DTU Library

Engineering a CTL-Tailored Replicon RNA Vaccine against PRRSV

Welner, Simon; Werder, Simea; Nielsen, Morten; Lund, Ole; Gerber, Markus; Liniger, Matthias; Ruggli, Nicolas; Summerfield, Artur; Larsen, Lars Erik; Jungersen, Gregers

Publication date:
2016

Document Version
Peer reviewed version

[Link back to DTU Orbit](#)

Session: Concurrent Session 3 - Vaccines & Adjuvants

Engineering a CTL-Tailored Replicon RNA Vaccine against PRRSV (#61)

Simon Welner [1] , Simea Werder [2] , Morten Nielsen [3 4] , Ole Lund [4] , Markus Gerber [2] , Matthias Liniger [2] , Nicolas Ruggli [2] , Artur Summerfield [2] , Lars E Larsen [1] , **Gregers Jungersen** [1]

1. *National Veterinary Institute, Technical University of Denmark, Frederiksberg, Denmark*
2. *Institute of Virology and Immunology, Mittelhäusern, Switzerland*
3. *Instituto de Investigaciones Biotecnológicas, Universidad Nacional de San Martín, Buenos Aires, Argentina*
4. *Department of Systems Biology, Technical University of Denmark, Lyngby, Denmark*

The development of vaccines against porcine reproductive and respiratory syndrome virus (PRRSV) has been hampered by the high mutation rate and the multiple immunoevasive strategies of the virus.

With the overall aim of designing a broad coverage vaccine that induces an effective CTL response against PRRSV, we have used a bioinformatics approach to identify common PRRSV type 2 epitopes predicted to react broadly with predominant swine MHC (SLA) alleles.

All possible 9- and 10-mer peptides derived from 104 wild-type strains were analyzed *in silico* for their predicted binding affinity to 3 common SLA class I alleles and ranked according to genomic conservation and SLA binding coverage. Of the 53 top-ranked peptides, 33 were verified *in vitro* as high affinity binders. Polyepitope gene cassettes of these peptides, flanked by an upstream ubiquitin sequence and a downstream FLAG tag, were cloned into a classical swine fever virus (CSFV)-derived replicon vector. Virus replicon particles (VRP) were rescued by transfection of a complementing cell line with replicon RNA. Polyepitope expression and subsequent proteasomal degradation was confirmed indirectly by increased FLAG-tagged protein detection in the presence of a proteasome inhibitor.

Finally, a vaccination-challenge experiment using 18 SLA-matched pigs is currently being conducted until July 2016 in which a test group and a control group are being vaccinated twice with VRPs expressing PRRSV epitopes and non-sense control epitopes, respectively, before challenged with live wild type PRRSV. The induced epitope specific cell-mediated immune responses are being monitored by ELISPOT, flow cytometry and cytotoxicity assays, and the degree of protection against infection will be characterized by qPCR and antibody analysis. The results will be available for IVIS.

This study exemplifies how bioinformatics epitope prediction, recombinant SLA molecules and RNA virus replicon design can be used to engineer a replicating non-propagating vaccine tailored to deliver conserved and immunogenic CTL epitopes.

Close View