

PROCESS INTENSIFICATION FOR PRODUCTION OF A PESTE DES PETITES RUMINANTS VIRUS (PPRV) VACCINE

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Peste des Petites Ruminants Virus (PPRV) is a highly contagious disease affecting small ruminants in Africa and Asian countries, with negative/significant economic impact. Aiming to eradicate the disease, a novel/scalable PPRV vaccine production process is clearly needed. Built upon work previously done at iBET, a new production process is herein proposed using Vero cells growing on microcarriers, serum-free medium (SFM) and stirred-tank bioreactors (STB). This includes a new method for cells detachment from microcarriers, and perfusion culture for reducing turnaround time.

The PPRV vaccine production process was developed in 2L STB (BIOSTAT DCU-3, Sartorius) using Nigeria 75/1 strain. Engineering correlations (e.g. shear stress and Eddy size) were used to optimize culture conditions. Vero cells were adapted to grow in ProVeroTM-1 SFM (Sartorius). A new enzymatic and mechanical method for in situ cell detachment from microcarriers was designed. PBS washing, TrypLE Select and trypsin inhibitor concentrations, and stirring rates were the variables explored. Perfusion culture was evaluated in 2L STB (equipped with spin-filter) in order to reduce seed-train preparation time. PPRV were clarified using depth filtration (Sartopure PP2, Sartorius).

Vero cells were adapted to ProVeroTM-1 SFM, reaching growth rates of 0.03 h⁻¹ (similar to serum-containing cultures). The new in situ cell detachment method was successfully implemented, with yields above 80%; no impact on cell re-attachment or virus productivity was observed. A two-fold increase in maximum cell concentration was obtained using perfusion when compared to batch culture. Combining the new in situ cell detachment method with perfusion culture will enable the scale-up to 20L STB directly from a 2L STB, surpassing the need for a mid-scale platform and thus reducing seed-train preparation time. The potential of depth filtration for PPRV clarification (upon microcarriers sedimentation) could be confirmed, with yields up to 90%. Process scalability will be validated at the 20L scale in Sartorius BIOSTAT C-Plus (using engineering correlations such as shear stress and Kolmogorov-Eddy size as scale-up criteria) by comparing cells growth, metabolic and PPRV production kinetics to those achieved in 2L STB.

In conclusion, the novel/scalable vaccine production process herein proposed has potential to assist the upcoming vaccination program for eradication of PPRV disease.