Production of the virus-like particles of nipah virus matrix protein in Pichia pastoris as diagnostic reagents

ABSTRACT

The matrix (M) protein of Nipah virus (NiV) is a peripheral protein that plays a vital role in the envelopment of nucleocapsid protein and acts as a bridge between the viral surface and the nucleocapsid proteins. The M protein is also proven to play an important role in production of virus-like particles (VLPs) and is essential for assembly and budding of NiV particles. The recombinant M protein produced in Escherichia coli assembled into VLPs in the absence of the viral surface proteins. However, the E. coli produced VLPs are smaller than the native virus particles. Therefore, the aims of this study were to produce NiV M protein in Pichia pastoris, to examine the structure of the VLPs formed, and to assess the potential of the VLPs as a diagnostic reagent. The M protein was successfully expressed in P. pastoris and was detected with anti-myc antibody using Western blotting. The VLPs formed by the recombinant M protein were purified with sucrose density gradient ultracentrifugation, high-performance liquid chromatography (HPLC), and Immobilized Metal Affinity Chromatography (IMAC). Immunogold staining and transmission electron microscopy confirmed that the M protein assembled into VLPs as large as 200 nm. ELISA revealed that the NiV M protein produced in P. pastoris reacted strongly with positive NiV sera demonstrating its potential as a diagnostic reagent.

Keyword: Nipah virus; Genetic engineering; Pichia pastoris; Virus-like particles; Diagnosis