

## ABSTRACT

### **Effect of Locust Bean Gum Concentration, pH of Media, and Fermentation Temperature on the Production of Mannanase by *Bacillus subtilis* ATCC 6633**

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Mannanase is a hydrolase enzyme that works by hydrolyzing the  $\beta$ -1,4-manosidic bond between mannose and mannose. This enzyme has many uses in the pharmaceutical, pulp and paper, and bioethanol industries. Mannanase is obtained through a substrate containing many hemicelluloses such as galactomannan which is found in *Locust Bean* Gum. Potential sources of mannanase are microorganisms such as bacteria. *Bacillus subtilis* is a Gram-positive bacteria that produces the mannanolytic enzyme. The purpose of this study was to determine the effect of several factors on mannanase production such as substrate concentration, pH of media and fermentation temperature. The method used in this research is agar-well diffusion method. The tests were carried out in sequence starting from the variables of LBG concentration, media pH and the last fermentation temperature. This research was conducted in several steps, as follows: bacterial rejuvenation, making starter *Bacillus subtilis* ATCC 6633, making mannanase production media with various concentration, pH and fermentation temperature and mannanase activity test using *Congo Red*. The results showed that the mannanase activity emerged as a clear zone surrounding the bacterial colony. The diameter of the clear zone is converted to the mannanolytic index. The results showed that the most optimum mannanolytic index was formed at LBG concentration 0.25% with mannanolytic index  $2.89 \pm 0.09$ . The most optimal mannanolytic index was formed at pH 7 with mannanolytic index  $2,62 \pm 0,04$ . The most optimal mannanolytic index was formed at 37°C fermentation temperature with a mannanolytic index of  $2.62 \pm 0.04$ .

**Keywords** : Mannanase, Enzyme, *Bacillus subtilis* ATCC 6633, Locust Bean Gum, Production, Concentration, pH, Temperature.