Abstract

Esterase gene from Delftia sp. (cloned in pEam TA vector) was over expressed in E.coli (BL21). The maximum activity shown by enzyme was 50 U/min/ml. The optimum temperature and pH was 40°C and 8.0 respectively. The enzyme was not thermostable and showed pH stability at alkaline pH in the range of 7.0-9.0. This esterase exhibited maximum activity at 10 min incubation time and 150mM substrate concentration. Mg, Ni, Mn and K ions enhanced the enzyme activity 120, 104.2, 102 and 112% respectively. While in presence of Ba, Na and Ca it showed residual relative activity of showed 96.8%, 73 and 71% respectively. Cu, Co, Fe and Hg strongly inhibited the enzymatic activity by showing residual relative activity 1, 2, 14 and 5% respectively. EDTA strongly inhibited the enzymatic activity by decreasing the activity to 1%. But when used with Mg, Ni, and Mn the inhibitory effect was restored almost completely and activity recorded was 76.34, 92.42 and 84.11% respectively. SDS strongly inhibited the enzymatic activity by decreasing it to 2% in one hour and Tween 20 and Tween 80 stimulated activity 105 and 115% initially and after one hour showed 96.6% and 99.2% activity respectively. Triton X did not affect enzyme activity. Esterase activity was enhanced by IPTG even after 72 hours Lactose initially activity increased but after 48 and 72 hours it decreased as compared to IPTG. SDS page results indicated the purification of crude enzyme and also presence of an extra protein band (35.86 kDa) in transformed sample. V max, Km values were 55.2 U/ml/min, 26.5 mM and 54.8 U/ml/min and 25.24 mM from Lineweaver-Burke and Michaelis-Menton plots respectively.