RESEARCH ABSTRACT

SWINE HEALTH

Title: Development of fluorescent recombinant antibodies to detect African swine fever virus in tissue samples and infected cells. **NPB #11-022**

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Industry Summary:

The present project pretends to develop new reagents to solve an important gap in the African swine fever virus (ASFV) diagnosis. This virus is nowadays a real threat for Europe and, potentially, may spread to Asia or even other continents where the virus may produce a tremendous impact in the pig production.

The serology of ASF has been resolved by the use of recombinant proteins in the diagnostic tests. These proteins have been validated and represent a better alternative to antigens obtained from infected cells (use of infectious virus for their generation). Recombinant antigens showed an improved sensitivity and specificity for antibody detection in chronically infected or innaparent carrier pigs and allowed the standardization of reagents production and tests interpretation.

However, for the control or eradication of this important swine disease (no vaccine is available) is necessary to combine serology surveys with techniques for virus detection in samples of potentially infected pigs. Rapid detection means a minimization of disease spread risks to other animals or farms. Actually, the virus detection has to be done by PCR analysis (detection of viral DNA). This methodology detects accurately the virus presence in pig tissues, but needs a reference confirmatory technique because frequent false positive results, specially in laboratories with a reduced training level. The OIE recommend the virus isolation and the virus detection in animal

tissues by antibody immunofluorescence. There are not available commercial universal reagents (antibodies) to carry out the immunofluorescence tests on tissue cuts or tissue explants. Additionally, the virus has to be isolated in primary pig macrophage cultures and, frequently, it takes several days and even weeks, depending of the virus titers in body fluids or organs, before the observation of the characteristic cytopathic effect or the haemadsorption reaction.

The main objective of this project was to develop recombinant antibodies that could be used as reagent for sensitive detection of the virus in biological samples or infected cell cultures used for virus isolation. These antibodies, labeled with fluorescent molecules, will allow the virus detection using different technologies. These recombinant antibodies will avoid the use of sera from infected animals (potential risk of virus contaminations) or the use of monoclonal antibodies directed to variable epitopes of the virus that could fail in the detection of any specific virus strain. Antibodies would be produced by a cost-efficient system based on baculovirus vectors (a common system to produce biologics) and insect larva (living biofactories) instead insect cells. The larva system, only used for the moment by a reduced number of companies and research laboratories, is one of the most efficient and cost-effective system to produce any recombinant protein. These reagents (recombinant labeled antibodies) could be sent, without any risk, to reference diagnostic laboratories and would facilitate the standardization of results, independently of the expertise of profesionals in ASF diagnosis. The limitation of the source of these antibodies would not be a problem for diagnostic laboratories in contrast to the limited source of antibodies obtained from immunized or naturally infected pigs.

The conducted research during the granted 1 year project has generated different recombinant antibodies which are able to accurately detect ASFV in cell cultures. These antibodies were generated in insect larvae (IBES technology) with excellent productivities. Those useful reagents will be tested during the next months in samples from experimentally infected pigs to certify their sensitivity in virus detection and will also be tested in diagnostic laboratories from endemic regions (South Africa and Russia). Once those reagents were validated, those will be transferred to a company for their commercialization in any potentially affected country, including USA. Results obtained will also be published during the next months to disseminate the scientific information to the veterinary scientific community.

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