Cloning, Heterologous Expression and Purification of the SpaA Antigenic Region, a Candidate for a Subunit Vaccine Against Swine Erysipelas

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Swine erysipelas is one of the major diseases in swine-producing areas, causing great economic losses worldwide. Both attenuated and killed vaccines are available to prevent the disease, but the exposure of the animals to vaccines containing the whole microorganism *Erysipelothrix rhusiopathiae* can aggravate arthritic problems. In this context, many efforts have been done to develop a subunit vaccine that would be safer and able to prevent this disease. Here we report the cloning of the 1026 pb fragment codifying for the antigenic region of the *E. rhusiopathiae* SpaA protein, the heterologous expression in *E. coli* and the purification of the recombinant protein by affinity chromatography. The fragment was amplified by PCR from the *E. rhusiopathiae* chromosomal DNA and cloned into pGEM-T vector. After sequencing, the *spa*A fragment was cloned into the pET28a expression vector and used to transform *E. coli* BL21(DE3). The recombinant SpaA (rSpaA) protein was over-produced as inclusion bodies, which were washed with Triton-X100 and solubilized in urea for purification in denaturation conditions, using a Ni^{2+} sepharose column. Antibodies against the recombinant protein will be produced in mouse and tested for reaction with the *E. rhusiopathiae* SpaA surface protein by *Western blot*. The potentiality of the purified rSpaA as a subunit vaccine to prevent swine erysipelas will be evaluated in murine models in the next steps of this work.

Key-words: *Erysipelothrix rhusiopathiae*, recombinant protein, swine erysipelas, vaccine, SpaA

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