

SWINE HEALTH

Title: Development of a surveillance system to monitor the genetic variability and molecular epidemiology of swine bacterial pathogens – NPB #06-030

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Scientific Abstract:

The **objective** of this project was to standardize genotyping techniques for the major bacterial pathogens affecting the swine industry. Additionally, we created a surveillance system and a genomic database that contains the genotype, date of isolation, geographical location, age of affected animals, tissue, lesions associated with isolation, (sero)type, and antibiotic resistance profile. This project was extended until 2008 and expanded to include additional bacterial isolates. **Methods:** Genomic fingerprints were obtained for the following pathogens using the repetitive-element-based PCR (Rep-PCR): *S. suis* (n=140), *A. suis* (n=104), *A. pleuropneumoniae* (n=34). Each bacterial population structure and diversity was evaluated using computer-based analysis. Dendrograms identifying groups of related strains were constructed for each pathogen and the diversity of each species was calculated using the Simpson's index of diversity. **Results:** Primers targeting different repetitive elements were used, including BoxA, Enterobacterial Repetitive Intergenic Consensus (ERIC), and Repetitive element Consensus (REP). Best results were obtained using ERIC and BoxA primers. Results using the REP primers were inconsistent and this primer was not used in the analysis. Better reproducibility was obtained using the highest annealing temperature tested (50°C) for both primers, and further analysis was performed using this temperature. Diversity and discriminatory power were defined using reference strains and field isolates for each of the pathogens tested. The diversity index for *A. pleuropneumoniae* reference strains was D=0.96 for the ERIC-PCR, D=0.71 for the Box-PCR, and D=0.96 for the combined dendrogram based on both genotyping methods. For *Actinobacillus suis*, the diversity index was D=0.64 for ERIC-PCR, D=1.00 for the BOX-PCR, and D=1.00 for the combined dendrogram. For *Streptococcus suis*, the ERIC-PCR diversity index was D=0.93, the Box-PCR index was D=0.92, and the combined dendrogram had a diversity index of D=0.97. These experiments demonstrated the need to standardize and validate each of these techniques for each of the organisms tested. Following this analysis, further testing of *A. pleuropneumoniae* field isolates was performed by ERIC-PCR, whereas BOX and ERIC-PCR were used to genotype *A. suis* and *S. suis* isolates. Serotyping was also used to define *S. suis* strains groups, as the dendrogram containing ERIC-PCR, BOX-PCR, and serotyping increased the diversity index to D=0.98. Dendrograms containing the most relevant pathotypes were constructed and are available for consultation at the site www.molecularbacteriology.com.

These research results were submitted in fulfillment of checkoff funded research projects. This report is published directly as submitted by the project's principal investigator. This report has not been peer reviewed

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