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Generation and characterization of attenuated PEDV vaccine candidates

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Key words
porcine epidemic diarrhea (PED), vaccine, live attenuated vaccine, reverse genetics, coronavirus

Aim of the study
Based on the results and developed techniques of the FSVO-funded project 1.15.05 (Establishing preparedness for PEDV outbreaks) we will generate and characterize attenuated candidate PEDV vaccine strains. The parental strain will be provided by our industrial partner and will be attenuated and cloned (reverse genetics) at the IVI. Subsequently 3 PEDV vaccine candidate strains with different degrees of attenuation will be produced for in vivo experiments. The characterization of the candidate vaccine strains regarding attenuation and immunogenicity will be performed at the facilities of the industrial partner.

Material and methods
Based on the PEDV genome sequences of the parental strains we have used transformation-associated recombination (TAR) in yeast to clone full-length cDNAs of several PEDV genomes. Rescue of recombinant PEDV was performed using the established T7-polymerase-mediated in vitro transcription and electroporation of the resulting RNA into Vero cells. Virus growth was done in presence of trypsin in the cell culture medium to facilitate spread of PEDV in cell culture. Assessment of the vaccine candidates will be performed at the animal facilities of the industrial partner.

Results and significance
Several PEDV vaccine candidates have been successfully cloned using the TAR cloning in yeast. The clones have been sequence verified by NGS. Several clones have also been equipped with a reporter gene (green fluorescent protein gene) to monitor and facilitate virus replication and production. The obtained viruses will be assessed concerning their attenuation *in vivo* and subsequently the efficacy to protect from wild-type virus infection will be assessed. These studies have been significantly delayed due to the SARS-CoV-2 pandemic that precluded shipment of live viruses.
We expect that several of the produced vaccine candidates will show sufficient attenuation to move forward to assess the vaccine efficacy. The reverse genetic system to produce the vaccine candidates is versatile and can be applied to modify and adapt the vaccine. This will allow to update the vaccine in case novel strains may emerge. It is also possible to extend this approach to other coronaviruses that cause severe disease in pigs.

Publications, posters and presentations
A publication is planned after the in vivo experiments have been performed.

Project 1.19.07:

Project duration *1.1.2020 – 31.7.2022*