Short-term mission scientific report

STSM theme/name: Epidemiology, risk assessment and diagnosis in ASF

Host institutes:

INGENASA (C/ Hermanos García Noblejas 39, 28037, Madrid, Spain).
UCM (Facultad de Veterinaria. Universidad Complutense de Madrid. Avenida Puerta de Hierro s/n 28040 Madrid, Spain);
INIA (Ctra Algete-El casar s/n. Valdeolmos, Madrid, Spain);

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Period of research stay: 1.02.2018 – 30.04.2018

Grantee: PhD student Svetlana Konnova

Purpose of the visit: learning different diagnostic techniques used in the laboratory for ASFV diagnosis and study different methods of epidemiology analysis and risk assessment.

Diagnosis:

Diagnosis part of short – term mission were in INGENASA and UCM. We analyzed an experimental serum collection in order to detect the presence of IgG and IgM antibodies and antigens, as an indirect indicators of infection with different types **ELISA**, frequently used method in surveillance programs, such as OIE ELISA (indirect test 2013), indirect ELISA (prototype OIE 2017) for detection antibodies against ASFv. Also we performed blocking immunoenzimatic assay for detection of antibodies of African swine fever virus (ASFV) in porcine serum (INGENASA kit) and ASFV ELISAs against an early antigen of the virus for Ig G and IgM detection and carefully analysed and compare obtained results.

On the other hand, I studied rapid and easy to perform tests, such as **Lateral Flow Assay (LFA)**, usually used for antibody/antigen detection in serum/blood samples. The test is based on the use of a MAb against VP72 protein of ASFV, the major viral capsid protein and highly immunogenic. This method has been proven to be very useful as a pen-side test in field conditions, in order to have a rapid and qualitative diagnosis. In laboratory we performed all stages of test – producing, from preparing of labeled microspheres, which are covalently linked to a control protein, and preparation of chromatographic strips to the corresponding antigen/antibody and interpretation of results. By learning these two techniques I

had a wide overview of ASF diagnosis and more accurate knowledge for interpretation of diagnosis data.

Also we used Luminex for ASF-diagnosis by **X-MAP technology** to investigate up to 50 different antigens in one sample. Systems using xMAP Technology perform discrete assays on the surface of color coded beads known as microspheres, which are then read in a compact analyzer. Using multiple lasers or LEDs and high-speed digital-signal processors, the analyzer reads multiplex assay results by reporting the reactions occurring on each individual microsphere.

Different types of conventional and real-time **PCR** also used for the molecular diagnosis ASF. During my short – term mission I performed Taq –Man PCR with isolated samples (Protocol by King et al), Taq – Man PCR UPL, SYBR Green PCR and compared obtained results of analysis.

Also I visited reference laboratory in UCM and listen a lecture about organisation of BSL3 laboratory.

Epidemiology and risk assessment:

Epidemiological part of STSm included training in epidemiological methods to improve surveillance and preventive measures against ASF.
I studied how to collect data correctly in Excel tables for future analysis and what factors are crucial for epidemiological investigations. We used for analysis data on ASF outbreaks in domestic pigs and wild boar in the Saratov region for the last 7 years obtained from the Department of Veterinary (Saratov region, Russia) and The Federal Service for Veterinary and Phytosanitary Surveillance, to describe the situation and main causes and mechanisms of the disease spread in the region.
We used Spatial analysis to analyze this data:

1) Distance between wild boar and domestic pig notifications using point distance tool (Arc GIS 10.3 Analysis tool).
2) The distributional trend and spreading dynamic of ASF notifications was assessed using the Standard Deviational Ellipse (SDE) by location and by location pondered by pig census. SDE creates standard deviational ellipses to summarize the spatial characteristics of geographic features (central tendency, dispersion, and directional trends) helping to understand the spatio-temporal patterns of the disease (Ward et al 2008). This analysis was performed using spatial statistics toolbox of ArcGIS 10.3.1 ESRI Inc. TM (ESRI, Redlands, CA, USA).

Great experience in using Arc GIS 10.3, under the guidance of competent specialists, allowed me to prepare and perform poster presentation on GARA meeting, Italy, 11-13 of April 2018 and take first prize in nomination «Best Poster Presentation from early Career Scientist». By the results, obtained during this Short Term mission I with colleagues from CISA-INIA Ana de la Torre, Irene Iglesias

and Marta Martinez prepared the draft of paper «African swine fever in Saratov region in 2011-2017, Russian Federation», which we are going to publish until the end of the year.

Personal experiences:

The short-term mission has allowed me to broaden my horizon in terms of both practical hands-on work as well as personal connections. It has been useful to work in another laboratory and seeing how they organize their work from the small differences in lab procedures to experiencing new techniques. In addition, it has been exciting to get to know new people and interact both in a lab and office setting plus in more informal surroundings after normal working hours. It has been good to discuss my future researches with scientists that can provide new inputs and ideas. The short-term mission has given me an excellent platform for networking, and to achieve results that otherwise would have taken a lot longer to obtain. I believe that a solid network is essential for a scientific career where co-operation between other scientists within your field is necessary and that has definitely been achieved.