

Chapter 1 : Applications of enzymes in food processing | Horticulture International

Benefits of enzymes in food production processes. Enzymes are natural proteins that act as catalysts for biochemical reactions. Enzymes are processing aids: the enzyme itself is not functional in the final product.

Print Introduction Enzymes are protein molecules that are present in all living things. They speed up and target chemical reactions, in many cases increasing the rate of reaction millions of times. For example, they aid digestion, metabolise and eliminate waste in humans and animals, and play a crucial role in muscle contraction. Enzymes have been used unknowingly in food production, e. They can be obtained by extraction from plants or animals or by fermentation from micro-organisms. They are usually purified but may contain varying traces of the other naturally occurring constituents of these three sources. They are normally added to perform a technological function in the manufacture, processing, preparation and treatment of foods. Examples include enzymes used to break down the structure of fruit so that manufacturers can extract more juice, or to convert starch into sugar in alcohol production. Historically enzymes are considered to be non-toxic and not of safety concern for consumers since they are naturally present in ingredients used to make food. However, food enzymes produced industrially by extraction from plant and animal tissues, or by fermentation of microorganisms, are assessed for safety. In , the CEF Panel published guidance to applicants on the data needed for safety evaluation of food enzymes. This explains the format of a formal application for the safety assessment of a food enzyme, the administrative and technical data required, and the range of toxicological tests generally required. Among the data required, applicants must submit information on the identity of the source materials, the manufacturing process, an assessment of dietary exposure and toxicological data except in those cases outlined above. In , EFSA published a supporting explanatory note that further clarifies and provides practical examples of data requirements. Information, including the microbial strain used for enzyme production and the intended use, should be sent to: Main work in progress CEF Panel and its working groups Role EFSA adopts scientific opinions and provides scientific advice for risk managers on the safety of enzymes used in food and feed. The Authority has already substantial experience in evaluating the safety and efficacy of enzymes used in feed materials. However this activity falls under a different legal framework and requires the consideration of different scientific information. Since the entry into force of new European legislation in , the Authority has also taken on an important role in providing independent scientific advice to support the authorisation process for food enzymes. EFSA has two main functions in relation to food enzymes: Evaluating all enzymes currently marketed or intended to be marketed in the European Union EU during a submission period defined by EU legislation. Assessing applications for the authorisation of new enzymes after an EU list of approved enzymes has been established. EU framework Previously, food enzymes other than those used as food additives were not regulated at EU level or were regulated as processing aids under the legislation of Member States. Only France and Denmark have required safety evaluations for enzymes used as processing aids before they could be used in food production. Due to differences between national rules on the assessment and authorisation of food enzymes, new EU framework legislation on food enzymes was adopted in This legislation has the aim eventually to establish an EU list of enzymes. Until such a list is established national rules on the marketing and use of food enzymes and food produced with food enzymes will continue to apply in EU countries. A food enzyme will be included in the EU list if it does not pose a health concern to the consumer; there is a technological need for its use; and its use does not mislead consumers. This regulation establishes a submission period for existing food enzymes starting from 11 September These provisions apply only to enzymes with the same catalytic class, manufactured substantially by the same process and originating from the same source, and for enzymes: Also, microbial cultures traditionally used in the production of food cheese, wine , which may incidentally produce enzymes but are not specifically used to produce them, are not considered food enzymes. More information on food enzyme legislation and the submission of food enzyme applications:

Chapter 2 : Enzymes in Food Processing: A Condensed Overview on Strategies for Better Biocatalysts

Enzymes play a vital role in the food industry to aid nutritional value in food & beverages products. Advanced Enzymes ensures that their Enzymes for Food Processing improve food quality through high nutritional value, flavor, and texture.

This is an open access article distributed under the Creative Commons Attribution License , which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. Abstract Food and feed is possibly the area where processing anchored in biological agents has the deepest roots. Despite this, process improvement or design and implementation of novel approaches has been consistently performed, and more so in recent years, where significant advances in enzyme engineering and biocatalyst design have fastened the pace of such developments. This paper aims to provide an updated and succinct overview on the applications of enzymes in the food sector, and of progresses made, namely, within the scope of tapping for more efficient biocatalysts, through screening, structural modification, and immobilization of enzymes. Targeted improvements aim at enzymes with enhanced thermal and operational stability, improved specific activity, modification of pH-activity profiles, and increased product specificity, among others. This has been mostly achieved through protein engineering and enzyme immobilization, along with improvements in screening. The latter has been considerably improved due to the implementation of high-throughput techniques, and due to developments in protein expression and microbial cell culture. Expanding screening to relatively unexplored environments marine, temperature extreme environments has also contributed to the identification and development of more efficient biocatalysts. Technological aspects are considered, but economic aspects are also briefly addressed. Introduction Food processing through the use of biological agents is historically a well-established approach. The earliest applications go back to 6, BC or earlier, with the brewing of beer, bread baking, and cheese and wine making, whereas the first purposeful microbial oxidation dates from 2, BC, with vinegar production [1] [2]. Coming to modern days, in the late XIX, century Christian Hansen reported the use of rennet a mixture of chymosin and pepsin for cheese making, and production of bacterial amylases was started at Takamine latter to become part of Genencor. Pectinases were used for juice clarification in the s, and for a short period during World War II, invertase was also used for the production of invert sugar syrup in a process that pioneered the use of immobilized enzymes in the sugar industry [1]. Still, the large-scale application of enzymes only became really established in the s, when the traditional acid hydrolysis of starch was replaced by an approach based in the use of amylases and amyloglucosidases glucoamylases , a cocktail that some years latter would include glucose xylose isomerase [1 , 2 , 4 , 5]. From then on, the trend for the design and implementation of processes and production of goods anchored in the use of enzymes has steadily increased. Part of this market is ascribed to enzymes used in large-scale applications, among them are those used in food and feed applications [10]. These include enzymes used in baking, beverages and brewing, dairy, dietary supplements, as well as fats and oils, and they have typically been dominating one, only bested by the segment assigned to technical enzymes [11 , 12]. The latter includes enzymes in the detergent, personal care, leather, textile and pulp, and paper industries [10 , 13]. A relatively large number of companies are involved in enzyme manufacture, but major players are located in Europe, USA and Japan. The pace of development in emerging markets is suggestive that companies from India and China can join this restricted party in a very near future [15] [17]. Tapping for Improved Biocatalysts 2. General Aspects and the Screening Approach Roughly all classes of enzymes have an application within the food and feed area, but hydrolases are possibly the prevalent one. Representative examples of the enzymes and their role in food and feed processing are given in Table 1. The widespread use of enzymes for food and feed processing is easily understandable, given their unsurpassed specificity, ability to operate under mild conditions of pH, temperature and pressure while displaying high activity and turnover numbers, and high biodegradability. Enzymes are furthermore generally considered a natural product [18 , 19]. The whole contributes for developing sustainable and environmentally friendly processes, since there is a low amount of by-products, hence reducing the need for complex downstream process operations, and the energy requirements are relatively low. Life-cycle assessment LCA has confirmed, that within the range of

given practical case studies, including food and feed processing, the implementation of enzyme-based technology has a positive impact on the environment [3]. LCA is a methodology used to compare the environmental impact of alternative production technologies while providing the same user benefits [20]. An overview of enzymes used in food and feed processing adapted from [10 , 12 , 13 , 68]. Some of the broad generalizations on the limitations of enzymes for application as biocatalysts in commercial scale, namely, their high cost, low productivity and stability, and narrow range of substrates, have been rebutted [21 , 22]. Aiming at improving the performance of biocatalysts for food and feed applications, particular care has been given to increasing thermal stability, enhancing the range of pH with catalytic activity and decreasing metal ions requirements, as well as to overcoming the susceptibility to typical inhibitory molecules. Some examples of strategies taken to improve the performance of relevant enzymes for food and feed are given in Table 2. Along with these different strategies focused on the enzyme molecule namely, protein engineering, enzyme immobilization , the developments in recombinant DNA technology that occurred in the s also had a huge impact on the application of enzymes in food and feed. By allowing gene cloning in microorganisms compatible with industrial requirements, this methodology enabled cost-feasible production of enzymes that were naturally produced in conditions that prevented large-scale application namely, enzymes from plant or animal cells, such as transglutaminase or even slow-growing microorganisms. When successfully implemented, the undertaken approaches allow: Methodologies with a high level of parallelization, anchored in computer-monitored microtiter plates equipped with optic fibers and temperature control have also been developed. These provide high-throughput capability for a speedy and detailed characterization of the performance of enzymes [25]. Particular focus was given to the prediction of the long-term stability of enzymes under moderate conditions using short-term runs up to 3 hours. Some examples of strategies undertaken to improve the performance of enzymes with applications in food and feed. One of the methodologies to obtain improved biocatalyst relies on in-vitro modifications, which will be addressed latter in this paper; another approach relies on screening efforts, which has been consistently undertaken, as summarized recently [26 – 31]. Some focus is given to extremophiles, particularly thermophiles, since operation at high temperatures roughly above 45 – minimizes the risk of microbial contamination, a particularly delicate matter under continuous operation. Furthermore, the extension of some reactions in relevant food applications is favored at relatively high temperatures namely, isomerization of glucose to fructose , although care should be taken to avoid an operational environment that may lead by-product formation namely, Maillard reactions. Examples of screened enzymes include the isolation of amylases, with some of them being calcium independent [32 – 38]; amylopullulanases [39]; fructosyltransferases [40]; glucoamylases [41]; glucose xylose isomerases [42 , 43]; glucosidases [44 , 45]; inulinases [46 – 49]; levansucrases [50]; pullulanases [51 , 52]; and xylanases [53 , 54]. Other examples of these enzymes, with some of which able to retain stability under temperatures of or higher, were reviewed by Gomes and Steiner [55]. The majority of enzymes used in food and feed processing is of terrestrial microbial origin, and screening-efforts for isolation of promising enzyme-producing strains have accordingly been performed in such background [3 , 5 , 56]. From some years now, marine environment has also been tapped as a source for useful enzymes from either microbial or higher organisms origin [57 – 60]. Other examples of useful enzymes for food and feed, but isolated from higher organisms [59 , 69], are given in Table 3. Some of these enzymes are actually psychrophiles, hence performing best at low temperatures [30]. Examples of enzymes isolated from various marine higher organisms with potential of application in food and feed adapted from [68 , 69]. Operation at low temperatures is also welcome since it also reduces the risk of microbial contamination, enables some processes to be carried out with minimal deterioration of the raw material. These include protein processing, such as cheese maturing and milk coagulation with proteases [59 , 80]; milk processing with lactase for lactose-free milk [81 – 83]; clarification of fruit juices with pectinases to produce clear juice [84]; or production of oligosaccharides [85]. Since extremophiles are often difficult to grow under typical laboratory conditions if not nonculturable at all, different approaches have been developed in order to assess the potential of enzymes from such microorganisms. One approach relies on the generation and screening of target genes from DNA libraries, which can be obtained from mixed microbial population

from environmental samples. Recombinant microorganisms can then be obtained using mesophiles as hosts where the genes of interest from extremophiles have been expressed [86]. In order to screen the huge number of DNA-libraries typically generated for the intended property, high-throughput methods have been implemented [87]. These methods are also widely used when protein engineering is carried out. This will be addressed in the following section. Several enzymes namely, -amylases; pullulanases currently used in food processing, namely, in starch hydrolysis, are actually produced by recombinant microorganisms. Despite some complexity in the implementation of their use in large-scale applications, partly resulting from lack of uniformity in the US and EU legislation, quite a few enzyme preparations have been accepted for industrial use [88 , 89]. Beyond Screening Taking advantage of the knowledge gathered on molecular biology, high-throughput processing, and computer-assisted design of proteins, in-vitro improvement of biocatalysts have been consistently implemented [90 – 93]. Some of the research efforts in this area has focused on the biochemical and molecular mechanisms underlying the stability of enzymes from extremophiles [31 , 94 – 96]. Such knowledge is also particularly useful for protein engineering of known enzymes, aiming at enhancing stability without compromising catalytic activity [97]. Enhancing the stability of enzymes is of paramount importance when implementation of industrial processes is foreseen, since it allows for reducing the amount of enzyme used in the process. Given that thermostability is determined by a series of short- and long-range interactions, it can be improved by several substitutions of amino acids in a single mutant, where the combination of each individual effect is usually roughly additive [98]. The targeted improvements have not been restricted to thermostability, but they have also addressed other features, such as broadening the range of pH where the enzyme is active, or lessening the temperature of operation while retaining high activity [91 , 99]. Two methodologies can be used for protein engineering [97]. In order to control the pathway of the process, either a screening test for the assessed feature is performed after each round of modification, or selective pressure is applied [–]. This methodology, which allows for a high throughput, has been extensively applied, aiming for more efficient biocatalysts [–]. Some relevant examples in the area of food and feed processing include the following. This pattern is often reported when glucose isomerases from hyperthermophilic strains operate in mesophilic environments. Large-scale glucose isomerization is carried out at 55°C and slightly alkaline pH [1 , 31]. This set of conditions results from the optimal range of pH typically 7. The latter result from by-product and color formation occurring when the reaction is carried out at alkaline pH and high temperatures [31 ,]. There is therefore interest in selecting an enzyme able to operate efficiently at temperatures close to those currently used but at a lower pH. The mutant glucose isomerase 1F1 obtained by Sriprapundh and coworkers displayed a roughly 5-fold higher activity at and pH 5. The encouraging results obtained suggest the soundness of the approach to obtain a mutant glucose isomerase competitive with those currently used, while being able to operate in a slightly acidic environment and. IM [], of the amylosucrase from *Neisseria polysaccharea* [], of the glucoamylase from *Aspergillus niger* [], of a phytase from *Escherichia coli* [,], and of a xylanase from *Bacillus subtilis* []. Amylases and glucoamylases are enzymes used in starch processing, which involves temperatures typically in excess of ; hence, improving thermal stability without decreasing enzyme activity is of relevance. Starch liquefaction is performed at in the presence of -amylase, upon which the effluent reaction stream has to be cooled to , so that glucoamylases can be used. In order to avoid, or at least minimize, the cooling step, thermostable glucoamylases are aimed at. Furthermore specific activities and catalytic efficiencies remained unaltered, when mutant and wild type were compared []. Kim and coworkers obtained also a multiply-mutated amylase R26Q, SN, IV, MT, AV, QL, PL which displayed an optimal reaction temperature higher than that of the wild-type and a half-life of roughly min at , a temperature at which the wild-type ThMA was fully inactivated in less than 1 minute. Actually, the mutant was claimed to be the only amylosucrase usable at. At the latter temperature, the mutant enabled the synthesis of amylose chains twice as long as those obtained by the wild-type enzyme at , for sucrose concentrations of mM. The mutant thus allowed for a process with increased yield in amylose chains 31 g L⁻¹ , lower risk of contamination, enhanced substrate and product solubility and overall productivity []. Phytases are added to animal feeds to improve phosphorus nutrition and to reduce phosphorus excretion, by promoting the hydrolysis of phytate into myoinositol and inorganic phosphate. Thermal stable enzymes are needed, since

feed pelleting is carried out at high temperature 60 to. Phytases produced by thermophiles do not provide a suitable approach, since they have low activity at the physiological temperature of animals []. No significant changes in the pH activity profile were observed, but for some mutants, containing a K46E substitution, that displayed a decrease in activity at pH 5. Accordingly, these enzymes can be used in dough making, in baking, in brewing and in animal feed compositions. When the latter contain cereals namely, barley, maize, rye or wheat , or cereal by-products, xylanases improve the break-down of plant cell walls, which favors the ingestion of plant nutrients by the animals and consequently enhances feed consumption and growth rate. Furthermore, the use of xylanases decreases the viscosity of xylan-containing feeds [,]. As referred for phytases, the formulation of commercial feed often involves steps at high temperatures. Xylanases added to the the formulations hence have to withstand these conditions, while they are to display high activity at about , which is the temperature in the intestine of animals. However, most xylanases are inactive at temperatures exceeding , hence the need for enhancing thermal stability [,]. Miyazaki and coworkers obtained a triple-mutant xylanase Q7H, N8F, and SC which retained full activity for 2 hours at , whereas the wild-type enzyme was inactivated within 5 minutes under the same conditions. The mutation also led to a increase in the optimal temperature for reaction and enhanced activity at higher temperatures, albeit at the cost of decreased activity at lower temperatures, as compared to the wild-type enzyme []. Amylosucrases can be used for the modification or synthesis of amylose-type polymers from sucrose, but their industrial application is somehow thwarted by the low catalytic efficiency on sucrose and by side reactions leading to the formation of sucrose isomers. Van der Veen and co-workers engineered mutant enzymes through error-prone PCR that displayed increases in activity up to 5-fold and in overall catalytic efficiency up to 2-fold, when compared to the wild-type enzyme.

Chapter 3 : Fish Feed And Novel Enzyme Mixture | Manufacturer from Hyderabad

Included in the book is a history of the role of enzymes in food processing, enzyme characterization, a discussion of different classes of enzymes including lipases and proteases, commercial enzyme production, and the processing of particular foods such as meat, vegetables, fruit, baked goods, milk products, and beer.

Chapter 4 : AMFEP: Association of Manufacturers and Formulators of Enzyme Products

Advanced Enzymes offers eco-safe solutions for a variety of industries such as Textiles, Leather, Detergent, Paper & Pulp. Go ahead with Most Trusted Non-Food Processing Enzymes Manufacturers. About Us.

Chapter 5 : Enzymes in Food Processing - Google Books

An important role of Enzymes in the food industry is to accelerate the chemical changes and thus saves the processing time and money. It imparts high nutritional value, flavor, and texture to the food. Food Processing can be categorized as follows: Fruit & Vegetable Processing - Some of the common.

Chapter 6 : Products “ Advancedenzymes

Abstract. Food and feed is possibly the area where processing anchored in biological agents has the deepest roots. Despite this, process improvement or design and implementation of novel approaches has been consistently performed, and more so in recent years, where significant advances in enzyme engineering and biocatalyst design have fastened the pace of such developments.

Chapter 7 : Enzymes for food processing - Products - DSM

The Enzymes for Food Processing research study covers all aspect of the Enzymes for Food Processing market

globally, which starts from the definition of the Enzymes for Food Processing industry and develops towards Enzymes for Food Processing market segments.

Chapter 8 : Food Enzymes | Freshness, texture improvement and process optimisation - DuPont | Danisco

Food Enzymes Present in plants, animals and microorganisms, enzymes are proteins that function as catalysts for the thousands of chemical reactions that take place in all living cells. These natural substances are ideal for use in the modern food industry.

Chapter 9 : Home - Amano Enzyme

Without enzymes, those reactions simply would not occur or would run too slowly to sustain life. For example, without enzymes, digestion would be impossible. Enzymes are proteins, vital components of any living organism: microorganisms, plants, animals and us, humans.