Genetic and antigenetic variability of *Mycoplasma hyopneumoniae* field strains and its implication for improved diagnostics of enzootic pneumonia (EP)

Désirée Mayor[1], Friederike Zeeh[2], Peter Kuhnert[1]
[1]Institut für Veterinär-Bakteriologie, [2]Institut für Tierpathologie, Länggass-Strasse 122, Dept. für Klinische Veterinärmedizin, [3]Schweineklinik, Bremgartenstrasse 109a, Universität Bern, CH-3001 Bern

Schlüsselwörter
Molecular typing, enzootic pneumonia, *Mycoplasma hyopneumoniae*, MLST

Problemstellung und Zielsetzung
Genetic and antigenetic variability is known for *Mycoplasma hyopneumoniae*, and can pose a problem for proper diagnosis of this porcine pathogen. We recently showed that different types of *M. hyopneumoniae* can be distinguished by real-time PCR (ABC, REP or ABC/REP positive). In this project we explored this variability by isolation and characterization of *M. hyopneumoniae* strains and the analysis of sera concurrently taken from the same animals. The genetic variability was investigated by newly developed sequence based typing methods which also allow epidemiological inferences.

Material und Methoden
Self-made optimized Friis medium was used for isolation of *M. hyopneumoniae* strains. Southern-blot analysis was done to confirm the real-time PCR results of isolated strains. Antigenic variability was assessed by immunoblots. Two typing methods one based on sequencing of the polyserine encoding region of the P146 adhesin-like protein gene, and the other based on multilocus sequence typing (MLST) using 7 housekeeping genes were established. Serum samples and lungs were gained at the slaughterhouse from EP-positive pigs. Lungs swabs were used for cultivation of previously determined PCR types of expected *M. hyopneumoniae*. Additionally, samples from previous studies originating besides Switzerland from other countries were taken for applying the newly developed typing methods.

Ergebnisse und Bedeutung
Southern blot analysis of isolated strains using the corresponding DNA-probes confirmed the PCR results since the ABC and REP genes were indeed absent in PCR negative isolates. Immunoblot analysis of whole cell proteins and serum gained from infected pigs showed, however, no big differences in recognized antigens between these different strains. When the MLST data of the sample set was compared to the other P146 based molecular typing approach, full congruence between the two typing methods was observed. Results showed a high diversity of *M. hyopneumoniae* with different types circulating in the Swine population. However, there was always a single strain observed in affected Swiss farms and farms in close geographic or operational contact showed identical clones, indicating that a specific strain is responsible for an outbreak. This might be considered in further programs to control the disease in Switzerland.

Publikationen, Poster und Präsentationen
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