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Poster

Overexpression and purification of the CSFV-E2 recombinant protein for diagnostics from biofactories using biotechnological methods

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ABSTRACT

The production and purification of the E2 protein for develop of a diagnostic tool for detection of classical swine fever virus (CSFV) has raised interest by the current impossibility to distinguish between vaccinated and infected animals. The production of the recombinant E2 protein is important for the development of a diagnostic kit that allows the early detection of animals infected with the swine fever virus.

Bionaturis is developing a recombinant E2 molecule using the FlyLife platform. For the production, the E2 protein was cloned in recombinant baculovirus, which were used for protein overexpression by infecting Trichoplusia ni larvae. The addition of a histidine tail to the original protein is also a key question to facilitate E2 protein purification. By using SDS-PAGE and anti-CSFV-V8 Wester blot analyzes, a 44kDa size band of the E2 protein was observed as expected. The results indicated that overexpression efficiently took place, yielding a large amounts of the desired protein.

In conclusion, a protein extraction and purification protocol has been developed using affinity chromatography and denaturing conditions and a yield of 0.30 mg/larvae and 86% purity has been obtained. The use of a histidine tail to facilitate purification has allowed an increase in purity. Finally, a step of refolding the denatured protein has been carried out and its activity has been validated by an in vitro potency test by specific antibody recognition (ELISA).

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