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Purification and characterization of transglutaminase from Tropical tilapia (*Oreochromis niloticus*)

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Abstract

Transglutaminase (TGase) from Tropical tilapia (*Oreochromis niloticus*) was purified to electrophoretic homogeneity using successive chromatographies of DEAE-Sephacel, Sephacryl S-4 HR and HiTrap Heparin with a yield and purification-fold of 12.9% and 69.8, respectively. The molecular weight (MW) of the purified tilapia TGase was estimated to be 85 kDa using sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE). The isoelectric point (pI) of tilapia TGase was 6.53. Optimal temperature and optimal pH of tilapia TGase were 37–50 °C and 7.5, respectively. Optimal concentrations of $CaCl_2$ and dithiothreitol (DTT) were at 1.25 and 5 mM, respectively. The activity of TGase towards monodansylcadaverine (MDC) decreased as the NaCl concentration increased. Chelating agents, ethylenediaminetetraacetic acid (EDTA) and ethylene glycol-O,O′-bis(2-aminoethyl)-*N*,*N*,*N*′,*N*′-tetraacetic acid (EGTA), inhibited TGase activity. Tilapia TGase was strongly inactivated by ρ-chloromercuribenzoic acid (PCMB), *N*-ethylmaleimide (NEM), iodoacetamide (IAA), Cu^{2+}, and Zn^{2+}, suggesting a thiol group at the active site.

Keywords: Transglutaminase; Tilapia (*Oreochromis niloticus*); Purification