Oligomerization state of the multimerization domain of Nipah virus phosphoprotein

ABSTRACT

Oligomerization of the phospho (P) proteins of paramyxoviruses plays important roles in the viral genome replication and transcription. The multimerization domain of the P proteins (PMD) was found to be responsible and sufficient for the oligomerization process. However, the oligomeric status of Nipah virus (NiV) P protein is still unknown. Therefore, the aim of this study was to produce the PMD region of NiV in bacteria in order to determine the oligomeric status of the P protein. The NiV PMD region was cloned into pTrcHis2-TOPO vector and expressed in Escherichia coli. SDS-PAGE and Western blot analysis confirmed the protein expression upon IPTG induction at 37 °C for 5 h. Immobilized metal affinity chromatography (IMAC) purification was developed to purify the recombinant histidine-tagged NiV PMD which yielded a purity of 97%. Chemical cross-linking revealed that the PMD of NiV formed tetramer. The stability of the oligomer was studied with dynamic light scattering while mass spectrometry confirmed the exact molecular masses of the cross-linked products. Circular dichroism analysis revealed that the NiV PMD contained high -helical content (55%) suggesting the presence of a coiled coil structure.

Keyword: Nipah virus; Phosphoprotein; Multimerization domain; Oligomerization; Coiled coil structure