Haemophilus parasuis – a battle of epidemiology and diagnostics

aemophilus parasuis (H. parasuis) is classified in 15 different serovars and between these serovars there is a difference in virulence. H. parasuis is of increasing importance across the globe, both on low and high health status farms. In the past clinical symptoms were often seen after stress, for example after transportation.

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Typically unprotected, expensive replacement gilts would be inserted in a H. parasuis infected herd.
Resident breeding stock in such a farm can contain healthy carrier animals, which do not show any clinical signs. Newly inserted gilts would contract the disease, or symptoms would appear in young piglets after maternally derived immunity had weaned off.

Nowadays it is a much more commonly encountered disease on all types of farms. This puts more emphasis on correct diagnosis, and healthy carriers must be differentiated from clinically infected individuals. H. parasuis normally has a low prevalence, especially in endemic infections, only affecting a small part of the population.

Acute disease with much higher morbidity can be observed in negative populations. When samples are

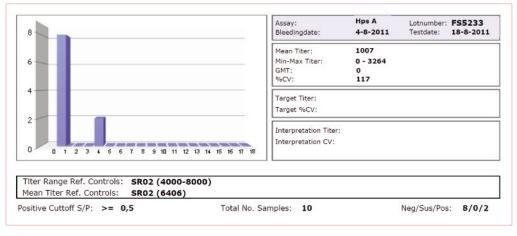


Fig. 1a. H. parasuis serological profile of an infected herd. Only two samples above cut-off level (prevalence of 20%). These are the H. parasuis affected pigs.

collected for diagnostic work or monitoring this epidemiological aspect has to be taken into account in the sample size.

Diagnostic tools

Culture of H. parasuis in a laboratory requires special techniques. The chance of isolating H. parasuis is even smaller when the animal is treated with antibiotics before samples are taken.

Even if the culture is positive, there is no way of discriminating between healthy carriers and infected animals, as H. parasuis is a normal inhabitant of the upper respiratory tract of pigs.

For this same reason PCR assays on samples from the upper respiratory tract can be positive, without the herd showing any clinical symptoms. Only when using a PCR that contains a virulence marker, can assessment of the risk of clinical disease be made.

Serological examination can be just as inconclusive, as most commercial ELISA test kits are based on Lipo-Poly-Saccharides (LPS). Antibodies against LPS are formed in both healthy carriers and clinically infected animals. Maternally derived antibodies against LPS can cause difficulties in interpretation of samples from young piglets.

Diagnostic tools are used to confirm clinical suspicion or to check

the efficacy of an intervention program, so any false positive or inconclusive results are detrimental to the diagnostic process.

Sending in samples for analysis is always an investment of time, effort and money. These resources will go to waste if the results cannot be used to improve the health management on the farm.

It is clear that additional diagnostic tools are needed in this true battle between epidemiology and diagnostics!

Diagnostic breakthrough

A more recent discovery in H. parasuis antibody typing shows that pigs with clinical infection and vaccinated animals have different antibodies from animals that are colonised but healthy.

These specific antibodies are not seen when H. parasuis is only present without causing clinical symptoms. This makes it possible to differentiate between colonised and infected or vaccinated animals (Table 1).

These specific antibodies are directed against the OppA (Oligopeptide permease A) protein. OppA is an intracellular protein, which is presented by macrophages to the immune system during an Continued on page 26

Table 1. Summary of titers observed (cut-off Hps OppA ELISA: titer=1071, cut-off Hps LPS ELISA: S/P=0.6).

Category	Average titer Hps OppA ELISA	Average S/P ratio Hps LPS ELISA
Healthy/non-vaccinated carrier animals	79	0.82
Animals after one vaccination	337	
Animals after two vaccinations	7780	0.60
Clinically suspect, negative	236	0.50
Clinically suspect, positive	2027	0.75

Continued from page 25 infection, inducing an antibody response.

The same antibodies are present after vaccination with H. parasuis vaccines, irrespective if an a-virulent live or an inactivated (whole-) bacteria vaccine is used.

What makes the OppA antibodies so interesting is that research points out that OppA is present in all H. parasuis serotypes, but not in other bacteria like Actinobacillus, Bordetella, Streptococcus, etc.

This discovery is the beginning of a whole new era of H. parasuis diagnostics.

Response to vaccination

The OppA protein induces a serological response in infected and in vaccinated animals and it is thought that this response is correlated with protective immunity. Depending on the dynamics of H. parasuis on the farm, it can be beneficial to implement a vaccination program for sows and/or piglets.

Vaccination can induce protective immunity, but when no follow-up vaccination is given this immunity wears off over time and the sows will no longer transfer MDA to the piglets.

When present, MDA will protect the piglets against an early infection with H. parasuis. This passive protection will last for a couple of weeks depending on the antibody titers in the sows and the colostrum uptake of the piglets. When these piglets are raised on H. parasuis infected farms, vaccination of the piglets is indicated and the vaccination response can be monitored.

When using H. parasuis vaccines and this highly specific OppA ELISA

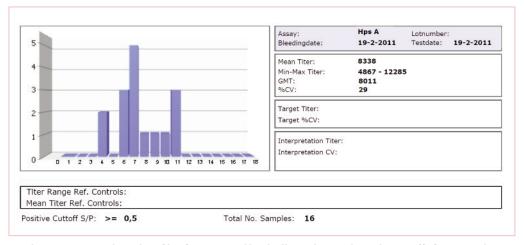


Fig. 1b. H. parasuis serological profile of a vaccinated herd. All samples are above the cut-off after a two shot vaccination.

to check the post vaccination serological profile, it is possible to visualise vaccination responses and create profiles of breeding stock and their piglets.

By using the BioChek Hps qPCR kit on swabs or tissues the presence of H. parasuis in the herd can be detected, as well as the presence of a virulence marker.

With the correct bacteriological procedures the involvement of H. parasuis in the infection can be shown. These three diagnostic tools provide different information and are fully complementary.

Differentiation between vaccination and infection

Typical for an H. parasuis infection is the low prevalence, usually no more than 10-15% in endemically infected herds. This means that only a small part of the population will

become clinically infected, show symptoms and produce antibodies against OppA. This also means that collecting an appropriate number of samples, preferably 20 or more, is essential to properly investigate the presence of H. parasuis field infections. If the sample size is too small, an endemic infection can easily be missed.

After vaccination the antibody response is much larger, with more animals seroconverting. After a two shot vaccination it can be expected to have as much as 95% of the animals positive. This information can then be used when interpreting a serological profile.

When a low prevalence of positive animals in the OppA ELISA is seen, it is likely to be due to field infection (Fig. 1a). High prevalences are expected after a successful two shot vaccination (Fig 1b).

If the herd is showing a much lower prevalence after vaccination,

it is recommendable to evaluate the vaccination procedure and timing.

The H. parasuis OppA ELISA is a highly specific tool to monitor herds for vaccination response and field infection. The BioChek H. parasuis qPCR shows the presence of the pathogen, and provides additional information on the presence of a virulence factor.

By this means a better evaluation on clinical relevance can be made. Bacteriological examination of affected organs together with the clinical expression, will provide a solid answer but is expensive, labour intensive and not always successful. With the BioChek Hps OppA ELISA and Hps qPCR diagnostic opportunities are now greater than ever before.

More information is available from the author on request info@biochek.com