Istituto Zooprofilattico Sperimentale delle Venezie

Sampling procedures and sample delivery instructions for the isolation of *Mycoplasma hyosynoviae*

Introduction

Mycoplasma hyosynoviae is a microorganism belonging to the genus Mycoplasma spp. that infects exclusively the pig, causing arthritis in growing and finishing-age swine. It is mainly localized at the tonsillar level and is transmitted by direct contact or via aerosol over short distances.

Sows carrying *M. hyosynoviae* are primarily responsible for transmission to piglets, but this can also occur among piglets themselves in the post-weaning stages. After colonizing the tonsils, *M. hyosynoviae* can spread and cause synovitis and arthritis in pigs from 10 weeks of age, with mechanisms not yet known. Typical symptoms consist of lameness, reluctance to move, 'sitting dog' position, joint swelling, yellow to brown synovial fluid, sometimes with the presence of fibrin.

Culture isolation, that, unlike other bacterial species, takes from one to three weeks, is chosen to confirm the diagnostic suspicion and allows to obtain the viable microorganism, a fundamental requirement for proceeding with further investigations such as the study of sensitivity to antibiotics by means of MIC (Minimum Inhibitory Concentration) method, the production of stable vaccines, experimental infections or other laboratory activities.

The MIC determination of the *M. hyosynoviae* isolates takes at least three weeks and allows the clinical veterinarian to choose the most appropriate drug for the context. On the basis of these brief considerations, we point out some practical measures to be applied in order to increase the isolation rate.

Figure 1. Swollen joint (top) compared to normal joint (bottom).

Figure 2. Example of sero-fibrinous arthrosynovitis.

When to sample

Attention should be paid to symptomatic animals not subjected to antibiotic treatment, in particular with macrolides, tetracyclines, fluoroquinolones and aminoglycosides which, being effective against mycoplasmas, may potentially affect the result of isolation.

What to sample

In case of arthritis in subjects over 10 weeks of age, the preferred sample for the isolation of *M. hyosynoviae* is joint fluid taken aseptically.

How many samples to collect

For the detection of *M. hyosynoviae,* 2 to 5 joint fluid samples taken from animals with evident clinical signs are sufficient.

How to sample

Sampling is an important critical point, capable of negatively affecting the entire isolation procedure; the main cause of mycoplasma isolation failure is bacterial contamination resulting from incorrect sampling procedure. We recommend to:

1. Remove the skin at the level of the affected joint;
2. Withdraw the joint fluid by inserting the needle into the joint;
3. Transfer the inflammatory fluid into a sterile test tube and uniquely identify the sample (s) delivered to
the laboratory.

The procedure described refers to the sampling of deceased or euthanized animals.

Figure 3: example of joint fluid sampling

How to store and send the samples to the laboratory

Once the samples have been collected, it is important to deliver/send them to the laboratory as soon as possible to preserve the viability of the mycoplasma present in the sample and thus ensure diagnostic success. Samples can be delivered to any IZSVe facility.

Temperature changes must be minimized, refrigerating the sample immediately if it is delivered within 24 hours (to avoid the proliferation of contaminating bacteria); alternatively, the sample can be frozen.

In case of special requests, please contact the laboratory directly in order to find ad hoc solutions for specific conditions or problems encountered.

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