

PORK SAFETY

Title: Phage therapy to reduce transport and lairage associated increases in *Salmonella* infections: evaluation of delivery methods - NPB #09-131

Investigator: Paul Ebner

Institution: Purdue University

Date Submitted: 11/5/10

Scientific Abstract

We previously produced an anti-*Salmonella* phage cocktail that reduced colonization in swine when the pigs were exposed to an environment heavily contaminated with *Salmonella*, similar to what might be seen in a transport trailer or processing facility holding pen. In this study we increase the efficacy of the phage treatment by (1) expanding the host-range of the cocktail; (2) developing a more cost-effective microencapsulation technique; (3) improving the delivery method. We collected samples from wastewater treatment facilities and isolated 20 distinct phages belonging to either the Siphoviridae or Myoviridae families and identified 10 phages that together lysed a mixed culture of *Salmonella enterica* Typhimurium, Enteritidis, and Kentucky. The phages were microencapsulated using two sodium-alginate-based methods that only reduced the cocktail titer by 1.0–1.5 logs (premicroencapsulation: $10.4 \log_{10}$ PFU/mL; post-microencapsulation method one: $9.2 \log_{10}$ PFU/mL; post-microencapsulation method two: $8.9 \log_{10}$ PFU/mL). Microencapsulated phages remained stable at both 4°C and 22°C for up to 14 days with no appreciable drop in titer (mean titer: $8.9 \log_{10}$ PFU/mL). Twenty-one pigs (~35 lbs) in three replicates were randomly placed into three groups: feed (F), gavage (G), and control (G). The F group was direct fed a 14 phage cocktail daily for five days (1.2×10^{11} PFU/day). On the fifth day, the G group received the same phage cocktail by gavage (1.2×10^{11} PFU), while C pigs received a mock treatment with no phage. All pigs were then challenged with 5×10^8 CFU of *Salmonella enterica* Typhimurium. At six hours post-challenge, all pigs were euthanized and ileal and cecal samples and mesenteric lymph nodes were collected and analyzed for the challenge organism. Pigs in the F group were significantly less likely to shed *Salmonella* at 2h (38.1%) and 4h (42.9%) post-challenge compared to pigs in both the G (2h: 71.4%; 4h: 81.1%) and C (2h: 71.4%; 4h: 85.7%) groups ($P < 0.05$). Likewise, the concentrations of *Salmonella* in the ileal ($2.0 \log_{10}$ CFU/mL) and cecal ($2.7 \log_{10}$ CFU/mL) samples were significantly lower than ileal ($3.0 \log_{10}$ CFU/mL) and cecal ($3.7 \log_{10}$ CFU/mL) samples from C pigs. Taken together, these data indicate that direct feeding phages is a practical and effective means of reducing *Salmonella* colonization and shedding in pigs.

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.

For more information contact:

National Pork Board • PO Box 9114 • Des Moines, IA 50306 USA • 800-456-7675 • Fax: 515-223-2646 • pork.org
