

## **Direct recovery of recombinant nucleocapsid protein of Nipah virus from unclarified Escherichia coli homogenate using hydrophobic interaction expanded bed adsorption chromatography**

### **ABSTRACT**

A direct recovery of recombinant nucleocapsid protein of Nipah virus (NCp-NiV) from crude Escherichia coli (E. coli) homogenate was developed successfully using a hydrophobic interaction expanded bed adsorption chromatography (HI-EBAC). The nucleic acids co-released with the recombinant protein have increased the viscosity of the E. coli homogenate, thus affected the axial mixing in the EBAC column. Hence, DNase was added to reduce the viscosity of feedstock prior to its loading into the EBAC column packed with the hydrophobic interaction chromatography (HIC) adsorbent. The addition of glycerol to the washing buffer has reduced the volume of washing buffer applied, and thus reduced the loss of the NCp-NiV during the washing stage. The influences of flow velocity, degree of bed expansion and viscosity of mobile phase on the adsorption efficiency of HI-EBAC were studied. The dynamic binding capacity at 10% breakthrough of 3.2 mg/g adsorbent was achieved at a linear flow velocity of 178 cm/h, bed expansion of two and feedstock viscosity of 3.4 mPa s. The adsorbed NCp-NiV was eluted with the buffer containing a step gradient of salt concentration. The purification of hydrophobic NCp-NiV using the HI-EBAC column has recovered 80% of NCp-NiV from unclarified E. coli homogenate with a purification factor of 12.5.

**Keyword:** Expanded bed adsorption, Hydrophobic interaction chromatography, Recombinant nucleocapsid protein, Nipah virus, Escherichia coli, Phenyl ligand