Epitope Presentation System Based on *Cucumber Mosaic Virus* Coat Protein Expressed from a *Potato Virus X* –based Vector

The *Cucumber mosaic virus* Ixora isolate (CMV) coat protein gene (CP) was placed under the transcriptional control of the duplicated subgenomic CP promoter of a *Potato virus X* (PVX)-based vector (Fig.1). *In vitro* RNA transcripts were inoculated onto *Nicotiana benthamiana* plants and recombinant CMV capsid proteins were identified on Western blots probed with CMV antibodies 5-7 days post-inoculation. PVX-produced CMV CP subunits were capable of assembling into virus-like particles (VLPs), which were visualized by electron microscopy (Fig.2). We further used the PVX/CMVCP system for transient expression of recombinant CMV CP constructs containing different neutralizing epitopes of *Newcastle Disease Virus* (NDV), economically important pathogen of poultry. NDV epitopes were engineered into the internal βH-βI (motif 5) loop of CMV CP. Both crude plant extracts and purified VLPs were immunoreactive with CMV antibodies as well as with epitope-specific antibodies to NDV, thus confirming the surface display of the engineered NDV epitopes. Our study demonstrates the potential of PVX/CMVCP as an expression tool and as a presentation system for promising epitopes. High-level heterologous expression of the CMV CP from PVX vector and production of compact, assembled VLPs may further contribute both to the development of vaccine/biomaterials delivery and epitope presentation systems as well as to the study of the biology of CMV, an economically important virus of many crops worldwide.

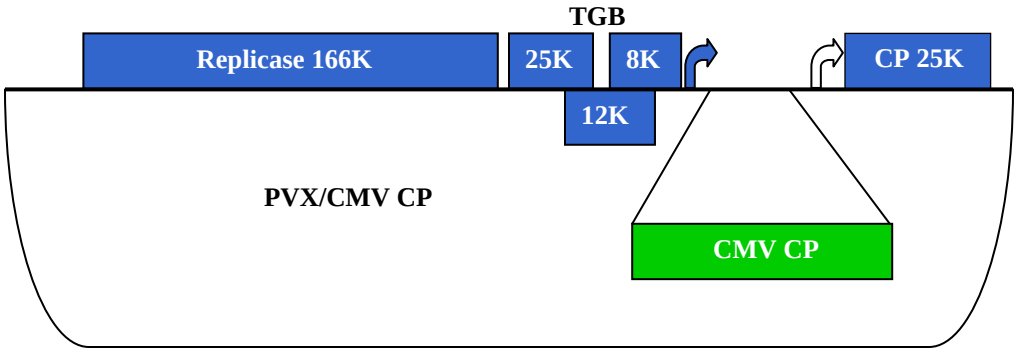

Fig.1. Schematic representation of a PVX vector containing CMV CP gene.

Closed arrow: duplicated PVX CP subgenomic promoter. Open arrow: PVX CP subgenomic promoter. TGB: triple gene block.

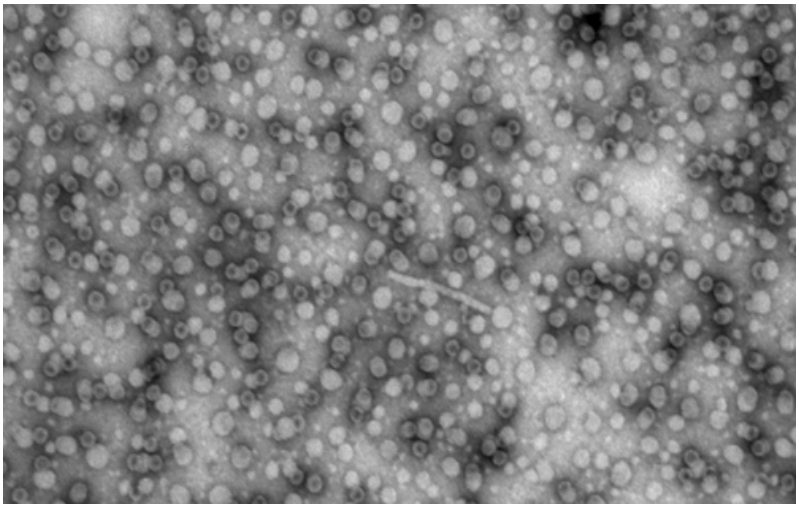

Fig.2. **Purified preparation of CMV VLPs derived from plants, infected with PVX/CMV-CP recombinant virus**

Natilla A., Hammond R.W., and Nemchinov L.G. 2006. Epitope presentation system based on Cucumber mosaic virus coat protein expressed from Potato virus X vector. Archives of Virology, Jul; 151(7):1373-86. Epub 2006 Feb 20.