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Carbon nanotubes molybdenum disulfide 3D nanocomposite as novel nanoscaffolds to immobilize *Lens culinaris* β-galactosidase (*Lsbgal*): Robust stability, reusability, and effective bioconversion of lactose in whey

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ABSTRACT

Multiwalled carbon nanotubes molybdenum disulfide 3D nanocomposite (MWCNT-MoS_2 NC) was successfully synthesized via eco-friendly hydrothermal method. The microstructural characterization of synthesized nanocomposite was carried out using different spectroscopic and microscopic techniques. Nanocomposite was activated using glutaraldehyde chemistry and used as a platform to immobilize *Lens culinaris β*-galactosidase (*Lsbgal*) which resulted in 93% of immobilization efficiency. Attachment of *Lsbgal* onto nanocomposite was confirmed by AFM, FE-SEM, FTIR, and CLSM. The nanobiocatalyst showed broadening in operational pH and temperature working range. Remarkable increase in thermal stability was observed as compared to soluble enzyme. Nanobiocatalyst showed outstanding increase in storage stability, retained 92% of residual activity over a period of 8 months. This offers good reusability as it retained ~50% residual activity up to 21 reuses and exhibited higher rate of lactose hydrolysis in whey. MWCNT-MoS_2 NC conjugated to biomolecules can serve as a potential platform for fabrication of lactose biosensor.

1. Introduction

Lactose sugar is one of the valuable assets of basic nutrients as it plays a crucial role in growth of animal during early stages of development. It influences the absorption of calcium, phosphorous, magnesium, and also helps in promoting the growth of acidic microflora of intestinal tract (Atkinson, Kratzer, & Stewart, 1957). However, when the consumption of lactose exceeds over the hydrolyzing capacities of *β*-galactosidase present in the gut, then symptom of lactose intolerance like abdominal pain, bloating, flatulence, vomiting, etc. arise (Shukla & Wierzbicki, 1975). The decrease in residual activity of *β*-galactosidase with the onset of ageing affects nearly 70% of world population (Adam, Rubio-Texeira, & Polaina, 2005). Thus, lactose intolerant people start avoiding milk and dairy products which later results in diseases like osteoporosis, cataract, and galactosemia.

In dairy industry *β*-galactosidase plays a crucial role in production of lactose reduced/free products by hydrolyzing lactose into glucose and galactose that improves the sweetness, and flavor of the dairy products (Richmond, Gray, & Stine, 1981). Nowadays, the production of Galacto-oligosaccharide (GOS) via transgalactosylation reaction by *β*-galactosidase has become popular because of its extraordinary nutraceutical and pharmaceutical properties that helps in the maintenance of microenvironment of the gut by restoring the growth of beneficial bacteria (Mahoney, 1998).

Besides milk, cheese whey is another major source of lactose sugar as it constitutes about 5–6% of the total whey. Earlier, González Siso (1995) had reported that manufacturing of 1 kg cheese produces 9 L of whey as waste. When the large quantity of lactose containing whey is dumped into water bodies, it increases the biological oxygen demand (BOD) and chemical oxygen demand (COD) (Geiger et al., 2016), leading major environmental concern. Lactose being a high calorie (energy) food cannot be dumped into waste stream.

Abbreviations: Lsbgal, *Lens culinaris β*-galactosidase; MWCNT-MoS_2 NC, Multiwalled carbon nanotubes molybdenum disulphide nanocomposite; NBCs, Nanobiocatalyst; FE-SEM, Field Emission-Scanning Electron Microscopy; TEM, Transmission Electron Microscopy; CLSM, Confocal Laser Scanning Microscopy; FTIR, Fourier Transformed Infrared Spectroscopy; AFM, Atomic Force Microscopy; RSM, Response Surface Methodology

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