

Optimization of Inoculum for α -amylase production by locally isolated *Bacillus Licheniformis*

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Single stage inocula are commonly used in most laboratory scale studies for the production of α -amylase by *Bacillus licheniformis*. Inocula at an age of 12 or 24h are commonly used as single stage inocula. This study was aimed to check the combined effect of two different aged inocula on the production of α -amylase using locally isolated *Bacillus licheniformis*. In this study α -amylase production using single and inoculum with two different aged inocula and the effect of different volume ratios of inoculum with two different aged inocula on α -amylase production were studied. Fermentation medium used contained (gL^{-1}) soluble starch, 2.0; peptone, 2.0; $(\text{NH}_4)_2\text{SO}_4$, 2.0; NaCl, 1.0; KH_2PO_4 , 1.0; K_2HPO_4 , 2.5; FeCl_3 , 0.01; MgCl_2 , 0.01; and CaCl_2 , 0.01 at pH 7.0. The effect of storage period of the *Bacillus licheniformis* in agar slant before inoculation was studied and 24h old bacteria gave the highest α -amylase activity ($40.2 \pm 0.42 \text{UmL}^{-1}$) at 48h.

When the age of the inoculum in the activation medium was 12h, highest α -amylase activity was produced ($45.8 \pm 0.51 \text{UmL}^{-1}$) at 48h, whereas the inoculum of 18h produced the α -amylase activity of $36.7 (\pm 0.18) \text{UmL}^{-1}$. When the inocula with two different ages (12h old, 5mL) and (18h old, 5mL) were mixed in equal volumes and used, α -amylase activity produced was $54.9 \pm (0.44) \text{UmL}^{-1}$ at 48h, which was 48% higher than the activity produced with 18h old inoculum and 19.5% higher than the activity produced with 12h old inoculum where the inocula were prepared from 24h old slant culture. When the effect of volume ratios of inocula prepared from two different aged inocula were studied on α -amylase production, the highest α -amylase activity ($53.3 \pm (0.34) \text{UmL}^{-1}$) was obtained with the inocula of 12h and 18h old in the volume ratio of 1:1 (5mL: 5mL). By optimizing the inoculum α -amylase production was improved by 1.52 fold.

Key words: Inocula, α -amylase, *Bacillus licheniformis*, activity, fermentation, culture.