Review Article: Amylase producing microorganism and industrial application

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ABSTRACT

Enzymes are the most important products obtained for human needs mainly through microbial sources.Amylase is hydrolysing enzyme which breakdown starch to simple sugar form, it is present in saliva of the human and some other mammals. Base on structure and application of enzyme it can classified in three categories. Pancreas and salivary gland make amylase to hydrolyse dietary starch into disaccharides and trisaccharides than other enzymes converted to glucose (energy source). Amylase is an extracellular enzyme, which is involved in the starch processing industries where it breaks starch into simple sugar constituents.

Amylase has extensive application in starch processing, brewing and sugar production, in textile industries and in detergent manufacturing processes.

Keywords: Amylase enzyme, Amylase producing bacteria, industrial important enzyme

Sreelekshmi Mohan M. R et al.,(2019) reported that Amylase is an industrially important enzyme used for the hydrolysis of dietary starch into disaccharides and trisaccharides and ultimately to glucose. Amylase producing bacteria isolated from soil samples collected from the evergreen and deciduous forest of Jawaharlal Nehru Tropical Botanic Garden and Research Institute (JNTBGRI), Palode, Thiruvananthapuram, Kerala, India. Bacterial colony which showed the maximum zone of clearance in starch agar plates were isolated and cultured separately. The screening was done on the basis of hydrolysis of starch by amylase on agar plate containing 1% starch. Bacteria were identified as *Bacillus sps*. And apply as starch hydrolyser in industries. Kumar Pranay (2019) reported that Secondary screening based on amylase production in starch broth medium led to the selection of six amylolytic strains of *Bacillus* sp. The selected strains were grown in four different fermentation media (FMI-FMIV) in order to screen for three most efficient amylolytic strains for optimization and characterization. FMIV was the best basal medium as it provided required nutrients, which stimulated highest amylase production in bacterial strains within shortest incubation time (24 hours). Molecular identification based on 16S rDNA sequence revealed that three most efficient strains [BCM36 (KR1), BCM33 (KR2), and BCM25 (KR3)] belonged to *Bacillus* sp.

MengistuFentahun and PagadalaVijayaKumari (2017) studded that thermostable amylases are the most important enzymes in present with potential industrial applications,isolate and characterized thermophilic amylases from Bacilli found in starch rich soil. Amylase producing bacilli were isolated and their enzymes were also characterized. Effect of temperature, pH, substrate and salt concentration on amylases activity were determined. All amylases produced by different isolates were hydrolyzed greater than 91% of starch after 60 h of fermentation. Amylases from all bacilli isolates were shown hydrolysis capacity of starch ranging from 91.4 to 95.7%. The optimum enzyme activity of amylase from Isolate-2 was extended from pH 7 to 8 with starch hydrolysis efficiency of 98% but other isolates enzyme activity reaches 99.5 to 100% at pH 8. The crude amylase extract has an activity

with inversely proportional with substrate concentration. The bacterial dry weight increases as the course of incubation time increases and NaCl concentration greater than 5 molar significantly decreases the activity of the crude amylase extract. Amylases of this finding with thermophilic, alkalophilic and halophilic characteristics have wide range of huge potential for industrial applications. Besides, further purification of the crude extract could be conducted to meet thermophilic amylase enzyme requirements of pharmaceuticals and clinical sectors.M. S. Audre Preena et al.,(2017) reported that amylases have potential application in a wide number of industrial processes such as food, fermentation and pharmaceutical industries. decayed brinjal (*Solanummelongena*) was collected for bacterial isolation. Eight bacterial isolates were isolated and screened for amylase production, one isolate (B6) displaying the highest activity was selected for further study. The enzyme was purified by ion exchange chromatography and its specific activity was found to be 0.83 U/ml/min. The determination of the α-amylase activity was performed by FT-IR analysis.

Mankar S. D and Barate D. L (2018) noticed that amylases are one of the main enzymes used in industry. Such enzymes hydrolyze the starch molecules into polymers composed of glucose units. Amylases have potential application in wide number of industrial processes such as food, fermentation and pharmaceutical industries. Microbes are the most preferred sources of enzymes due to their broad biochemical diversity. From the 9 soil samples, 65 isolates out of which 29 isolates were selected based on amylase activity as clear zone on starch agar. Out of 29 isolates 10 isolates show excellent zone of hydrolysis in the secondary screening. The isolates showed prominent activity were further identified by standard conventional methods, which showed most of them belonging to genus *Bacillus* followed by Pseudomonas spp., Serratiamarcescens and Staphylococcus aureus.

SathyaRengasamy and UshadeviThangaprakasam (2018) reported that potent amylase producing Streptomyces isolated from the marine source. Soil samples were collected from less explored mangrove regions of Muthupet, Tamilnadu. Streptomyces was performed by serial dilution plate technique using starch casein agar (SCA) (pH 7.2 and temp 28 °C). Preliminary screening and quantification of amylase activities were analysed in selected Streptomyces isolates by starch agar plate and dinitrosalicylic acid (DNS) method respectively. Totally 65 isolates were separated from the marine soil. Among them, 23 strains showed different morphological features. These strains were subjected to amylase activity. Eight Streptomyces isolates (S1-S8) exhibited positive for amylase activity. The zone of clearance was exhibited in the range of diameters between 4-20 mm. Fermentation

was prompted with inorganic salt starch agar, international Streptomyces project (ISP-4) media at 28 °C and incubated in an orbital shaker at 250 rpm for 96 h (pH 7.5). The quantitative estimation of amylase activity was exhibited selected eight isolates in the range between 2.4±0.002-5.9±0.005 (U/ml). The Streptomyces species S4, S5 and S6 exhibited strong amylase activity in both qualitative and quantitative level. Amylase producing Streptomyces are originated in mangroves and it proved Streptomyces sp. S6 has a more efficient source of amylase production.

Md. Rayhan Islam et al (2016) Alpha amylases (α-amylases) are one of the most imperative enzymes for producing simple sugar units from complex sugar molecules. Amylolytic bacterial strains were isolated from soil samples of tannery wastes collected from Hazaribagh, Dhaka and subsequent partial characterization was performed. Bacterial isolates were primarily screened for αamylase activity on starch agar medium. Based on microscopic and biochemical properties of isolates, α-amylase activity of bacterial isolates were determined to find out two best producers of the enzyme. Subsequent molecular identification of these two α-amylase producing bacterial isolates using 16s rRNA sequence analysis showed that isolates were *Bacillus*amyloliquefaciens and B. subtilis respectively. In submerged fermentation the B. amyloliquefaciens showed the highest activity (2.13 U/ml) while B. subtilis showed the second highest activity (1.89 U/ml). Characterization of the enzyme produced by B. amyloliquefaciens revealed that the maximum activity demonstrated at incubation time 25 min, pH 7.0 and temperature 500C. This newly isolated B. amyloliquefaciens could be exploited for the industrial production of α-amylase with commercial implications.

Tole S. B.,(2016) reported that amylase can be derived from several sources such as plants, animals and microbes. The microbial amylases meet industrial demands because it is economical when produced in large quantities. Amylase producing bacteria isolated from the soil samples collected from TuljaBhavani Temple of Tuljapur. Total 20 bacterial isolate were isolated from soil samples and screened their amylolytic activity on starch agar plate method. Among 09 bacterial isolates, only 03 isolates showed the amylolytic activity. The enzyme production was done by using amylase production media at 37°c in shake flask culture. The enzyme was extracted by centrifugation and then it was purified by ammonium sulphate precipitate 40%. The partially purified fraction was used for the charcterisation studies of amylase.It was 1 fold purified fraction with 90.2 % yield.

Shyam Sunder Alariya (2013) reported that Amylases are among the most important enzymes and great significance in present day industry. Starch degrading bacteria are most important for industries such as food, fermentation, textile and paper. Thus isolating and manipulating pure culture from various soil and waste materials has manifold importance for various biotechnology industries. In the present investigation bacterial strains were isolated from soil sample and growth pattern as well as optimum growth condition was determined. Characteristic feature of the strains indicates them as *Bacillus* subtilis, Pseudomonas florescens, E. coli and Serratiamarscens. The optimum temperature for production was 35-40 ^{0}C, whereas maximum growth was observed at 1% dextrose concentration but increases with increase in substrate concentration. The pH range was found to be 7 and incubation time 48hrs with 1ml as inoculum for optimum growth. Other optimum parameters include Yeast extract as Nitrogen source, Calcium chloride as chloride and Manganese sulphate as sulphate source for amylase production.

B. Sundarapandiyan and S. Jayalakshmi (2017)studied that isolation, screening and optimization of the enzyme production by using the marine bacterium *Bacillus* subtilis SJ33. They observed the growth parameters showed the profound influence on the amylase production on the candidate species such as the maximum production of the enzyme obtained at 48 hrs of incubation and the optimum was pH 7. The temperature also influenced on the maximum production of the amylase, the optimum was 35°C for higher production of the enzyme. The optimum salinity of 2.0% showed to maximum production of the amylase. In the study, the starch and peptone was found to the best carbon and nitrogen sources to maximum production of the amylase. With optimized parameters in the mass scale medium maximum growth of 2.89 OD and enzyme activity of 64U/ml/min was obtained at 48 hrs. of incubation.

T. Panneerselvam1 and S. Elavarasi (2015) studied that *Bacillus* subtilis is an aerobic, Gram-positive, endospore forming bacterium that has the ability to produce and secrete the hydrolyzing carbohydrate enzyme, - amylase. used in various industries to rapidly degrade complex polysaccharides (e.g. starches) into smaller oligosaccharides. Starch is an abundant carbon source in nature, and -amylase (1, 4-a-D- glucanohydrolase), which hydrolyzes a-1, 4-glucosidic linkage in starch-related molecules, is one of several enzymes involved in starch degradation. Amylase is the enzyme which breaks down starch into glucose molecules and commonly called as glycoside hydrolase enzymes. Amylases are among the most important industrial enzymes and also have great significance in Microbiology studies. In this paper,

we screened soil bacteria and an isolate alkalophilic*Bacillus* subtilis was found to produce an alkaline amylase in different media *Bacillus* subtilis strain isolated from garden soil was tested for its abilities to hydrolyze the structural polysaccharides. The strain grows well at 37 o c and the 2% starch concentration, with PH near neutral. The enzyme activities were observed at 2% starch concentration. The present review was focused on bacterial amylase and this review assesses the following chapters: Amylase, Microorganisms and amylases, Physiology of amylases, Fermentation studies on bacterial amylase production and Commercial application of amylases.

CONCLUSION

Plants, animals and certain microorganisms produce amylase and use for various biochemical reactions. The most stable and reliable source of amylase is obtained from microbes compared to other sources it can produce large scale at industrial level, it can essential for conversion of starches into oligosaccharides. A large number of microbial α-amylases have applications in different industrial sectors such as food, textile, paper and detergent industries. The enzymes from microbial sources are more stable and obtained cheaply. Isolation and screening of amaylase producing microorganisms from different sample and can apply various industrial applications.

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