Risk factors for the detection of *Salmonella* in ileocolic lymph nodes in US slaughtered pigs.

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Abstract

*Salmonella* harborage at slaughter can be viewed as a risk for human health through contamination of the pork food chain. Better understanding of herd level factors associated with this harborage would be useful to prioritize further study of epidemiology and control of *Salmonella* in pork production. Ileocolic lymph node samples collected at slaughter from 115 Midwest US swine herds were assayed for *Salmonella enterica*. A subset of these herds was collected sequentially one or two additional times. Herd characteristics and management factors were assessed by a written survey. Risk factors were screened at the univariate sequentially stepwise analysis including herd / sample as a random statistical effect. Pigs at increased risk of *Salmonella* harborage at slaughter included those placed in finisher barns at heavier weights (OR 1.2 per 10 kg increased weight), those from larger herds (OR 2.0 comparing upper quintile to lower quintile of herd size), those from herds that allowed visitors with recent (<8 h) contact with other herds (OR 2.2), or those fed pelleted feeds (OR 2.1). Further investigation of these risk factors and potential biological mechanisms will require further study.

Introduction

*Salmonella enterica* has been commonly identified on U.S. pig farms, with 38.2% of farms testing positive in a 1995 survey of the major swine producing states in the U.S. (Anon., 1997). Risk factors for *Salmonella* shedding have been identified in different countries and production systems. Risk factors identified in studies reviewed by Funk and Gebreyes included not using automated liquid feeding of by-products, not having membership of an Integrated Quality Control production group, trough feeding, high proportion of solid flooring surfaces, concurrent Lawsonia or Porcine Reproductive and Respiratory Syndrome virus infection, and infrequent removal of sow dung failure to empty manure pits. More recently *Salmonella* shedding or harborage in tissues at or near the time of slaughter slaughtered was associated with provision of dry-only diets or use of bowl rather than nipple drinkers.

The following study was designed to assess for a broad range of potential risk factors for *Salmonella enterica* harborage in the ileocolic lymph nodes of slaughtered pigs from Midwest US swine herds. Ileocecal lymph nodes were chosen for microbial *Salmonella* culture since they drain an area of the GI tract often colonized by *Salmonella*, were conveniently and economically collected at slaughter, and because harborage in these nodes has been associated with increased shedding on farm and in fecal samples collected at slaughter (Bahnsen et al., 2005, Bahnsen et al., 2008).

Material and Methods

Samples were collected from pigs at two US slaughter plants. Thirty pigs were sampled from each herd studied; only herds supplying ≥ 30 pigs on a given day were enrolled. Herds were identified prior to sampling from a pre-existing slaughter plant herd supplier list, a U.S. state pork-producer association, coordinated marketing groups, and swine-dedicated veterinary practices. Pigs were sampled on the basis of availability of technical personnel at the time of delivery to the slaughter plant. The target was to sample up to 150 herds for the first sample collection period. At 3-9 month
intervals these same herds were sampled up to two additional times as they marketed additional pigs to the same slaughter plants. The slaughter plant deliveries were a part of normal marketing practices of the herds.

After humane slaughter and evisceration, GI tracts were moved to a separate area. A minimum of 10 g from the ileocolic lymph node chain was aseptically collected. After the overlying mesentery was removed the lymph node tissue was extracted using sterile gauze. For the first two sample collections each lymph node was split, with ≥5 g frozen at -70°C. Samples (~1 g) from each of five pigs were pooled in plastic bags, smashed by a mallet, then blended with tetraethionate broth using a paddle blender. For the third sample collected, 2 g of lymph node tissue was cultured from each pig individually within 24 h of collection.

After completing pooled sample cultures, a subset of frozen paired to samples contributing to culture positive pools were identified. These were thawed overnight at 2°C, then individually cultured using the laboratory process described for fresh samples, except that 2 g lymph node from each individual was blended with 20 mL tetraethionate broth.

A survey, mailed to herd managers the day after collection of samples, included questions on facilities, husbandry, management, and slaughter-transport practices. Non-respondents were sent a reminder card, a second copy of the survey, and were contacted by telephone.

For samplings for which culture results were at the individual-pig level, *Salmonella* prevalence was defined as the number of culture positives / number sampled. Where only pooled sample were tested, prevalence was estimated from a formula derived by regression analysis. (Bahnson et al., 2006). Statistical models were developed using SAS PROC GLIMMIX, including herd of origin as a random effect to adjust for expected clustering of results within herd. Putative risk factors were screened for univariate association; those with *p* < 0.3 were considered for a multivariate model using forward stepwise approach, with *p* < 0.05 to enter the model, and *p* > 0.10 to leave.

**Results**

Valid survey responses and microbiologic data were available for 115 herds for sample 1, 77 herds for sample 2, and 19 herds for sample 3. *Salmonellae* were detected in 73% of sample sets. Mean *Salmonella* prevalence across herds and samples was 8.4%; the unweighted mean prevalence by sample number was as follows: Sample 1, 6.4%; sample 2, 9.0%, and sample 3, 18.3%. The mean herd size, reported as the number of slaughter weight pigs sold in the prior 12 months was 9,275. The mean weight at slaughter and at entry to the barn from which pigs were sold to slaughter (finisher) was 112.2 and 33.9 kg, respectively. Strict batch pig flow was reported by 42% and 16% of respondents by barn and by herd site, respectively. Dry feed was provided in 62% of finisher groups, while 15% reported pelleted feeds. Birds, mice and rats were reported in 39%, 47% and 97% of finishing facilities, respectively. *Salmonella* vaccine was administered to pigs post-weaning in 6% of samples. Thirty variables passed the screening criteria to be considered in multivariate statistical models.

The final model described increased risk of harborage associated with pelleted finishing feeds (OR = 2.1, 95% CI 1.06-4.2), allowing visitors with same day contact to other herds (OR 2.2, 95% CI 1.15-4.3), increasing weight of pigs entering the finishing phase and increasing herd size. Finisher entry weight was associated with odds ratio of 1.2 (95% CI 1.01-1.58) per 10 kg increased weight. The upper and lower quintiles of herd size were 11,755 and 1,600 slaughter weight pigs sold per year, and was associated with a 2.0 (95% CI 1.3 – 3.2) when comparing the larger to the smaller herds.

**Discussion**

The prevalence of *Salmonella* (8.4%) is comparable to that of three other reports on intestinal lymph node tissues collected at slaughter from pigs of Midwest US herds. One study of 73
Midwest US herds estimated 14.5% prevalence (Bahnsen et al., 2006a) and a second study of 25 herds which 3.4% prevalence (Carlson and Blaha).

Pelleting of feed can increase feed to gain efficiency. However, feed form has been linked to changes in Salmonella carriage in observational and experimental settings. Pelleted feed has been associated with higher Salmonella seroprevalence in commercially produced pigs (Leontides et al., Wong et al.) and increased bacterial culture prevalence in an experiment (Mikkelsen et al.). Although feed pelleting has been shown to effect adherence of S. Typhimurium on the intestinal epithelium (Hedemann et al.), a more complete understanding of the underlying mechanisms have not been explained.

Herd marketing larger numbers of pigs per year tended to have higher Salmonella prevalence. Etiologic or explanatory biological mechanisms by which herd size is associated go beyond the findings of the current report; however, further investigation seems warranted, especially given the increasing market share of larger herds in US pork production.

Conclusions

Salmonella shedding in ileocecal lymph nodes in slaughtered pigs was associated with provision of pelleted feed, allowing entry of visitors with recent (<8 h) contact with other herds, increasing weight of pigs when entering the last (finisher) pre-slaughter growth phase, and increasing herd size. These findings suggest the need for further study of these factors and potential underlying mechanisms of effect.

References


