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Tracking *Mycoplasma hyorhinis* prevalence in the farrowing house and nursery using a newly developed real-time PCR

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New Developments

Introduction

Mycoplasma hyorhinis has recently emerged as an important cause of mortality in nursery pigs.[1] Approximately 50% of the cases with polyserositis received at the MNVDL from September 2009 to May 2010 show the involvement of this pathogen based on isolation or PCR.[2] Several protocols for the detection of *M hyorhinis* DNA by gel-based PCR have been published; however, the majority of them are validated for the detection of this pathogen in cell cultures. There are no quantitative PCR (qPCR) protocols available for this pathogen and there is limited information on aspects regarding the epidemiology, control and eradication. The application of qPCR could be employed to study the prevalence of *M hyorhinis*, the transmission of the agent and the efficacy of treatment and control interventions.

Materials and methods

A set of primers and a TaqMan probe were designed with Primer3 Software. The analytical sensitivity of the qPCR was analyzed by testing 10-fold dilutions of extracted DNA of an ATCC *M hyorhinis* strain. The analytical specificity was evaluated by testing 19 bacterial species frequently isolated from swine. A total of 45 clinical samples with polyserositis submitted to the MNVDL for *M hyorhinis* testing were analyzed. To evaluate the more appropriate sampling method, a total of 30 nasal swabs and 30 oropharyngeal swabs from pigs with clinical signs suggestive of *M hyorhinis* systemic infection were tested. To define the prevalence of *M hyorhinis* colonization in different age groups, three breeding farms, including the nursery flow, with a history of isolation of *M hyorhinis*, were selected for this study. Sixty nasal swabs from sows and 60 from piglets of 0, 7, 14 and 21 days of age, as well as 30 nasal swabs from nursery pigs (1-8 weeks post weaning) were collected. Oral fluids were also collected from one pen with 35 pigs at 1 through 8 weeks into the nursery.

Results

The newly developed qPCR test was more sensitive than gel-based PCR, detecting 40×10^4 CFU/reaction. The qPCR detected exclusively *M hyorhinis*. Twenty-one of the 45 clinical samples from pigs with lesions characteristic of *M hyorhinis* infection were positive by qPCR compared to only 14 positive by the gel-based test. A total of 26 nasal swabs and 20 tonsil swabs were positive for the presence of *M hyorhinis*, indicating that nasal swabs are more sensitive to detect this pathogen.

Nasal swab and oral fluid samples from the study herds are being tested at this time.

Discussions

A sensitive and specific qPCR for the detection of *M hyorhinis* in clinical samples has been developed. This test can be used to detect *M hyorhinis* in clinical samples obtained from diseased pigs, to characterize the prevalence of colonization at different ages and to evaluate the efficacy of control and treatment interventions. Although nasal swabs were more sensitive for the detection of *M hyorhinis*, the presence of this pathogen in oropharyngeal samples likely allows its detection in oral fluids (rope testing).

References

1. Leuwerke B. 2009. *Mycoplasma hyorhinis* – Field experiences in diagnosis and control. Allen Leman Swine Conference Proceedings. Saint Paul, MN. 36:89–90.

2. Rovira, A. 2009. Review of *Mycoplasma hyorhinis*. Allen Leman Swine Conference Proceedings. Saint Paul, MN. 36:87–88.