P7 - Expression of African swine fever virus proteins in a live-attenuated pseudorabies virus vector

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African swine fever virus (ASFV) causes a lethal disease of domestic pigs and wild boar. It is endemic in Sub-Saharan Africa, and currently spreading throughout Europe and Asia. Up to now, no vaccines are available, and little is known about immunogenicity of the predicted 150-160 different ASFV proteins.
Therefore, we expressed several of them in the pseudorabies virus vaccine strain Bartha (PrV-Ba), using an efficient mutagenesis system based on CRISPR/Cas9-supported rescue of a defective, GFP-expressing plasmid clone (Hübner et al., 2018, J Virol Methods 262:38-47). Transgenes were inserted at the nonessential glycoprotein G locus (US4) of PrV-Ba. Optionally the upstream protein kinase gene US3 was additionally deleted to improve attenuation. Protein expression under control of the strong CAG promoter was optimized by codon adaptation. Up to now, 12 ASFV gene products, including membrane (p12, p22, p54, pE199L, p285L, CD2v), capsid (p72), other structural (p11.5, p30, pA104R), as well as abundant nonstructural (pK145R, pB602L) proteins were individually expressed. Since pB602L is a chaperone required for proper processing of p72, both proteins, spaced by the “self-cleaving” teschovirus 2A peptide, were also co-expressed.
The mutations did not inhibit PrV replication in cell culture, and abundant transgene expression was demonstrated using monospecific antisera. In immunized pigs, the tested p30, p22 and pE199L mutants induced ASFV-specific antibodies. Thus, our PrV-based vector system allows the analysis of immune responses to single or multiple ASFV proteins in the context of a nonlethal virus infection of swine, and might provide a platform for vaccine development.

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