Effective use of partially purified α-amylase produced by Brevibacillus Borstelensis R1 in Detergent and Textile Industry

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Abstract— The main objective is to test the efficiency of partially purified α-amylase produced by Brevibacillus borstelensis R1 in automation dishwashing, laundry and to test the capabilities of partially purified α-amylase as desizing agent in textile industries. Submerged fermentation was used in α-amylase production. In textile industry, the α-amylase acted as a better desizing agent on sari, towel and navaar samples than caustic soda. Alpha-amylase together with the detergent proved to be more efficient to degrade the residues of starchy foods. The α-amylase was found to be useful in automation dishwashing and laundry industries.

Key words: Brevibacillus borstelensis R1, α-amylase, Textile industry, Automation dishwashing; Laundry industry

I. INTRODUCTION

Alpha-amylase has applications in starch processing, desizing of textiles, paper sizing, detergent additive, bread improvement, ethanol production, sewage treatment, effluent treatment and other fermentation processes [1-4]. Detergent industries are the primary consumers of enzymes, in terms of volume and value. Enzymes used in laundry detergent must be inexpensive, stable, and safe to use. The use of enzymes in detergent formulations enhances the detergent’s ability to remove tough stains and make the detergents environmentally safe. Automation dishwashing in restaurants and house hold usage is common. The demand is high for α-amylase detergents use in automatic dishwashing. Amylases are used in the formulation of enzymatic detergents and 90% of all liquid detergents contain amylase enzymes [5-8].The enzymes in detergent formulation used in laundry and automatic dishwashing degrade the residues of starchy foods such as potatoes, custard, chocolate, gravies, etc. to dextrans and other smaller oligosaccharides [9,10].

Amylases are used in textile industry for desizing process. Prior to weaving of yarn into fabric, the warp yarns are coated with a removable sizing agent to lubricate and protect the yarn from abrasion during weaving. Historically, the main sizing agent used for cotton fabric has been starch because of its excellent film forming capacity. Before the fabric is dyed, the applied sizing agent must be removed. Before the discovery of amylase enzymes, the only method to remove the starch-based sizing was extended treatment with caustic soda at high temperature. However such chemical treatment does not totally effective in removing the starch, appropriate desizing of starch is now possible by application of alpha-amylase. Recently, α-amylase from Bacillus strain is employed in textile industries as desizing agent [11-14].

II. MATERIALS AND METHODS

A. Estimation of amylase by DNS method

Maltose produced by the hydrolytic activity of α-amylase on α-1, 4 linkages present in polysaccharides, reduce 3, 5 dinitro salicylate to an orange red colored 5-nitro 3-amino salicylate which can be measured at 520nm. The starch substrate [0.5ml of 0.5% in 0.1M phosphate buffer (pH 6.8)] was mixed with 1% (0.2ml) NaCl in a test tube and pre incubated at 37°C for 10 minutes. The supernatant collected from the centrifugation of the production media was used as enzyme source, 0.5ml of this was added to the reaction mixture. The reaction was terminated by the addition of 1.0 ml of 3, 5-dinitrosalicylic acid reagent [1.0 gm DNS in 0.8% NaOH, 60% Na K tartrate] after incubation at 37°C for 15 minutes. The contents were mixed well and kept in boiling water bath for 10 minutes. Then they were cooled and diluted with 10 ml of distilled H2O. The color developed was read at 520nm. One unit of enzyme activity is defined as the amount of enzyme that releases 1.0 mmol of reducing sugar (maltose) per minute under the assay conditions [15].

B. Textile industry

In the small scale weaving industry at Peddapurum, East Godavari District, Andhra Pradesh, India, the common process of sizing of the cotton yarn was observed. Porridge was produced by thorough boiling of rice at 100°C. The porridge was allowed to cool for few minutes and applied to cotton yarn by using brush, then allowed to dry for few minutes. The yarn was used to weave saris, towels, bed sheets and navaar. The sample piece of a sari, towel and navaar were taken to test the desizing effect of the enzyme and caustic soda. The sample pieces were subjected to the partially purified α-amylase (100U/mg protein) produced by B. borstelensis strain R1 and caustic soda (5%) by soaking for 15 minutes. The softness of the samples was tested.

C. Automation Dishwashing (Restaurants)

The automation dishwashing (Manf. Winterhalter, Germany) used at Dasapalla Group of Hotels for washing vessels used in rice cooking, starchy vegetable curries, fries, soups etc. Plates, cups, glasses were subjected to different washing detergent preparations; partially purified α-amylase (100U/mg protein), detergent (5%) separately and mixture of α-amylase and detergent (5%). Hot water cleaning served as control in automated dish washing machine.

D. Laundry industry

Stains of jaggary, starch gravies, rice porridge and chocolate on different sample pieces of cloth were subjected to treatment with partially purified α-amylase (100U/mg protein), detergent (5%) separately and α-amylase mixed with detergent (5%). The cloth pieces with and without...
stains were taken as positive and negative controls respectively. The efficiency to remove the stains by the enzyme was observed after incubation for 30 minutes at 25°C.

III. RESULTS

Partially purified α-amylase from *Brevibacillus borstelensis* R1 was 3.9 fold purification, when the crude enzyme was subjected to (NH₄)₂SO₄ precipitation and gel filtration. Alpha amylase proved it’s potency in many fields: in automation dishwashing, laundry and textile industries.

A. Textile industry

In textile industry, the amylase plays an important role in desizing. The present study was carried out to observe the desizing affect of the α-amylase (100U/mg protein) on the sari, towel and navaar samples. The results of the experiments were noticed for softness of the samples as shown in figures 1A-I. The desizing effect of α-amylase was better than the regularly used caustic soda.

![Fig. 1: Desizing effect of enzyme on a sample of sari, towel and navaar. A sample of sari treated with (A) starch, (B) 5% caustic soda and (C) α-amylase (100U/mg protein). A sample of towel treated with (D) starch, (E) 5% caustic soda and (F) α-amylase (100U/mg protein). A sample of navaar treated with (G) starch, (H) 5% caustic soda and (I) α-amylase (100U/mg protein)](image)

B. Automation dishwashing (Restaurants)

Alpha-amylase was used commercially in automation dishwashing in Desapalla Group of Hotels. The α-amylase (100U/mg protein) and detergent were proved to be more efficient to degrade the residues of starchy foods such as potatoes, gravies, custard, chocolate and soup than treated with detergent alone or with α-amylase (100U/mg protein) alone (Figures 2A&B).

![Fig. 2: Automation Dishwashing (A) Vessels washer and (B) cup & Glasses washer](image)

C. Laundry Industry

In removing jaggary, starch gravies, rice porridge and chocolate stains in laundry industry- the stains treated with α-amylase (100U/mg protein) and detergent were proved to be more efficient than the stains treated with only detergent or with only α-amylase (100U/mg protein) (Figure 3A-X).

![Fig. 3: Destaining effect of α-amylase (A) Jaggary stain (B) positive control, (C) treated with α-amylase (D) treated with detergent (E) treated with α-amylase & detergent and (F) negative control. (G) Starch gravies stain (H) positive control (I) treated with α-amylase (J) treated with detergent (K) treated with α-amylase & detergent and (L) negative control. (M) Rice porridge stain: (N) positive control (O) treated with α-amylase & detergent and (P) negative control. (S) Chocolate stain: (T) positive control (U) treated with α-amylase (V) treated with detergent and (W) treated with α-amylase & detergent and (X) negative control](image)

IV. DISCUSSION

In textile industry, α-amylase produced from *B. borstelensis* R1 used as a good desizer when applied on the sample pieces of sari, towel and navaar. Similarly, amylases from *Bacillus* strain employed in textile industry were reported to show promising results [16-21]. Alpha-amylase along with detergent worked more effectively in automatic dishwashing in removing tough stains. Similar work on the role of amylases in modern detergents was carried out by Olsen & Falholt [22], Kirk *et al.* [23], Chi *et al.* [24], Hmidet *et al.* [25] and Mukherjee *et al.* [26].

V. CONCLUSIONS

In textile industry, the α-amylase acted as a better desizing agent on sari, towel and navaar samples than caustic soda. Alpha-amylase, when commercially used in automation dishwashing in local group of hotels, the α-amylase together with the detergent proved to be more efficient to degrade the residues of starchy foods such as potatoes, gravies, custard, chocolate and soups.

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