Effect of different carbohydrates on the production of pectin methyl esterase by solid state fermentation

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Pectin methyl esterase (PME) enzyme one of the main commercially available enzymes which is produced by solid state fermentation (SSF) process. Pectic methyl esterase are of major importance to the food industry because of their effect on the texture of foods such as apples, peaches, and tomatoes and in the preparation of wines and fruit juices. Optimum enzyme production depends upon environmental condition as well as nutritional factors. In the current study SSF medium was enriched with different carbohydrates to improve enzyme yield. Maximum yield of PME was obtained with addition of 5% (w/v) lactose in the fermentation medium compared to control. Maximum pH change was observed in the control during fermentation. Highest residual sugar was available after fermentation in the medium containing glucose at a level of 5% (w/v).

Key words: Tissues, solid state fermentation, texture, carbohydrates

INTRODUCTION

Pectinases are a group of enzymes that degrade smaller pectin-containing substances into fractions thus resulting into reduction of viscosity and facilitate extraction, maceration, liquefaction, filtration and clarification processes. Pectinases are widely used in fruits and vegetables processing industry as reported by Kareen et al. (2007), for processing of wine, coffee and tea fermentation as studied by Nagodawithana et al. (1993) and also in the textile industry. Submerged fermentation (Smf) system has been extensively employed for production of enzyme. However, an alternative technique of enzyme production is solid state fermentation (SSF). Higher product yield and easier down stream processing makes SSF advantageous than Smf (Blandino, et al., 2002). Due to excessive increasing cost of enzyme production SSF are becoming a viable alternative technique for large-scale production of enzyme as reported by Louboudy, et al. (2001). Filamentous fungi are the most important group of microorganisms used in SSF processes owing to physiological, enzymological biochemical properties. The hyphal mode of fungal growth and their good toterance to low water activity and high osmotic pressure conditions make them efficient and competitive for bioconversion of solid substrates. Moreover, use of agricultural wastes to produce extra cellular enzymes of importance makes the process economic also. The production of extra cellular pectinases is induced by ago-industrial wastes which contain appreciable quantities of pectin (Martins, et al. 2002, Sinha et al., 2009. In the present study the effect of addition of different nitrogen sources and also metal salts on the production of PME has been studied.

MATERIALS AND METHODS

Microorganism : The strain of *Penicillium notatum* was used as a culture.

Production of crude enzyme extract: After fermentation (30°C for 5 days) the solid medium was soaked with distilled water (1:10, w/v) for 4 hrs. at 120 rpm. and fltered by centrifugation at 10000 rpm at 5°C for 15 minutes. The clear centrifugate was used as crude enzyme extract.

Enzyme Activity: According to the method described by Balaban et al. (1991), enzyme assay of pectinase was done. The amount of acid produced was nutralised by 0.02(N) NaOH soln. PME activity is defined as milli equivalents of ester hydrolysed per minute per ml of enzyme.

10 ml of 0.1% (w/v) carbohydrate solution of glucose, maltose, lactose and amino acids were added in the medium and assay was done for PME activity at 30°C after 5 days of fermentation. Calculation was made considering total activity as 100%.

RESULTS AND DISCUSSION

In general any fermentation system depends on environmental and nutritional factors. Nutritional parameters such as carbohydrates, nitrogen, salts, amino acids, vitamins can increase the

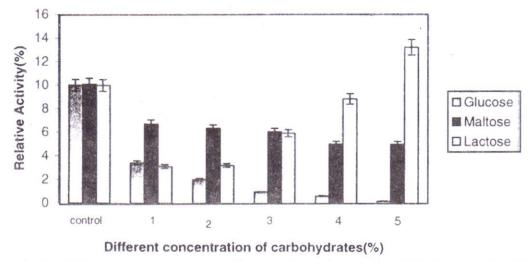


Fig. 1: A graph of effect of different carbohydrates on production of pectin methyl esterase by solid state fermentation. All the results were expressed in mean ± SD from n = 3

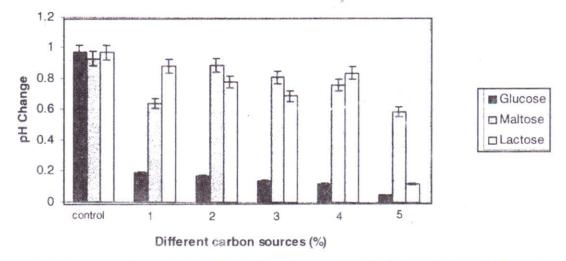


Fig. 2: A graph of effect of different carbohydrates on pH changes during production of pectin methyl esterase by solid state fermentation. All the results were expressed in mean ± SD form n = 3

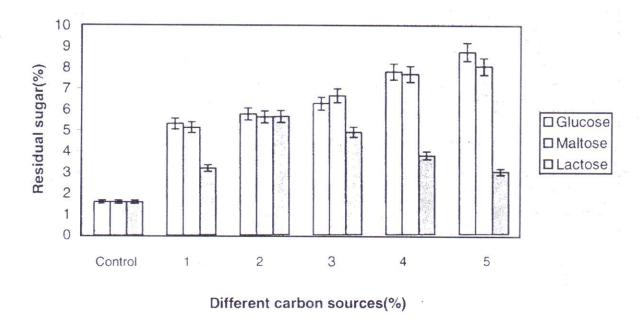


Fig. 3 : A graph on effect of different carbohydrates on residual sugar during production of pectin methyl esterase by solid state fermentation. All the results were expressed in mean \pm SD from n = 3

enzyme production in the medium if added at proper level. Earlier publication as reported by Sinha et al. (2009) revealed the environmental condition for optimum production of PME. The effects of addition of different carbon sources such as glucose, maltose and lactose on the production of PME in the SSF system are shown in Fig. 1. From relative activities (total activities considered as 100%), it was observed that 5% (w/v) lactose in the medium improved the enzyme production compared to control whereas 5% conc. of glucose supressed enzyme production highly. It was cleared that glucose and maltose had no effect on PME production. Only higher concentration of lactose (5%) had effect on PME production. Maximum pH changes occurred in the control set followed by medium containing 2% (w/v) maltose and lowest was medium containing 5% glucose and the pH of the crude enzyme was lower than that of the subtrate. It implied that some appreciable amount of acids was generated during fermentation. Residual sugar of the enzyme medium was measured by DNS methods. Highest residual sugar (total considerd as 100%) was found in the media containing 5% glucose followed by 5% maltose. Carbon sources was consumed by organism during fermentation as a source of food. A possible reason for this type of result may be due to the necessity of some

carbohydrates in particular concentration for the growth of the microorganism which are further utilised by the organism and produce desired enzyme.

The commercially available PME is accepted as a one of the most important food enzymes in the world. Different food wastes (wheat bran, orange peel, sugar cane bagasse etc.) can be used for production of PME. Optimum yield of PME was obtained at a concentration of 5% of lactose.

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REFERENCES

Balaban, M.O.; Arreola, A.G.; Marshall, M.; Peplow, A; Wei, C.I. and Cornell, J. 1991, Inactivation of pectinesterases in orange juice by supercritical carbon dioxide. *J. Food Sci.*: 56: 164-169.

- Blandins, A., Iqbalsyah, T. Pandialla, S. S., Cantere, D., and Webb, C. 2002. Polygalacturonase production by *Aspergillus awamori* on wheat in solid state formentation. *Appl. Microbial, Biotechnol.* **53.** 164-169.
- Kareen, S.O. and Adebowale, A.A. 2007. Clarification of orange juice by crude fungal pectinase from citrus peel. *Nigerian Food Journal*; 25: 130-137.
- Kunte, S. and Shastri, N.V. 1980. Studies on extracellular production of pectolytic enzymes by a strain of Alternaria alternata. Indian Journal of Microbiology 20: 211-215.
- Louboudy. S.S.; El-Gamal, M.S.; Ammar, M.S.; and Ali Moo. 2001 Microbial utilization of Eichhornia crassipes for Pectinases and Cellulases Enzymes production under solid state fermentation (SSF) conditions. Fourth Int. Sci

- conf. Science development and Environment. Faculty of Science, Al-Azhar University, Cairo, Egypt 27-29 March abstract. 32
- Martins, E.S.; Silva, D.; Da Silva, R., and Gomes, E. 2002. Solid state production of thermostable pectinases from thermophilic *Thermoascus aurantiacus*. Proc. Biochem. 37: 949-954.
- Nagodawithana, T.; Reed, G and Taylor, S. 1993 Carbohydrates In *Enzymes in Food Process*, Ed. Begelis R, Academic Press, London, 121-158.
- Sinha P.: Gayen, S. and Ghosh, Uma. 2009 Production of Pectinase from Agro-industrial easte using Aspergillus niger in Future of Food Biotechnology in India, National Institute of Technology, Durgapur, ISBN:978-81-907839-8-9, 205-211.