
A REVIEW OF THE USE OF MICROBIAL AMYLASE IN INDUSTRY

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ABSTRACT

Amylases are one of the most often utilized enzymes in business. Enzymes like these hydrolyze starch molecules into polymers made up of glucose units. Amylases have the potential to be used in a variety of commercial activities, including food, fermentation, and pharmaceuticals. Amylases come from a variety of sources, including plants, animals, and microbes. In the industrial sector, however, enzymes derived from fungi and bacteria have prevailed. The enzyme amylase is required for the conversion of starches to oligosaccharides. Starch is a key component of the human diet and a significant storage product in a variety of commercially important crops, including wheat, rice, maize, tapioca, as well as potato. Maltodextrin, modified starches, and glucose or fructose syrups are all made using starch converting enzymes. A wide range of microbial amylases are used in a variety of industries, including food, textiles, paper, and detergents. Submerged fermentation has traditionally been used to make amylases, however submerged fermentation systems seem to be a viable technique. The thermo stability, pH profiles, pH stability, and Ca-independency of each amylase are essential in the development of the fermentation process. The synthesis of bacteria and fungi amylases, their distribution, structural-functional features, physical and chemical parameters, and their usage in industrial applications are all covered in this study.

KEYWORDS: *Amylase, Bacterial, Enzyme, Fungal Amylase, Starch.*

1. INTRODUCTION

Amylases catalyze the hydrolysis of inner 1, 4-glycosidic connections in starch to produce low molecular weight products like glucose, maltose, and malt triose units. Amylases are one of the most significant enzymes and are very important in biotechnology; they are a class of industrial enzymes that account for around 25% of the global enzyme market. They may be found in a variety of places, including plants, animals, and microbes. Today, a wide range of microbial amylases are commercially accessible, and they have almost entirely replaced chemical starch hydrolysis in the starch processing business. Microorganisms' amylases have a wider range of industrial uses than plant and animal amylases because they are more stable. The main benefit of utilizing microorganisms to make amylases is the low cost of mass manufacturing and the ease with which microbes can be manipulated to produce enzymes with desired properties[1].

Amylase comes from a variety of fungi, yeasts, and bacteria. In the industrial sector, however, enzymes derived from fungi and bacteria have prevailed. Amylases may be used in a variety of industrial activities, including food, fermentation, textiles, paper, detergents, and pharmaceuticals. Amylases from fungi and bacteria may be helpful in the pharmaceutical as well as fine-chemical industries. However, because to advancements in biotechnology, amylases are now used in a

variety of areas, including clinical, pharmaceutical, and analytical chemistry, as well as starch saccharification and the textile, food, brewing, and distilling industries. Amylases are one of the most common and essential types of industrial amylases, and the microorganisms that generate them are highlighted in this study[2].

1.1. Amylase's Structural or Functional Characteristics

Microorganisms, plants, and higher organisms all contain amylase (1,4-glucan-4-glucanohydrolase). The amylase is a member of the endo-amylase family that catalyzes the first hydrolysis of starch into shorter oligosaccharides by cleaving -D-(1-4) glycosidic linkages. -amylase cannot cleave either terminal glucose residues or 1,6-linkages. -amylase activity produces oligosaccharides of various lengths with a configuration and limit dextrins, which are a combination of maltose, malt triose, or branching oligosaccharides of 6–8 glucose units with both 1,4 and 1,6 links. Other amylolytic enzymes play a role in starch breakdown, however amylase is the most essential for the start of the process.

Amylase has a three-dimensional structure that allows it to attach to substrate and induce glycoside link breaking via the action of highly specialized catalytic groups. Human -amylase is a calcium-containing enzyme that is made up of 512 amino acids in a single oligosaccharide strand with a molecular weight of 57.6 kDa. A, B, and C are the three domains that make up the protein. The biggest domain is A, which has a conventional barrel-shaped superstructure. The B domain is placed between the A and C domains and is sulphide-bonded to the A domain. The C domain seems to be a separate domain with unknown function, having a sheet structure connected to the A domain by a simple polypeptide chain. The -amylase's active site (substrate binding) is positioned in a lengthy cleft between the carboxyl terminuses of the A as well as B domains. Calcium (Ca²⁺) is located between a And B domains and may function as an allosteric activator as well as a three-dimensional structural stabilizer. Asp206, Glu230, and Asp297 are thought to be involved in catalysis based on their ability to bind substrate analogs. The catalytic site is located at subsite 3 of the substrate-binding site, which has 5 subsites. Substrate may bind to the first glucose residue in subsites 1 and 2 and cause cleavage between the first and second or second and third glucose residues[3], [4].

1.2. Production of Amylase:

Submerged fermentation (SmF) as well as solid state fermentation (SSF) have been studied for the generation of -amylase and are dependent on a number of physicochemical variables. Because of the simplicity with which various factors like as pH, temperature, aeration and oxygen transfer, and moisture can be controlled, SmF has historically been employed for the synthesis of industrially significant enzymes. Because of its inherent potential and benefits, SSF systems seem to be promising. SSF is the ideal option for microorganisms to thrive and generate valuable value added products since it closely mimics their natural environment. SmF, particularly in the case of fungus, may be regarded a breach of their natural environment. According to the theoretical notion of water activity, fungi and yeast were deemed appropriate microorganisms for SSF, while bacteria were deemed unsuitable. Bacterial cultures, on the other hand, have been proven to be easily controlled and manipulated for SSF procedures. Other advantages of SSF over SmF include higher productivity, a simpler technique, lower capital investment, lower energy requirements and less water output, better product recovery, and the absence of foam build-up, as well as the fact that it is said to be the best process for developing countries. SSF has recently been tested to see whether it is the optimum system for generating enzymes. They discovered that SSF is suitable for the synthesis of enzymes and other thermolabile products, particularly when yields are greater than those achieved with SmF[5], [6].

1.2.1. Amylases Produced By Bacteria:

Amylase may be generated by a variety of bacteria; however commercially available amylase is mostly sourced from the *Bacillus* genus. *Bacillus licheniformis*, *Bacterial stearothermophilus*, or *Bacillus amyloliquefaciens* generate amylases that may be used in a variety of industrial applications, including food, fermentation, textiles, and paper. Thermostability is a desirable feature in the majority of commercial enzymes. Because of their stability, thermostable enzymes isolated from thermophilic organisms have found a variety of commercial uses. High temperatures are used for enzymatic liquefaction and saccharification of starch. Thermo stable amyolytic enzymes are being researched to enhance industrial starch degradation processes and are of considerable importance for the manufacture of important products such as glucose, crystalline dextrose, dextrose syrup, maltose, and maltodextrins. *Bacillus subtilis*, *Bacillus stear other mophilus*, *Bacillus licheniformis*, and *Bacillus amyloliquefaciens* are all known to generate thermo stable amylase and have been extensively utilized for commercial production of the enzyme for a variety of purposes[7], [8].

Some halophilic bacteria generate enzymes that have optimum activity at high salinities and may therefore be utilized in a variety of severe industrial processes where the concentrated salt solutions used would normally block many enzymatic conversions. Furthermore, most halo bacterial enzymes are very thermotolerant and can survive for extended periods of time at room temperature. *Chromohalobacter sp.*, *Halo bacillus sp. (4)*, *Haloarcula hispanica*, *Halo Monas meridiana*, and *Bacillus dipsosauri* have all been shown to produce halophilic amylases

1.2.2. Amylases From Fungi:

The majority of studies on fungi that generate amylase have been confined to a few species of mesophilic fungi, with efforts to define the culture conditions and select better strains of the fungus for commercial production. Terrestrial isolates of *Aspergillus* and *Penicillium* are the most common fungal origins. The *Aspergillus* species generate a wide range of extracellular enzymes, with amylases being the most important for industry. Filamentous fungus like *Aspergillus oryzae* and *Aspergillus Niger* generate large amounts of enzymes that are often utilized in the industry. Because of its capacity to produce a large number of high-value proteins and industrial enzymes, such as amylase, *A. oryzae* has gotten a lot of interest as a good host for heterologous protein synthesis

1.3. Amylase Purification

Industrial enzymes are often generated in bulk and need minimal downstream processing, making them very basic formulations. Purification of amylase is not required for commercial usage, however high purity amylases are required for pharmaceutical and therapeutic uses. Purified enzymes are also required for research into structure-function connections and biochemical characteristics. Different purification methods for enzymes have been explored, based on the target biomolecule's unique properties. Ion exchange, gel filtration, hydrophobicity interactions, and reverse phase chromatography are all used to purify amylase at the laboratory scale. Amylase extraction methods using organic solvents like ethanol, acetone, and ammonium sulfate precipitation and ultrafiltration have also been suggested[9].

1.4. Amylase's Industrial Applications:

1.4.1. Conversion Of Starch:

Amylases are most often employed in the starch business, where they are used for starch hydrolysis in the starch liquefaction process, which turns starch into fructose and glucose syrups. Gelatinization, which involves the dissolving of starch granules and the formation of a viscous suspension; liquefaction, which involves partial hydrolysis and the loss of viscosity; and saccharification, which involves the creation of glucose and maltose via additional hydrolysis.

Detergent manufacturing Enzymes are mostly used by the detergent industry, both in terms of volume and value. Enzymes are used in detergent formulas to improve the detergent's capacity to remove stubborn stains while also making it ecologically friendly. Amylases are the second kind of enzyme utilized in enzymatic detergent composition, and they are found in 90% of all liquid detergents. These enzymes are used in laundry detergents and automated dishwashing machines to break down starchy food residues such as potatoes, gravies, custard, chocolate, and other oligosaccharides into dextrans and other smaller oligosaccharides. Amylases have activity at lower temperatures and alkaline pH, allowing them to retain the required stability in detergents. Amylases' oxidative stability is one of the most significant requirements for their usage in detergents with a highly oxidizing washing environment.

1.4.2. Ethanol Production as a Fuel:

The most widely used liquid biofuel is ethanol. Starch is the most often utilized substrate for ethanol synthesis owing to its cheap cost and availability in most parts of the globe. In order to get fermentable sugars, starch must first be solubilized and then subjected to two enzymatic processes. Liquification and saccharification, in which starch is turned to sugar using an amylolytic microbe or enzymes like amylase, are followed by fermentation, in which sugar is transformed to ethanol using an ethanol fermenting microorganism such yeast *Saccharomyces cerevisiae*.

1.4.3. Industry of Food:

Amylases are widely used in the processed food business, including baking, brewing, making digestive aids, making cakes, and making fruit juices and starch syrups. In the baking business, -amylases are often utilized. These enzymes may be added into bread dough to break down the starch in the flour into smaller dextrans, which the yeast then ferments. The addition of amylase to the dough increases the pace of fermentation and lowers the viscosity of the dough, improving the volume and texture of the finished product.

1.4.4. Textile Manufacturing:

Amylases are employed in the dyeing process in the textile industry. To guarantee a quick and secure weaving process, sizing agents such as starch are added to yarn prior to fabric manufacturing. Starch is an appealing size since it is inexpensive, widely accessible in most parts of the globe, and readily removed. In the textile finishing business, starch is subsequently removed from the woven cloth using a wet-process. Desizing is the process of removing starch from a fabric, which acts as a strengthening agent to prevent the warp thread from breaking during the weaving process. The amylases remove the size in a selective manner, avoiding the fibers. Amylase derived from *Bacillus stain* has long been used in the textile industry. In addition to being an excellent coating for the paper, starch is a good sizing agent for the finishing of paper, increasing the quality and erasability. Paper stiffness and strength are improved as a result of the size. Amylases isolated from microorganisms used in the paper industry [10], [11].

2. DISCUSSION

Amylases are one of the most often used enzymes in the workplace. Enzymes like these break down starch molecules into glucose-based polymers. Amylases may be utilized in a wide range of commercial applications, including food, fermentation, and medicines. - Amylases are found in plants, animals, and microorganisms, among other places. However, enzymes produced from fungus and bacteria have won out in the industrial sector. The production of -amylase is required for the conversion of starches to oligosaccharides. Starch is an essential part of the human diet and a major storage product in a number of economically important crops, including wheat, rice, maize, tapioca, and potato. Starch converting enzymes are used to make maltodextrin, modified starches, and glucose and fructose syrups. -amylase has been widely used in starch-based industries for decades, and although there are a number of microbiological sources for efficient

synthesis of this enzyme, only a few carefully selected fungus and bacteria strains meet the criteria for industrial production. The search for new microorganisms capable of producing amylase is never-ending. Many authors have recently reported encouraging results in developing -amylase purification techniques, opening the path for high purity amylase applications in the pharmaceutical and healthcare sectors

3. CONCLUSION

The use of amylase in starch-based businesses has been widespread for decades, and although there are a variety of microbial sources for effective amylase synthesis, only a few strains of fungi and bacteria satisfy the requirements for industrial production. The hunt for novel microbes that can produce amylase is a never-ending activity. Many authors have lately reported promising findings in the development of amylase purification methods, paving the way for applications in the pharmaceutical and clinical industries that need high purity amylases. Industry of paper Amylases are used in the pulp and paper industry to modify coated paper starch, resulting in low-viscosity, high molecular weight starch. The coating process improves the writing quality of the paper by making the surface suitably smooth and robust. The natural starch's viscosity is too high for paper size in this application, but this may be changed by partly decomposing the polymer using amylases in batch or continuous operations.

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