International Scientific Working Group

ISWG update October 31st, 2018

Ongoing projects, regular and special

Regular projects

NIZO project (1601) – Determination of the effect of different preserving conditions on the survival of common contaminants of sheep casings

The final step in this project is completing the manuscript for publication as a scientific paper. When available, the expected EXCEL-based prediction model will be made available, together with a clear instruction on its use and a guideline for appropriate sampling.

Some difficulties have arisen in getting the scientific paper accepted for publication. Work is in progress to complete it. However, this will result in a delay in availability of the afore-mentioned outcome. The project is expected to be completed in the second quarter of 2019.

WBVR project (1602) – on inactivation of animal viruses and bacteria in casings

This extensive project is developed to include all contagious animal diseases with a known relevance to international casing trade and to determine if the standard treatment of salting casings or variations influencing time, temperature and pH will result in a sufficient inactivation of these diseases. In the past similar studies have already been done on e.g. foot-and-mouth disease and classical swine fever in experimentally infected animals. The aim now is not only to include more diseases and treatment variations, but also to reduce the number of animals needed by developing a suitable and validated non-animal model. This will make it easier in any future experiments to generate new results in case this is required.

- 2016, completion of the initial literature study on the inactivation of these diseases;
- 2017, completion of the subsequent pilot project, which focused on how / where to take samples from the intestines in order to ensure correct measurements. An important conclusion from this pilot project was to use full thickness of the intestine to use for your samples and not use cleaned casings. It was deemed the best approach to develop a worst-case scenario project design. The first manuscript, based on the experiments described above is in progress;
- 2018, completion of phase 2, focusing on the inactivation of classical swine fever virus in porcine intestines after treatment. Samples taken from a live-animal experiment. Results show that standard salting with NaCl for at least 45 days at temperatures over 12 °C are effective in the inactivation of CSFV in hog casings. With this prolonged exposure the application of P-salt would not be necessary. The subsequent project phase will focus on the validation of the non-animal (3D collagen) model by confirming the results of the live animal studies. After successful completion, all selected porcine diseases will then be tested in the 3D collagen model as phase four of this project.
- 2018, completion of the validation study of the 3D collagen model. Report pending.
- October 2018: regular WBVR project is put on hold due to various African swine fever virus (ASFV) outbreaks in China, Europe and the subsequent necessity to develop and execute a priority project on the inactivation of ASFV in a live-animal study.

International Scientific Working Group

Special project

WBVR project (1801) – Study to inactivation of ASFV in intestines of pigs

In preparation of this specific high priority project, extensive discussions were held with the WBVR on study design, size and scope, taking into account all relevant comments made by the ISWG delegates and industry members, resulting in a comprehensive project.
Relevant arguments included are:

- Confidence in acceptance of the outcome; the 2011 WBVR study (Wieringa et al., 2011), with the development and validation of the 3D collagen model as a replacement for live-animal experiments, was met by some hesitation from competent authorities. Due to the fact that the 3D model included results on FMDV and CSFV inactivation, which are were confirmed by previous live-animal studies, the results on ASFV inactivation were not challenged but not deemed sufficiently extensive to allow for acceptation without some discussion.
  However, the World Organisation for Animal Health (OIE) did already accept the outcome of the 2011 study and included a specific article in its Terrestrial Animal Health Code on the inactivation of ASFV in hog casings.

Article 15.1.23.
Procedures for the inactivation of ASFV in casings of pigs For the inactivation of ASFV in *casings* of pigs, the following procedures should be used: treating for at least 30 days either with dry salt (NaCl) or with saturated brine (Aw < 0.80), or with phosphate supplemented dry salt containing 86.5% NaCl, 10.7% Na_2HPO_4 and 2.8% Na_3PO_4(weight/weight/weight) at a temperature of 12°C or above.

  This article has provided the natural casing industry with a concrete argument and reference point. Unfortunately, some OIE members consider the Code articles as guidelines and not as formal basis to develop their countries' import requirements.
  As a result, a new live-animal study was developed to (re)confirm the efficacy of salting hog casings to reduce the import risk of AFSV to an acceptably low / negligible level.

- Study design: the aim of the study is to provide an answer on the inactivation of ASFV, applicable for the entire intestinal tract, including all parts that are used for casing production (runners, chitterlings fat ends etc.). To this end, four pigs will be infected, after the clinical onset of ASFV the animals (fever, specific symptoms, high virus blood titre) will be euthanised and samples taken from the intestinal tract. From a worst-case scenario perspective, manure-stripped small intestines will be used, with the mucosa included, to provide the actual samples (97 per animal). Subsequently the samples will be exposed to NaCl, P-salt or no salt at all, at different temperatures (4, 12, 20 °C) over a period of 60 days. After treatment, these samples are analysed to show how much virus is still present that can cause an infection. In addition, along the entire length of the intestinal tract (stomach to rectum), samples are taken to determine the actual virus concentrations before inactivation. This way we can determine if or how the virus concentrations can differ along the entire length of the intestinal tract.

- Sampling & testing: at days 2, 4, 7, 14, 21, 30, 45 and 60 the samples will be harvested and tested to determine the amount of virus present after the inactivation (Phase 2B – see below). Contrary to previous studies, the actual testing will take place shortly after the day of harvesting and not after the period of 60 days is completed. This means that if no virus can

be detected early on in the experiment, the remaining part need not be done. This also means that costs can be saved at that point in the experiment.

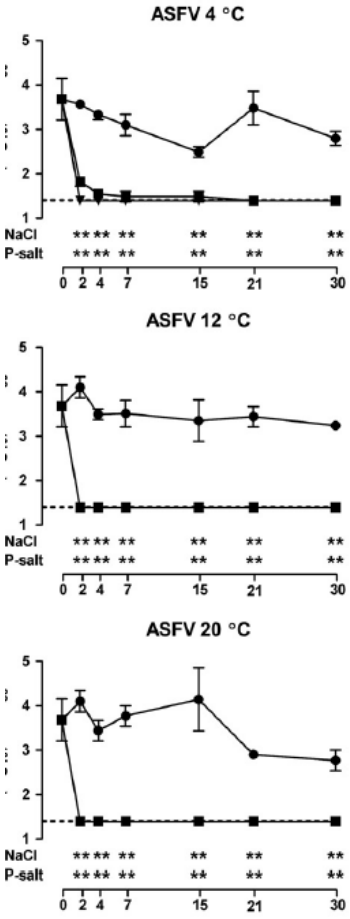
- Expected outcome: the 2011 study indicates how ASFV is inactivated over time when exposed to NaCl, P-salt at different temperatures. It is therefore expected that the live-animal experiment will have a similar result.

- Expected costs: based on the project's description the costs are divided over 4 different phases.

Phase	Net price (exc. VAT)
1	€ 92,990 (animal experiment)
2A	€ 107,256 (lab tests)
2B	€ 107,256 (lab tests)
3	€ 24,433 (scientific paper)
Total	€ 331,935

Phase 2B can vary as described above, depending on the number of samples that need to be analysed. Subsequently, the costs for this particular part of the project can be reduced.

- Planning: the project is scheduled to start at the beginning of November. Due to the quick start of the actual virus testing of the samples, preliminary results may be expected before the end of the year (2018). With the report and scientific manuscript finished in March 2019, a published paper may be possible by the end of Q2 2019. However, this is an indication only.



- As this is a special, high priority project, done to protect the interests of the global casing industry, the funding of this project is subject to additional financial support by the INSCA company members and association members. To this purpose a member assessment will be made and national and regional associations asked for their support.