Session Ecology and Management of Foodborne Agents

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049 - Shotgun metagenomic detection of *Salmonella enterica* in feedlot cattle compared to aerobic culture and PCR techniques.

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Authors

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Abstract

The objective of this study was to compare aerobic culture, polymerase chain reaction (PCR), and shotgun metagenomic approaches for ***Salmonella enterica*** identification in feedlot cattle feces.

Individual rectal fecal samples from cattle exposed (n = 15, Treatment) and not exposed (n = 15, Control) to parenteral tulathromycin were collected upon arrival at the feedlot and 11 days later. Each fecal sample (n=60) was processed with two distinct procedures: one for total DNA extraction and subsequent shotgun metagenomic sequencing as well as a standard aerobic culture protocol for ***Salmonella enterica*** identification in which tetrathionate broth was stored for PCR testing. After extracted DNA samples were sequenced, metagenomic reads were trimmed for increased quality and taxonomically classified with the Kraken software and the National Center for Biotechnology Information's RefSeq genome database.

Although low ***S. enterica*** prevalence restricted formal statistical comparisons, aerobic culture and PCR results had 100% agreement and indicated that 0.05% (3/60) of samples were positive for ***S. enterica***. On the contrary, metagenomic analysis reported that all samples (60/60) contained reads matching to ***Salmonella enterica***. Further examination of how Kraken and other sequence classifiers align reads to reference genomes and classify those reads to an appropriate taxonomic level revealed that plasmid sequences currently associated as being specific to a species also match a wide variety of newly published plasmid sequences and cause false positive classification of reads. To improve the accuracy of ***Salmonella enterica*** identification, we confirmed that all samples contained reads that aligned to the published genomic regions and not the associated plasmids.

This study shows that the traditional techniques of aerobic culture and PCR provide similar results and that shotgun metagenomic results for the identification of bacterial pathogens must be carefully evaluated to fit criteria suitable for its intended use. Additionally, the accuracy and sensitivity of read classification is contingent on the integrity and diversity of reference genome databases.