

# Quantification and Control of Bacterial Pathogens in Agricultural Watersheds

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## **Justification:**

Science-based knowledge on the fate and transport of pathogens in mixed-land use watersheds is critically needed, allowing for identifying important sources and targeting areas of a watershed for remedial efforts in minimizing pathogen transport. This project will develop fundamental baseline data for understanding pathogen sources and transport, and test the effects of various best management practices (BMPs) on reducing pathogen runoff.

## **Objectives:**

The overall objective of this project is to identify critical sources, transport pathways and losses of pathogens in dominant farming systems of NW Arkansas, where confined poultry operations are integrated with beef cattle grazing, with the goal of identifying cost-effective BMPs that will mitigate pathogen transport. Rainfall simulations will be conducted on small plots to evaluate the effects of time after manure application, type of manure, manure treatments, and weather on pathogen runoff. We will also use 28 small watersheds to determine the effect of BMPs, land use, and landscape position on pathogen transport by measuring the effect of: 1) grazing/pasture management, 2) poultry litter application methods, 3) chemical treatment of litter, 4) runoff type (sub-surface vs. overland), 5) treating biosolids with water treatment residuals, and 6) different landscape positions and flow pathways. We will also assess the viability of using indicator organisms by conducting correlation studies to see if *E. Coli* or *Enterococci* are related to specific pathogens in runoff.

## **Progress to date:**

Nutrient and runoff data from 21 of the watersheds has been collected for several years. Site selection for the seven other watersheds was made last year. Berms were constructed and automatic water samplers were installed at five of the sites. New culture methods were developed to enumerate *Salmonella typhimurium*; *E. coli* O157:H7 and *Campylobacter jejuni* in runoff water, since initial attempts were hampered by background interference (litter/soil bacteria). Optimum recoveries were obtained using Brilliant Green Sulfa (BSA), Rainbow Agar and Campylobacter Line Agar for *Salmonella*, *E. coli* O157:H7 and *Campylobacter*, respectively. A real-time PCR bi-plex system was also developed to simultaneously detect *Salmonella* and *E. coli* O157:H7 in water samples. *E. coli* primers and probes targeting the *eae* gene and *Salmonella* primers and probes targeting the *sipB/C* gene were combined into one reaction were able to detect as few as 37 cells/ml. Strong PCR inhibition in runoff water was overcome by adding at least 1.66 uL of BSA. Primers for *Campylobacter* are being incorporated into the bi-plex to create an assay for all three pathogens.

## **Impacts:**

This research will improve the sustainability of beef cattle/poultry operations through the development of cost-effective BMPs which reduce pathogen survival and/or transport.