Schmallenberg virus (SBV) - Consortium
Technical and scientific studies – final report March 2014

Five-country consortium of veterinary research institutes:

• Animal Health and Veterinary Laboratories Agency (AHVLA), United Kingdom;
• Veterinary and Agrochemical Research Centre (VAR-CODA-CERVA), Belgium;
• Friedrich-Loeffler-Institut (FLI), Germany;
• L'Agence nationale chargée de la sécurité sanitaire de l'alimentation, de l'environnement et du travail (ANSES), France;
• Central Veterinary Research Institute of Wageningen University and Research (CVI), The Netherlands (coordinator)
Studies of the Friedrich-Loeffler-Institut as partner of the consortium

- Studies of SBV pathogenesis in pregnant animals (cattle)
- Studies of diagnosis and infectivity of SBV-RNA positive bovine semen
  - Validation of diagnostic methods and German field study
  - Experimental infection of cattle and mice with SBV-RNA-positive semen
  - German and European method trial to evaluate the proficiency of methods for SBV-RNA detection in bovine semen and other matrices
- Studies of SBV pathogenesis in different mammalian host species
  - Transmission of SBV during winter
  - Dynamics of SBV infection in a cattle herd
  - Pathogenesis of SBV isolates in cattle
  - Pathogenesis of a German SBV isolate collected from cattle in 2012 for experimentally infected sheep
  - German field study and experimental infection of South American camelids with SBV
  - Experimental inoculation of poultry with SBV
- Determination of the role of wildlife (deer, wild boar etc.) in the epidemiology of SBV
- Screening of biting midges for SBV
- Collection of SBV case data

Presentation of selected results
Studies of SBV pathogenesis in pregnant animals (cattle)

Objectives:
1. Collection of experimental data about the pathogenesis of SBV for pregnant animals and their fetuses
2. Determination of the pathogenicity of the virus in fetuses at different gestation stages

Results:
- Two animal trials with in total 36 pregnant cattle were performed
- SBV was transmitted in most cases to the fetuses
- The vulnerable phase of SBV infection in pregnant cattle is approximately ranging between **day 60** and **day 150** of pregnancy

- Transplacental SBV-infection in cattle could be experimentally proven
- A very low percentage of malformed fetuses/abortion was observed (1 malformed fetus, 1 abortion)
Studies of diagnosis and infectivity of SBV-RNA positive bovine semen

Open questions:
1. Is there any excretion of SBV in semen?
2. If yes, is SBV in semen infectious?
3. Is sensitive, specific and robust detection of SBV genome in bovine semen samples possible?

Study design and results:
- Validation of molecular diagnostic methods
  - Protocols for highly sensitive RNA extraction and detection were developed and validated
  - Proficiency tests with all consortium partners as well as further national and European laboratories were performed
- Analysis of semen from naturally infected bulls from Germany
  - 29 (3.8%) of 766 semen batches from 11 of 95 bulls were positive for SBV-RNA
  - Intermittent virus excretion and concurrent occurrence of SBV-RNA in semen and SBV-antibodies in serum possible
- Animal trial
  - animal trials with cattle and mice
  - subcutaneous inoculation of semen
  - 5 of 11 cattle could be infected with 2 of 6 semen batches
Studies of SBV pathogenesis in different mammalian host species

1. Transmission of SBV during winter 2013 in Germany
   • study indicates that SBV transmission occurred in January 2013 at a low level
   • transmission of SBV seems possible, even during winter in central Europe

2. Dynamics of SBV infection within a cattle herd in Germany, 2011
   • rapid spread of SBV infection throughout the entire herd (100%) was observed between September and mid-October 2011

3. Pathogenesis of different SBV isolates in cattle and sheep
   • Viremia (duration, titer) and clinical signs were studied, and a general challenge model for sheep and cattle with infectious serum was developed and used in all pathogenesis studies

4. Studies of SBV infection, e.g. also in South American camelids
   • German field study of SBV infection in South American camelids 2012/2013
     – SBV antibodies (n=309 seropositive, n=4 doubtful) were found in two-thirds (62.4%) of 502 SAC in 61 of the 66 herds (92.4%)
     – no SBV-RNA was detected in any sample
   • SBV pathogenesis in South American camelids experimentally infected with SBV
     – No clinical signs were observed in the SAC
     – SBV genome can be detected in SAC for a short time after SBV-infection
     – SBV ELISA can be used for serological investigations
Determination of the role of wildlife in the epidemiology of SBV

- 27.7% of 1868 blood samples collected from mouflons (72% of 29), roe deer (42% of 499), fallow deer (36% of 44), red deer (29% of 226), sika deer (25% of 20) and wild boar (21% of 1079) between 2011 and 2013 were positive for SBV antibodies

- SBV antibodies found in wild cervids and wild boars, but not in mice and wild carnivores → free-ranging Artiodactyla, but not mice or wild carnivores, might play a role as reservoir in the epidemiology of SBV

Screening of biting midges for SBV

- 937 biting midge pools from 36 collection sites and 633 black flies dating from 2011-2013 were screened for SBV by RT-qPCR

- Two pools of midges were SBV genome positive
- All black flies samples were negative

Collection of SBV case data

- Case/Control study in Germany in cattle, sheep and goats

- “animals kept indoors permanently” as protecting factor, and different risk factors (see report) could be identified