ABSTRACT

Immobilization of α-amylase on magnetic nanoparticles, especially magnetite nanoparticles, has been the main approach studied to make this enzyme, economically and industrially, more attractive. Covalent cross-linking of enzymes to magnetite (Fe_3O_4) nanoparticles (MNPs) is one of the useful enzyme immobilization methods which provides repeated use of the catalyst, facilitates enzyme separation from the reaction mixture, and improves biocatalysts stability. The aim of this study was to immobilize α-amylase from *Thermoanaerobacterium saccharolyticum* onto MNPs via covalent attachment using carbodiimide (CDI). MNPs were synthesized by the co-precipitation method. The size and the structure of the particles were characterized by X-ray diffraction (XRD) and scanning electron microscopy (SEM). The effects of different operational conditions of direct α-amylase binding on MNPs in the presence of CDI were investigated by using the shaking method. Fourier transform infrared spectroscopy was used to confirm the success of immobilization. The optimum conditions and catalytic properties of immobilized α-amylase were also evaluated. The efficiency of immobilization and the residual activity of the immobilized α-amylase were dependent on the mass ratio of MNPs: CDI: α-amylase and the immobilization temperature. The optimum pH for the free and immobilized amylase was 6, and 7.0 respectively. The free and immobilized α-amylase showed maximum activity at 50°C and 60°C respectively. The retained activity for free α-amylase after 30 storage days was 61% whereas it was 85% for the immobilized α-amylase. In repeated batch experiments, the immobilized α-amylase retained a residual activity of 41% after 10 repeated uses. It was found that immobilized α-amylase have better starch digestion capacity at different physicochemical conditions than that of free α-amylase. These results open new avenues for applying this MNP immobilized enzyme in different industrial sectors, notably in the paper and brewing industries.

Keywords: α-amylase, Magnetite Nanoparticles, Immobilized Enzyme, Stability.