

Original Research Article

Amylase Production from Potato and Banana Peel Waste

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A B S T R A C T

Keywords

Amylase;
PB1; *BS9*;
Banana peel
waste; Potato
peel waste.

This project mainly aims at the use of potato and banana peel waste separately and in combination for production of industrially important enzyme amylase. It was observed that the amylase yield from the fermentation medium was 0.7 units/ml for *Aspergillus niger* and 0.8 units/ml for *Bacillus subtilis* which are known amylase producers. But one of the isolate i.e. *BS9* gave a maximum yield of 1.55 units/ml after 72 hrs. But the surprising results were obtained within just 24hrs. by *PB1* as 1.36 units/ml which can be considered as a very good yield. The additional nitrogen source peptone did not show any significant increase in the amylase yield.

Introduction

Amylases were one of the first enzymes to be produced commercially by microorganisms. Amylases are amylolytic enzymes; represents a group of catalytic proteins of great importance in carbohydrate metabolism (M. Nagajan *et al.*, 2010).

Amylase production has increased dramatically due to its wide spread use in food, textile, baking and detergent industries. Besides its use in the saccharification or liquification of starch, the enzyme is also used for the warp sizing of textile fibers, the clarification of haze formed in beer or fruits juices and for the pretreatment of animal feed to improve the digestibility (Shaista Kokab *et al.*, 2003).

Commercially amylase is produced by using Starch as a substrate and organisms used are *Bacillus subtilis* and *Bacillus diastaticus*. Asia is the largest producer of banana and potato. At present India is the biggest potato producer with an output of 20,280,000 tons (Andreas Schieber *et al.*, 2009).

Banana and potato fruit peels are organic wastes that are highly rich in carbohydrate content and other basic nutrients that support microbial growth. (Andreas Schieber *et al.*, 2009). This paper aims at amylase production by using banana and potato peels as a cheap substrate with the help of isolates like *BS9* and *PB1* which are compared with known organisms

like *Bacillus subtilis* and *Aspergillus niger* for the amylase yield using the same substrate.

Materials and Methods

Microbial cultures

Bacillus subtilis, *BS9*, *PB1*, maintained on nutrient agar slants with pH 7 at 37°C while *Aspergillus niger* maintained on PDA plates.

Screening of amylase producers

Screening of organisms from soil extract was carried out by using 5% potato and banana peel extract agar. The technique used for isolation was Four Quadrant Streaking method and after incubation well isolated colonies were selected and Gram stained. Organisms showing Gram Positive thick rods in chains were selected and purified using Nutrient Agar. From the different isolates obtained only two isolates as *BS9* and *PB1* were used for studying Amylase production

Substrate preparation

The banana and potato peels were separately grinded to form paste and used as substrate for production. Concentration of substrate maintained was 50%. After slight boiling, the mixture was filtered and the extract obtained was sterilized at 121°C for 15min. and then used for actual production.

Innoculum preparation

Suspension of *BS9*, *PB1*, *Bacillus subtilis* and *Aspergillus niger* were prepared separately in sterile tubes and were standardized to an O. D. 1 at a wavelength of 670nm.

PARAMETERS

pH

All the flasks were adjusted to pH 7 except the flask in which *Aspergillus niger* was inoculated. This flask was adjusted to pH 3.

Incubation period

Flask after inoculation was subjected to incubation for 3 days in which amylase was estimated after every 24 hrs at 37°C.

Temperature

The inoculated flasks were maintained at a temperature of 37°C.

Additional nitrogen source

0.5 gms of peptone was used as nitrogen source per 100ml of medium in the last three sets. Keeping all the above parameters constant total 6 different sets were prepared to compare the amylase production by using 50% of waste in each of the sets.

Method

Estimation of amylase was carried out using Khandeparkes method modified by Stress and Clones.

Result and Discussion

In 50% Banana peel extract broth *BS9* was found to give a higher yield as compared to other organisms after 72 hrs. i.e.1.55 units/ml. In 50% Potato peel extract broth *B. subtilis* gave a higher yield of 0.8 units/ml after 48 hrs.

Figure.1 50% Banana peel extract

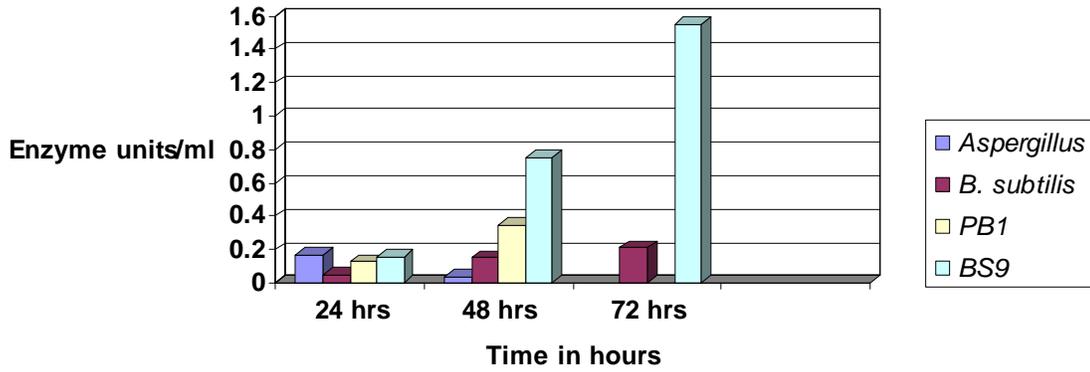


Figure.2 50% Potato peel extract-

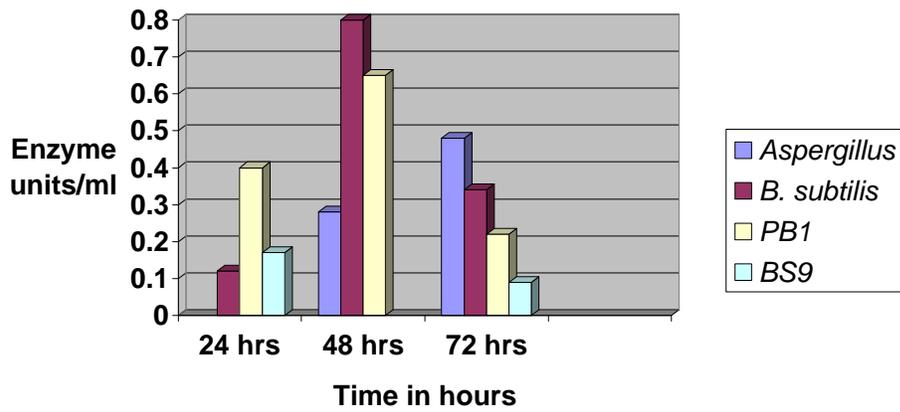


Figure.3 50% Potato + Banana peel extract

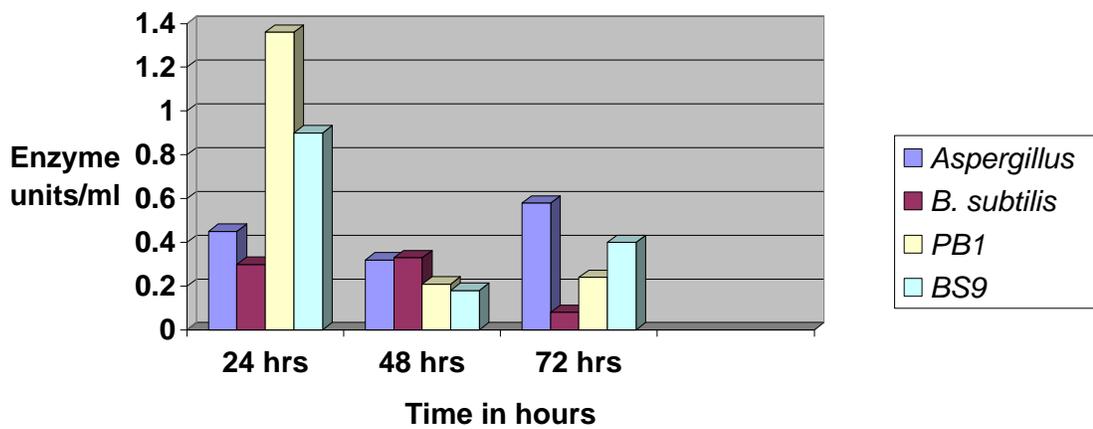


Figure.4 50% banana peel extract + peptone

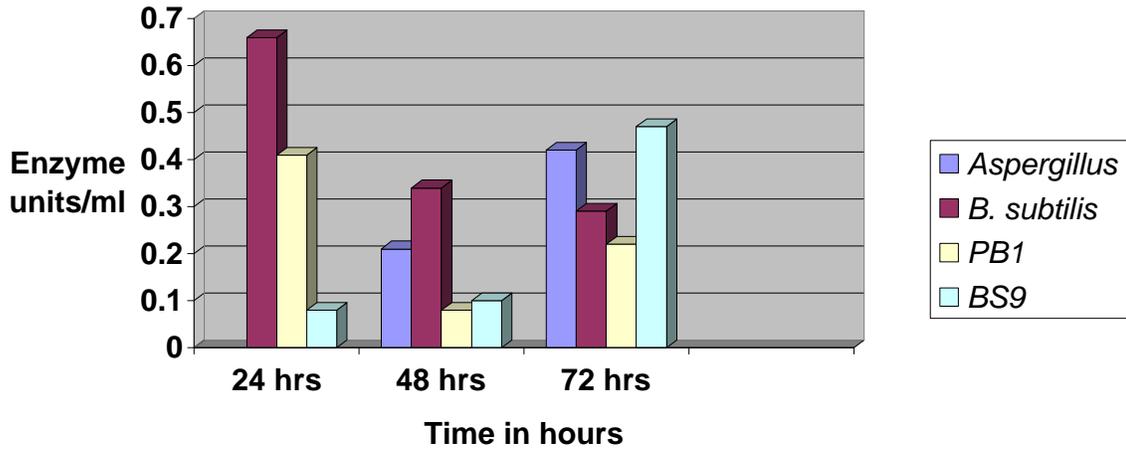


Figure.5 50% potato peel extract + peptone

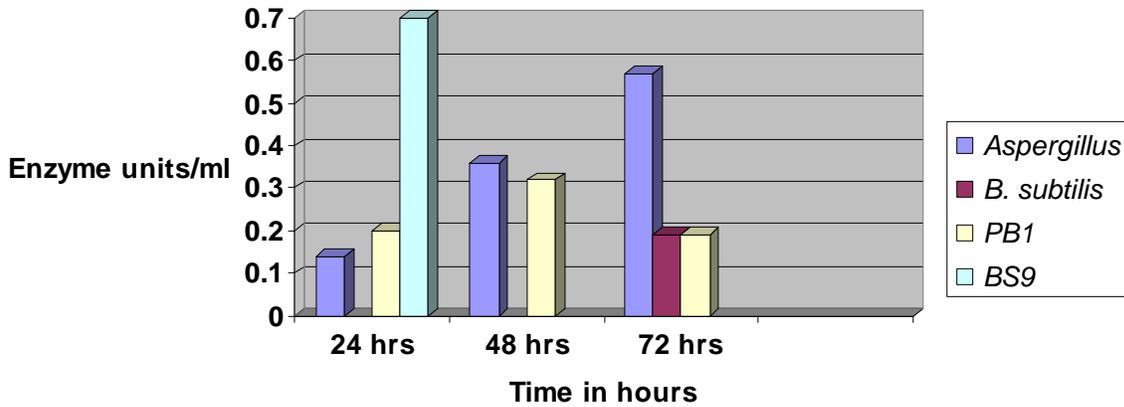
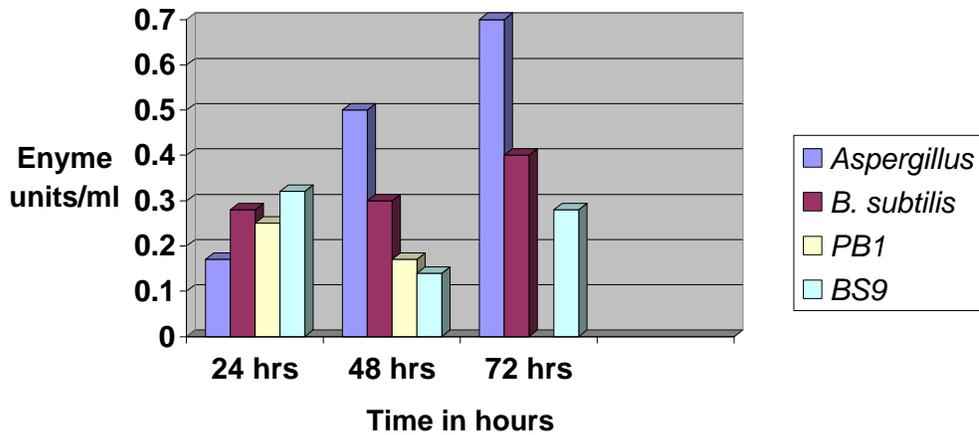


Figure.6 50% Banana + Potato peel extract + Peptone



In 50% Potato + banana peel extract broth *PB1* gave a higher yield of 1.36 units/ml after 24 hrs. In 50% Banana peel extract broth when Peptone was added as a nitrogen source *B.subtilis* gave a higher yield of 0.66 units/ml after 24hrs. In 50% potato + Peptone it was observed that highest yield was given by *BS9* i.e. 0.7 units/ml after 24hrs. In 50% Potato + Banana with peptone it was observed that *Aspergillus* gave a high yield i.e. 0.7 units/ml after 72 hrs (Figures 1-6). Amylase was produced by all the four organisms viz. *BS9*, *PB1*, *Bacillus subtilis* and *Aspergillus niger*. But the maximum activity of amylase was given by the isolates in absence of additional nitrogen source i.e. peptone.

In an initial experiment fresh mashed peel was used as a substrate to study the amylase production from four different organisms as *BS9*, *PB1*, *Bacillus subtilis* and *Aspergillus niger*. The isolates showed good growth on mashed peel and so it was selected to be used as substrate for amylase production. The concentration of medium was 50% only as further increase in the substrate did not increase the enzyme yield and the turbidity of substrate interfered with the optical density. While referencing it was observed that major research work was done on Potato and banana peels for the production of amylase but separately. This project indicates the use of equivalent mixture of potato and banana peels extract for Amylase production which acts as a cheaper substrate. The supplied nitrogen source i.e. peptone in particular sets showed no positive effect on the enzyme production (Swetha Sivaramakrishnan *et al.*, 2007). Potato and banana peel offers good option if research on the possibility of augmenting its nutritional status is carried out; otherwise the potential of these organisms to utilize the substrate could be

harnessed for effective waste management (J.P. Essien *et al.*). The potato and banana peels were used in combination for production of amylase which could also be produced efficiently by controlled fermentation techniques using different organisms. (Manikandan *et al.*, 2006). Amylase was produced by all the four organism used in present work. The maximum activity of amylase was given by *BS9* (1.55 units/ml) which was recorded after 72hrs at pH 7 by using 50% banana peel as a substrate. Thus, it can be concluded that this waste used in the project proves to be useful in production of industrially useful enzyme amylase.

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