, and environmental impact, ensuring that enzymes reach their full potential as reliable, evidence-based tools in shaping the future of food systems.

AUTHOR CONTRIBUTION

Author	Contribution
Muhammad Usama Aslam*	Substantial Contribution to study design, analysis, acquisition of Data Manuscript Writing Has given Final Approval of the version to be published
Esha Aslam	Substantial Contribution to study design, acquisition and interpretation of Data Critical Review and Manuscript Writing Has given Final Approval of the version to be published
Muhammad Shahbaz*	Substantial Contribution to acquisition and interpretation of Data Has given Final Approval of the version to be published

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