ABSTRACT

The present study deals with the enhanced production of cutinase (E.C. 3.1.174) by *Aspergiluus oryzae* ISL-9 under submerged fermentation and its immobilization on ZnO nanocrystals for improved stability. Submerged type fermentation was carried out to produce the enzyme while using the apple cutin as a substrate. The wild-type (ISL-9) was optimized for various parameters viz. substrate level (2%), sucrose concentration (1%), time of incubation (72 h), size of inoculum (3%) and pH (6). The activity was further enhanced by immobilization on ZnO nanocrystals. The UV-Vis spectrum showed an absorption band for both immobilized and free enzyme at 310 nm and 290 nm, respectively. The results of XRD showed the crystalline nature of ZnO nanocrystals. In FTIR, the observed band from 1120.6 cm^{-1}, 590.2 cm^{-1}, 601.7 cm^{-1} and 1105.2 cm^{-1} showed the stretching vibrations of nanocrystals of immobilized and free enzyme, respectively. The SEM images confirmed the particle size of the ZnO nanocrystals of 120-149 nm. The optimized parameters for the immobilization of cutinase on ZnO nanocrystals were enzyme concentration (0.8 ml for immobilized and free enzyme). ZnO nanocrystals concentration (30 mg) and procurement period (30 min for immobilized enzyme and 45 min for free enzyme). Further, the polyester hydrolysis and insecticide degradation potential of the cutinase produced by wild- type ISL-9 was also evaluated. Under the optimized conditions of 0.6 ml of enzyme concentration, 30 mg ZnO nanocrystals and 30 min incubation, an overall 16.236% of polyester was hydrolysed and 26.62% of insecticide was degraded by enzyme. The immobilized enzyme showed 1.19-fold and 1.35-fold increase in the enzyme activity for polyester and insecticide hydrolysis, respectively. The results are highly significant ($p \leq 0.05$) indicating the commercial availability of the process after the scale-up studies.