Abstract

The aim of the study was to purify and characterize the protease enzyme from bacterial strains, *Bacillus subtilis* and *Bacillus amyloliquefaciens*. Protease enzyme was partially purified by using ammonium sulphate precipitation. The protease produced by *B. subtilis* was purified at 70% with 1.92 purification folds and 92% recovery. However, the protease enzyme produced by *B. amyloliquefaciens* was purified at 50% saturation with 1.74 folds and 91.2% recovery. The partially purified enzymes were subjected to SDS-PAGE to determine their molecular weights. The molecular weights of the proteases were 27kDa and 37kDa of *B. subtilis* and *B. amyloliquefaciens*, respectively. The optimum temperature of the both protease enzymes was of 45°C and both of the enzymes remained stable up to 70°C with 30% residual activity. The protease obtained from *B. subtilis* showed highest activity at pH 8.0 while the protease produced by *B. amyloliquefaciens* showed the highest activity at pH 9.0. The protease produced by *B. subtilis* was more stable with the increase of pH up to 12 as compared to the protease produced by *B. amyloliquefaciens*. Both of the enzymes showed the highest specificities for casein as substrate. $FeCl_3$ and $MgCl_2$ enhanced the activity of the proteases produced by *B. amyloliquefaciens* and *B. subtilis*, respectively. PMSF inhibited the activity of both the proteases, indicating the serine nature of the enzymes.