Novel recombinant classical swine fever vaccines based on replicon-technology and vaccine delivery

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Key words
Pig, classical swine fever virus, vaccine, virus replicon particle, interferon-α/β, immune response

Aim of the study
Potent classical swine fever virus (CSFV) vaccines based on live attenuated virus are available. However, live virus vaccines raise safety concerns and do not allow serological differentiation of infected from vaccinated animals (DIVA). Novel CSFV replicon particles (CSF-VRP) lacking the Erns gene were developed earlier but had only limited efficacy (BVET projects 1.01.15 and 1.04.02). The aim of the present work was to explore the possibilities of improving the immunogenic properties of the CSF-VRP vaccine.

Material and methods
The CSF-VRP were modified to induce interferon-α/β (IFN-α/β) and to co-express granulocyte-macrophage colony stimulating factor (GM-CSF) as a potential genetic adjuvant. The long-term replication of the CSF-VRP in presence of IFN-α/β was evaluated in cell culture. To determine the immunogenicity of IFN-α/β–inducing VRP, the recall humoral and cellular immune responses were measured after antigen-specific stimulation of ex vivo peripheral blood mononuclear cells (PBMC) from CSFV-immune pigs. Finally, the primary immune responses and the contribution of IFN-α/β secretion and GM-CSF expression to protection against a challenge infection was evaluated in a vaccination experiment in pigs.

Results and significance
The capacity of modified CSF-VRP to induce IFN-α/β and to co-express foreign genes of interest such as luciferase and GM-CSF was demonstrated. Importantly, the IFN-α/β-inducing replicons delivered from CSF-VRP persisted in cells for several cell culture passages, independently of the antiviral pressure of IFN-α/β. The replication of the replicon RNA was essential for the VRP to induce a specific B-cell immune response in vivo, since UV-inactivated CSF-VRP were not immunogenic, even after a booster immunization. In the ex vivo PBMC-based assays, the CSF-VRP that were mutated to induce IFN-α/β mediated enhanced B- and T-lymphocyte secondary responses. In vivo, all modified CSF-VRP protected from clinical symptoms apart from transient fever, and were indistinguishable from the parent CSF-VRP in terms of B- and T-cell responses. When compared with the unmodified CSF-VRP, immunization with the IFN-α/β-inducing and GM-CSF-expressing VRP showed slightly reduced viral RNA levels in the blood of animals following challenge infection, indicating enhanced immunogenicity. Taken together, this study shows that CSF-VRP represent a safe vaccine capable of co-expressing bioactive proteins of interest. CSF-VRP have the advantage over inactivated and subunit CSFV vaccines in offering cycles of replication providing more antigen and prolonged expression of protein and thus better protection.

Publications, posters and presentations
Suter, R.; Summerfield, A.; Tohmann-Harwood, L.J.; McCullough, K.C.; Tratschin, J.-D.; Ruggli, N. (2010) Immunogenic and replicative properties of classical swine fever virus replicon particles modified to induce IFN-α/β and carry foreign genes. Vaccine. 4;29(7): 1491-503.
Suter, R. (2010) Classical swine fever virus replicon particles: a versatile and robust system for vaccine and gene expression applications. PhD thesis, GCB of the University of Berne, in preparation.
Suter, R. a total of five posters and oral presentations at national and international conferences.

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