

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