Characterization of T-cell memory responses associated with control of CBPP and identification of immuno-active proteins of *Mmm*SC

P. Totté, A. Mather, L. Reslan, V. Rodrigues, E. Viley, J. Frey, D. Du Plessis, L. Dedieu.

A better understanding of the basis of protective immunity and immunopathologic reactions associated with contagious bovine pleuropneumonia (CBPP), caused by Mycoplasma mycoides subsp. mycoides small colony (*MmmSC*), is needed to develop better vaccines. Here we report on the characterization of T-cell memory responses associated with protective immunity in cattle and their use thereof to identify immuno-active proteins of *Mmm*SC. Both effector memory (em) and central memory (cm)-like *Mmm*SC specific CD4+ T cells were present ex vivo and capable of proliferation and IFN-γ production upon re-stimulation in vitro with inactivated *Mmm*SC. It has been shown in mice and humans that priming of T(cm) is required for long-lived protective immunity, which is also an essential requirement of any improved vaccine against CBPP. Pre-sensitized T-cells were used to screen recombinant proteins of *Mmm*SC produced as his-tagged proteins and purified by chromatography. Three of them were found to trigger the proliferation of both CD4+ T(em) and T(cm) and induce the production of IFN-γ: lipoprotein A (LppA), a glucose-specific component of the PTS system (ptsG) and to a lesser extent a substrate-binding component of the ABC transporter (ABC). In contrast, although lppQ had no effect on its own, it strongly inhibited the proliferation of CD4 in response to *Mmm*SC by a mechanism apparently independent of direct cytotoxicity. In conclusion, the T-cell reagents and methodology described in this study will help identify immuno-protective antigens of *Mmm*SC that have potential for the development of a subunit vaccine against CBPP. In addition, these cellular tools can also be used to screen for immuno-inhibitory proteins whose corresponding genes could be targets for deletion/inactivation in the aim of developing improved attenuated vaccines.