International Journal of Biotechnology and Microbiology
www.biotechnologyjournals.com
Online ISSN: 2664-7680, Print ISSN: 2664-7672
Received: 19-08-2024, Accepted: 18-09-2024, Published: 03-10-2024
Volume 6, Issue 4, 2024, Page No. 1-5

The impact of biocatalyst in food processing industry

Prerana Belekar*, Sneha Desai, Srushti Wagh, Aakash Pawar
Department of Biological Sciences, School of Science, Sandip University, Nashik, Maharashtra, India

Abstract
Enzymes have long been integral to food manufacturing, significantly influencing food processing across various applications. The primary classes of enzymes-ligases, hydrolases, oxidoreductases, transferases, lyases, and isomerasesare derived from diverse sources and are instrumental in a wide array of food processing operations. Research in enzyme technology focuses on the separation, purification, and characterization of these biocatalysts, which facilitate biochemical reactions without undergoing consumptive changes themselves. Microorganisms such as fungi, yeasts, and bacteria are notable producers of enzymes with specialized functions. Enzymes not only serve biodegradable roles but also find extensive use in industrial applications, including the production of wine, beer, and whisky, where they operate effectively *in vitro*. The involvement of enzymes in biochemical processes spans plants, animals, microorganisms, and humans, underscoring their universal significance. The efficient absorption of enzymes during food processing has prompted extensive research aimed at addressing global food security challenges. Advances in biotechnology have introduced sophisticated screening methodologies for discovering novel food enzymes, enhancing food processing, reducing production costs, and tackling waste management, food safety, and packaging issues. This chapter provides a comprehensive overview of enzyme technology within the food industry, highlighting recent advancements and discussing the diverse sources and applications of enzymes in food processing.

Keywords: Enzymes, food manufacturing, microorganisms, enzyme technology

Introduction

1. Enzymes

Enzymes, recognized as biological catalysts or biocatalysts, have been integral to food processing since their early application (Koshland Jr, 1958) [25]. The term "enzyme" derives from the Greek words en (within) and some (yeast), a nomenclature introduced by Wilhelm Kühne in 1878 about yeast's ability to ferment carbohydrates into alcohol (Robinson *et al*, 2015) [36]. Enzymes, present in all living organisms—microbes, animals, and plants—are pivotal in numerous biochemical processes including the synthesis of sweeteners and the modification of antibiotics (Sharma *et al*, 2019) [38]. Microbial enzymes are favored in food processing due to their advantages of ease, cost-effectiveness, and consistent production (Ahmad *et al*, 2018) [4]. The utilization of enzymes in commercial food processing is expanding, with projections indicating continued growth in this sector (Mohammed Kuddus, 2019) [26]. These enzymes can be extracted from cells and serve as catalysts for various significant industrial applications.Enzymes, as proteins, facilitate biochemical reactions crucial for vital processes such as metabolism, respiration, and digestion. Historically, their catalytic properties have been harnessed in food preparation, notably in bread, wine, beer, and cheese production (Robinson *et al*, 2015) [36]. Typically, enzymes are extracted from edible plants and animal tissues (Csuk R *et al*, 1991) [15]. The Industrial Revolution further accelerated the development of the enzyme industry, particularly in the dairy, beverage, baking, and brewing sectors (Dr. R. Prakash Chandran, 2019) [17]. Enzymes act as catalysts within living cells by lowering the activation energy required for reactions, thereby increasing reaction rates without undergoing permanent chemical changes (Mohammed Kuddus 2019) [26]. Due to their biological origin, enzymes are also referred to as biocatalysts. Structurally, enzymes consist of one or more polypeptide chains composed of amino acids (Dr. R. Prakash Chandran 2019) [17]. Each enzyme's unique amino acid sequence dictates its folding pattern and resultant three-dimensional structure (Ramli *et al*, 2021) [33]. Enzymes demonstrate high substrate specificity, catalyzing particular reactions or narrow classes of reactions (Jackson *et al*, 2009) [21]**.** The active site of an enzyme, a specific region on its surface, binds to the substrate molecules, akin to a lock and key mechanism. This interaction facilitates the catalytic process, after which the enzyme releases the product(s) and remains unaltered, allowing it to catalyze subsequent reactions (Dong Tianfei 2020) [16]**.** Thus, enzymes can be reused multiple times. Enzymes regulate nearly all metabolic processes in the body, encompassing catabolic pathways (the breakdown of complex molecules) and anabolic pathways (the synthesis of complex molecules) (Ramli *et al*., 2021) [33].

2. Enzyme technology

Enzyme technology is now a crucial factor in the food industry, transforming different areas of food manufacturing, processing, and storage (Yang *et al*, 2023) [45]. Using enzymes in food technology has a major benefit as they improve product quality, boost efficiency, cut costs, and enhance sustainability in manufacturing processes (van Beilen *et al*, 2002) [41]. The growth of the global industrial enzymes market is evident due to the rising need for high-quality processed foods and beverages, with enzymes playing a significant role in this sector (Talens-Perales *et al*, 2015) [40]. Moreover, hydrolytic microbial enzymes like carbohydrases, proteases, and lipases play a vital role in improving yields and enhancing food quality, meeting consumer demand for healthier alternatives (Guerrand *et al*, 2017; van Beilen *et al*, 2002) [20, 41]. The enzyme technology has been widely embraced by the food industry to tackle different issues like increasing production, boosting nutritional content, and decreasing processing time(Al-Maqtari *et al*, 2019) [7]. In addition, the food industry can now utilize microbial enzymes for a wider range of applications, such as baking, brewing, dairy, and meat processing, thanks to advancements in enzyme production technology, ensuring that food products meet quality standards (Singh *et al*, 2019) [32, 39].

3. Food processing

Before formally identifying enzymes as biological catalysts, their utilization in food processing was limited. The earliest application of enzymes in food technology emerged with the practice of food fermentation, which also initiated their use in cheese production (Bilal *et al*, 2020) [11]. Enzymatic functions in food processing and preservation have been instrumental in advancing human nutrition and food technology (Ravindran *et al*, 2018) [35]. Enzymes have made significant contributions across various sectors within the food industry, including baking, cheese production, dairy processing, milling, cereal technology, vegetable and juice processing, oil and fat processing, and wine production(Abdullahi *et al*, 2021) [2]. Microorganisms are the primary and most ancient source of enzymes for food processing, with additional enzymes obtained from plant and animal tissues. Advances in science and biotechnology now allow for the tailored commercial production of enzymes, expanding their applications (Raveendran *et al*, 2018) [34]. Historically, microorganisms-such as bacteria, yeast, and fungi—and their enzymes have been integral to food preparation, enhancing flavor and texture, and providing significant economic benefits to the food industry **[].** The advantages of microbial enzymes include their ease of production, cost-effectiveness, and reliability (Ahmad *et al*, 2018) [4]. Enzymes such as amylase, protease, lipase, oxidoreductase, and isomerase are widely employed in various sectors, including wine and beverage production, dairy processing, starch conversion, fermentation, spice processing, and baked goods (Yang *et al*., 2023) [45]. Industrial enzyme production typically involves microbial strains optimized through mutagenesis and selection processes to maximize enzyme yields while minimizing byproducts (Greenwood, J. C. 2013) [19]. These microorganisms are often genetically engineered to produce high quantities of specific enzymes. The production of industrial enzymes involves submerged or solid-state fermentation of microorganisms, followed by cell lysis and filtration (Motta *et al*, 2023) [28]. The resultant crude enzyme extract undergoes purification through techniques such as centrifugation and precipitation (Al-Maqtari *et al*., 2019) [7]. Enzyme isolation and purification further involve separation methodologies, including centrifugation, flocculation, filtration, aqueous two-phase systems, and chromatographic and crystallization techniques (Christian, 2019).

4. Different sources of enzymes used in food processing

Food processing enzymes encompass both endogenous enzymes present within the food matrix and exogenous enzymes introduced to facilitate processing (Ozatay *et al*, 2020) [29]. These enzymes originate from a range of sources including microorganisms, plant tissues, and animal tissues and organs (A. M. Grumezescu *et al*, 2019) [1]. Among these, microorganisms are the most significant and promising source of industrial enzymes due to their historical precedence in enzyme discovery and their ongoing economic, practical, and yield-related advantages (Bilal *et al*., 2020) [11]. The extensive application of microbial enzymes addresses the high demand in various sectors such as baking, dairy, beverages, confectionery, fruit and vegetable processing, and meat processing. Microorganisms, including genera such as Bacillus, Aspergillus, Kluyveromyces, Trichoderma, Rhizopus, and Saccharomyces, produce a diverse array of enzymes utilized across the food industry (Ravindran *et al*., 2018) [25]. These microbial sources are favored for their economic efficiency, effectiveness, adaptability, and higher yield in industrial processes (Rachana Singh *et al*, 2019) [32, 39]. Conversely, plant and animal-derived enzymes, found in foods such as papaya, pineapple, bananas, figs, and bee pollen, are typically present in lower concentrations(A. M. Grumezescu *et al*, 2019) [1]. These sources include proteinases, amylases, lipases, and maltases, as well as specific enzymes like papain and bromelain (Ahlawat *et al*, 2018) [3]. While these enzymes can complement human digestive enzyme production, their extraction and purification pose significant challenges due to their limited abundance, potential contamination, and complex extraction processes (Ahmad *et al*., 2018) [4]. In contrast, microbial-derived enzymes offer enhanced safety profiles compared to plant and animal sources, as they are less likely to contain contaminants such as viruses or toxins (Raveendran *et al*., 2018) [34]. Additionally, purified microbial enzymes generally exhibit superior catalytic properties compared to their free enzyme counterparts. Free enzymes often demonstrate limitations including reduced stability, slower catalytic activity under certain conditions, and susceptibility to inactivation at higher substrate or product concentrations (Akanbi *et al*, 2020) [6]. Therefore, purified microbial enzymes provide improved performance and reliability in industrial applications.

Table 1: Enzyme subtypes found in food and their classifications based on catalyzed chemical reactions

Classes	Functions	Enzyme subclasses in food	References
Oxidoreductase	They facilitate reduction and oxidation processes.	Peroxidase, polyphenol oxidase, catalase, and glucose oxidase	(Binod *et al*, 2019) [12]
Transferase	These catalyze the transfer of a chemical group from one molecule to another.	Transglutaminase	(Kirk O *et al*, 2002) [24]
Hydrolase	They stimulate the breakdown of a bond.	Amylase, lactase, lipase, invertase, lysozyme and protease	(Schäfer T *et al*, 2006) [37]
Lyase	Without requiring any additional catalyst, these facilitate the dissolution of bonds.	Pectin lyase and pectate lyase	(Binod *et al*, 2019) [12]
Isomerase	They aid in the compound's isomerization by catalysis.	Glucose isomerase	(Binod *et al*, 2019) [12]
Ligase	The joining of two molecules is catalyzed by ligases.	_	(Binod *et al*, 2019) [12]

Table 2: The following is a list of some common enzymes used in the food industry

Enzyme	Functions	Application	Reference
Lactase	Preserves dairy products forlactose-intolerant individuals	Dairy products	(Van der Maarel MJ *et al*, 2002) [42]
Glucose oxidase	Converts glucose to gluconic acid to prevent Maillard reaction; used in dehydration processes	Dehydratedfood products	(Aiyer PV 2005) [5]
Cellulase	Produces fermentable feedstock from cellulose waste for ethanol or single-cell protein synthesis	Ethanol production, single-cell proteins	(Van der Maarel MJ *et al*, 2002;James J *et al*, 1996) [22, 42]
α-amylase	Helps dissolve carbohydrates in barley and other grains used in brewing	Brewing	(Jooyendeh H *et al*, 2009) [23]
Beta-glucanase	Breaks down glucans found in malt and other materials	Brewing, malt processing	(Mohd Azmi *et al*, 2023) [27]
Lipase	Accelerates the ripening of cheese by acting on curds or butterfat	Cheese production	(Van der Maarel MJ *et al*, 2002), [42]
Papain	Tenderizes meat	Meat tenderizing	(Fernandes *et al*, 2010) [18]
Chymosin	Aids in curdling milk by dissolving kappa-caseins during cheese-making	Cheese-making	(Chen *et al*, 2021) [7]
Microbial proteases	Used to make texturized proteins, meat extracts, fish meals, and other products	Texturized proteins, meat extracts, fish meals	(Fernandes *et al*, 2010) [18]
Pectinase	Eases fruit pulp extraction, improves juice clarity and filtration	Fruit juice extraction and clarification	(Pasha KM *et al*, 2013) [30]

5. Role of enzymes in the food industry

In recent decades, enzymes have become increasingly integral across various industries, particularly in food processing (Alvarez *et al*, 2020) [8]. Historically sourced from plants, animals, and microorganisms, enzymes now play a pivotal role in the production of dairy, meat, cereals, confectioneries, and beverages, enhancing quality, reducing costs, and shortening processing times (Binod *et al*., 2019; Zhang *et al*., 2021) [12, 46]. This review focuses on the application of hydrolytic enzymes, primarily proteases, amylases, and cellulases, which dominate industrial processes in dairy, starch, textile, and baking industries (Ozatay, 2020) [29]. Microbial sources, due to their high yield and cost-effectiveness, offer significant promise, further enhanced by recombinant DNA technology and process engineering (Atalah *et al*, 2019) [9]. Historically, biological agents have been used in food processing since 6000 BCE for products like cheese, wine, and bread (Al-Maqtari *et al*., 2019; Motta *et al*., 2023) [7, 2]. Enzymes are increasingly preferred over chemical additives for their natural origins and effectiveness in producing fiber-rich bread, glucose syrup, gluten-free foods, and natural sweeteners (Bas *et al*, 2019) [10]. Current advancements include genetic modification, enzyme isolation from unique environments, protein engineering, directed evolution, and high-throughput screening**.** Over 90% of industrial enzymes are derived from microorganisms, underscoring their commercial viability (Zhang *et al*., 2021) [46].

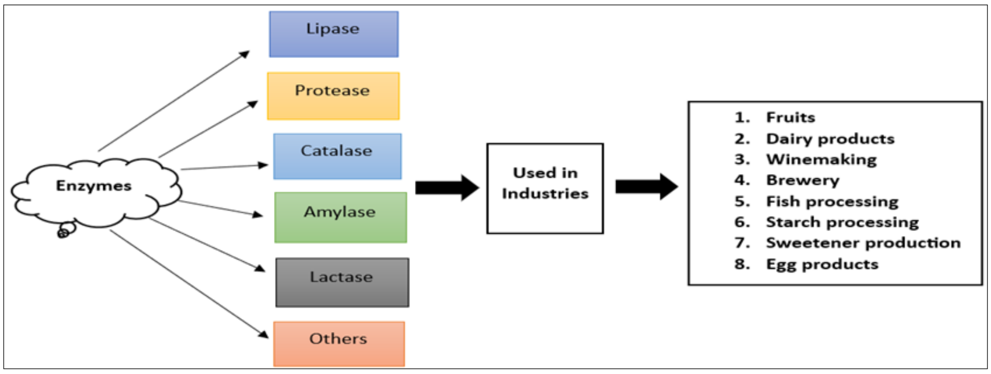

Fig 1: Role of enzymes in the food industry [75]

6. Enzyme technology showing recent developments

Recent advancements have demonstrated that nanotechnology, though still emerging, can be utilized to produce food processing enzymes, presenting both challenges and opportunities (Cacicedo *et al*, 2019) [13]. Concurrently, biotechnological progress has significantly expanded the use and market for industrial enzymes (Holban *et al*, 2019). Microbial enzymes are particularly notable due to their diverse catalytic functions, ease of synthesis, and genetic modifiability, leading to their widespread application in biotechnology (Binod *et al*., 2019; Zhang *et al*., 2021) [12, 46]. Enzymes, as natural biocatalysts, have historically been integral to food production and have been economically produced since the mid-20th century (Wiltschi *et al*, 2020) [43]. They offer substantial benefits to the food and beverage industries, including enhanced product quality, extended shelf life, nutrient release, and improved operational efficiencies.To maximize their industrial impact, enzymes must be highly competitive and efficient, necessitating the development of precisely engineered biological catalysts that are durable, stable, and selective under process conditions (Xavier *et al* 2018) [44].

Enzyme engineering focuses on optimizing these characteristics by modifying the enzyme's amino acid sequence. This process involves generating enzyme variant libraries, assessing desired properties in high-throughput formats, and designing genes using bioinformatics (Quaglia *et al*, 2017) [31]. Structural and sequencing data are analyzed and stored in databases, enabling the application of machine learning and artificial intelligence to extract actionable insights from this data (Singh *et al*., 2019) [32, 39].

Conclusion

The primary sources of enzymes utilized in food processing include microorganisms, plant and animal tissues, and organs. Enzymes are very efficient and quickly reduce complicated heterogeneous materials to simpler ones. Enzymes do not get consumed by biological reactions, although they can start them and/or speed up their rate. Advances in microbiology, genetic engineering, recombinant DNA technologies, and food processing give up new possibilities for use in nearly every aspect of food production. They are now used in food for purposes other than flavor development and catabolism. Industrial methods yield enzymes with specific and desirable features. In recent years, enzymes with high specificity, low-calorie content, and appealing culinary applications have been created. They are useful in enhancing quality, stability, shelf life, and sensory attributes in addition to serving as a processing aid. Additionally, they perform unique tasks in turning food waste into valuable and practical resources. A vast array of food processing applications involves their usages, such as baking, cheese production, dairy processing, milling, cereal technology, juice and beverage processing, vegetable processing, oils and fats processing, winemaking, syrup, sweetener, and chocolate production; infant foods; egg products; soft drinks; candies; flavor development; and meat tenderization, to name a few. Extremozymes have been discovered, therefore processing conditions are no longer a barrier to their use in food.

Acknowledgment

The author extends sincere gratitude toDr. Nissar Reshi Associate Dean, School of Science, Sandip University, Nashik, and Dr. Sandip Wagh, Head, of the Department of Biological Science at the School of Science, Sandip University, Nashik, Maharashtra, India, for their invaluable guidance and support throughout the preparation of this review article. Their expertise and insights were instrumental in shaping this work.

Disclosure statement

No potential conflict of interest was reported by the author(s).

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