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Multi-point covalent immobilization of β-galactosidase onto a polysiloxane-polyvinyl alcohol magnetic composite: an innovative biocatalyst for the production of prebiotic galacto-oligossacharides

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Beta-galactosidase (β-gal) is a dimeric enzyme with wide industrial applications and in the synthesis of oligosaccharides (GOS and FOS). β-gal from *Kluyveromyces lactis* was covalently immobilized onto a magnetic Polysiloxane-Polyvinyl Alcohol composite (mPOS-PVA), and the properties of the enzymatic derivative were duly evaluated. The mPOS-PVA is a hybrid inorganic-organic composite based on a polysiloxane and polyvinyl alcohol network obtained by sol-gel techniques and further magnetised with magnetite (Fe_3O_4), able to covalently retain proteins via reaction with glutaraldehyde. In this research effort, kinetic properties of both soluble and immobilized b-gal derivative onto mPOS-PVA were determined and duly compared. The enzymatic water insoluble derivative displayed the same optima pH (6.5) and T (50 °C) of the native enzyme. The apparent K_m and E_a for both soluble (7.377 ± 1.303 mM and 25.51 ± 8.72 Kj mol^{-1}) and immobilized enzyme derivative (7.841 ± 1.189 mM and 32.61 ± 5.82 Kj mol^{-1}) were quite similar. Furthermore, the mPOS-PVA β-gal derivative retained about half of the initial activity after being reutilized 20-times and incubated at 35 °C for 24 h, whereas the soluble enzyme lost all activity. Hence, mPOS-PVA proved to be an efficient support for β-gal immobilization and production of GOS at the industrial scale, due to both simplicity of matrix synthesis and immobilization protocol, and easy of removal from the reaction medium by simply applying a magnetic field.