PURIFICATION AND CHARACTERIZATION OF AN INFLUENZA VIRUS-LIKE PARTICLE VACCINE PRODUCED IN TRICHOPLUSIA NI LIVING BIOFACTORIES

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Trichoplusia ni pupae can serve as living biofactories for production of virus-like particle (VLP) vaccines through expression of only a few viral proteins such as hemagglutinin (HA) and matrix protein 1 (M1). Compared to other types of inactivated virus, split virus, LAIV and recombinant flu vaccines and their associated manufacturing methods, the use of T. *ni* for production of flu VLPs could potentially improve the safety, immunogenicity, and efficient and economic production of influenza vaccines. The goal of my work is to establish an effective method for purification of flu VLPs from T. *ni*. Extracts of T. *ni* pupae were prepared in phenylthiourea, PMSF, and formalin to prevent melanization and degradation of viral proteins. The extract was then differentially centrifuged at 1,500 × g and 11,000 × g, followed by a final discontinuous sucrose gradient (27-30- 60%) at 97,000 × g to obtain VLP particles. Samples at various steps of the purification process were analyzed using Coomassie blue stained SDS-PAGE, Western immunoblots with anti-HA and anti-M1 polyclonal antibodies, and transmission electron microscopy (TEM). Enrichment of the VLP proteins, HA and M1, was observed in successive steps of the purification process. TEM images showed that VLPs were indeed produced, and exhibited similar morphology to influenza viruses. These results demonstrate successful purification of a candidate VLP vaccine produced in T. *ni* pupae.