

# Distribution of *Salmonella* serovars in breeding, nursery, and grow-to-finish pigs, and risk factors for shedding in ten farrow-to-finish swine farms in Alberta and Saskatchewan

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## Abstract

The study objectives were to investigate *Salmonella* prevalence, serovar distribution, and risk factors for shedding in 10 purposively selected farrow-to-finish farms in Saskatchewan and Alberta. Pooled fecal samples from the breeding and grow-finish phases and individual fecal samples from breeding, nursery, and grow-finish pigs were cultured for *Salmonella*; serotyping of isolates was performed. Pig and pen characteristics were recorded for each pig and pen sampled.

Overall, 407/1143 (36%) of samples were *Salmonella* positive; within-farm prevalence ranged from 1% to 79%. Sows, nursery, and grow-finish pigs accounted for 43%, 29%, and 28% of positive samples, respectively. More *Salmonella* were detected in pooled pen than individual pig samples ( $P < 0.001$ ). Among 418 *Salmonella* isolates, there were 19 distinct serovars; the most common were *S. Derby* (28.5%), *S. Typhimurium*, var. Copenhagen (19.1%), *S. Putten* (11.8%), *S. Infantis* (6.8%), and *S. Mbandaka* (6.1%). Sows were more likely to shed *Salmonella* than nursery or grow-finisher (OR 2.9,  $P < 0.001$ ) pigs. Pelleted feed (OR 8.2,  $P < 0.001$ ) and nose-to-nose pig contact through pens (OR 2.2,  $P = 0.005$ ) were associated with increased *Salmonella* prevalence. Significant differences in serovar distribution were detected among production phases. The use of pooled pen samples is recommended as a more efficient means for accurate evaluation of *Salmonella* status in different phases of pig production. The breeding herd might be an important source of *Salmonella* persistence within farrow-to-finish farms and should be targeted in control efforts. The latter might also apply to the use of pelleted feed, which remains the most consistently reported significant risk factor for *Salmonella* shedding in pigs.

## Résumé

Les objectifs visés étaient d'étudier la prévalence de *Salmonella*, la distribution des sérovars, et les facteurs de risque pour l'excrétion dans 10 fermes de naisseurs-finisieurs choisies avec intention en Saskatchewan et en Alberta. Des échantillons de fèces regroupés provenant des groupes de reproducteurs et des animaux en croissance-finition ainsi que des échantillons individuels provenant des porcs reproducteurs, en pouponnière, et en croissance-finition ont été cultivés pour la recherche de *Salmonella*; le sérotypage des isolats a été effectué. Les caractéristiques des porcs et des enclos étaient notées pour chaque porc et enclos échantillonnés.

De manière globale, 407/1143 (36 %) des échantillons étaient positifs pour *Salmonella*; la prévalence intra-ferme variait entre 1 % et 79 %. De tous les échantillons positifs, 43 %, 29 % et 28 % provenaient respectivement des truies, des porcs en pouponnière et des porcs en croissance-finition. Plus de *Salmonella* étaient détectés dans les échantillons regroupés que dans les échantillons de porcs individuels ( $P < 0,001$ ). Parmi 418 isolats de *Salmonella*, il y avait 19 sérovars distincts; les plus fréquents étaient *S. Derby* (28,5 %), *S. Typhimurium* var. Copenhagen (19,1 %), *S. Putten* (11,8), *S. Infantis* (6,8 %) et *S. Mbandaka* (6,1 %). Les truies étaient plus susceptibles d'excréter *Salmonella* que les porcs en pouponnière ou en croissance-finition (OR 2,9;  $P < 0,001$ ). La nourriture en granule (OR 8,2;  $P < 0,001$ ) et le contact nez-à-nez des animaux entre les parcs (OR 2,2;  $P = 0,005$ ) étaient associés avec une augmentation de la prévalence de *Salmonella*. Des différences significatives dans la distribution des sérovars ont été détectées parmi les phases de production. L'utilisation des échantillons regroupés est recommandée comme étant un moyen plus efficace pour une évaluation précise du statut de contamination par *Salmonella* dans les différentes phases de production porcine. Le troupeau reproducteur pourrait être une source importante pour la persistance de *Salmonella* à l'intérieur des fermes de naisseurs-finisieurs et devrait être ciblé dans les efforts de réduction de l'infection. Ceci est également applicable à l'utilisation de nourriture en granule qui demeure le facteur de risque le plus fréquemment rapporté pour l'excrétion de *Salmonella* chez les porcs.

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## Introduction

*Salmonella* is an important challenge to the swine industry world-wide because of its implications for public health. Salmonellosis in humans results in high societal costs that include medical related expenses, losses associated with reduced or lost work productivity, and other costs (1,2). Although in North America pork is not considered a major source for human salmonellosis, *Salmonella* in pigs has become an important research priority over the past decade, primarily as a result of extensive implementation of *Salmonella* surveillance or monitoring programs in Denmark and other European countries. In Canada, Quebec has extensively investigated *Salmonella* in pigs (3,4), and recently initiated a provincial control program for *Salmonella* in pigs. Relatively large baseline studies were conducted in Ontario and Alberta (5,6), where approximately 40% to 60% of finishing swine farms were *Salmonella* positive, with the overall number of positive samples ranging from 11% (individual pigs) to 14% to 18% (pooled samples). A national, abattoir-based baseline study reported an overall pig carcass contamination prevalence of 4.2% (7), demonstrating that *Salmonella* carcass contamination rates within Canada are low when compared to the number of positive animals. Still, further improvement is needed and additional research into the epidemiology of *Salmonella* at both the farm and abattoir levels within Canada is required.

Swine production systems differ substantially among countries (8), and within Canada, among provinces and regions (5). In Canada, limited research has been conducted on the epidemiology of *Salmonella* in pigs, and the research that has been done to date has focused primarily on the finishing pig. Farzan et al (6) found that 46% (37/80) of farms in Ontario were *Salmonella*-positive. In western Canada, Rajić et al (5) reported that among 90 Alberta swine finishing farms producing  $\geq 2000$  pigs, 26% to 58% of farms studied were *Salmonella*-positive at any given time, and had low to moderate (1 to 4 positive samples, average 15 samples collected per farm) within-farm prevalence. Sorensen et al (9) examined the prevalence of *Salmonella* spp. in Alberta pigs at slaughter, reporting 35% positive cecal samples and 37 different serovars. Most recently, an examination of slaughter pigs from Saskatchewan abattoirs found 13% positive cecal samples (10). Only one study has investigated the distribution of *Salmonella* species in various pig production phases of 2 integrated production systems, where prevalence ranged from 17% to 66% (3). However, no study has investigated *Salmonella* serovar distribution throughout all phases (farrow-to-finish) of pig production in western Canada. The development and implementation of *Salmonella* control programs requires knowledge of the baseline prevalence and serovar distribution in targeted pig populations within a specific region, and thorough knowledge includes investigation of the breeding herd as well as finishing pigs.

Therefore, the objectives of this study were to evaluate *Salmonella* prevalence and serovar distribution in sows, nursery and grow-to-finish pigs, and risk factors for *Salmonella* shedding, using cross-sectional sampling on 10 purposively selected farrow-to-finish swine farms in Saskatchewan and Alberta.

## Materials and methods

### Farm selection

Ten farrow-to-finish swine herds (herd size  $n > 100$  sows) from Alberta (7 farms) and Saskatchewan (3 farms) were purposely selected by swine veterinarians, based on their presumed *Salmonella* positive status ( $n = 7$ ) or *Salmonella* negative status ( $n = 3$ ), and the producer's willingness to participate in the study. Purposeful herd selection was chosen to meet the objectives of a concurrent study evaluating diagnostic tests for *Salmonella* in pigs (unpublished data). Herds were presumed positive if either the herd veterinarian or producer observed clinical salmonellosis within the previous 12 mo, if *Salmonella* species were identified during routine testing, or if replacement breeding stock were purchased from known *Salmonella*-positive farms. Herds were presumed negative if none of these criteria were met. The number of herds and the number of samples used in the study were a function of logistic and financial constraints.

### Sample collection

Each herd was visited once from May through August 2004. Samples were delivered to the laboratory either within 2 h of leaving the farm, or held on ice overnight and delivered the following day.

*Collection of individual fecal samples.* On each farm, feces (minimum 10 g) were collected, from each of 10 randomly selected sows, directly from the rectum or from freshly voided feces on the floor. In the grow-to-finish area, 1 individual sample (minimum 10 g) was similarly collected from 1 pig in each of 30 different randomly selected pens. No individual fecal samples were collected from nursery pigs as most pigs were too small to collect 10 g feces directly from the rectum.

*Collection of pooled fecal samples.* Twenty pooled samples were taken from the breeding phase in each herd, by collecting a minimum of 5 g of feces from 5 different sows into a single container. Samples from individual sows, as described above, were not incorporated into the pooled sample. In both nursery and grow-to-finish phases, 1 pooled pen floor fecal sample was collected from each of 30 randomly selected pens or all pens on farm if there were  $< 30$  pens. For each pooled pen sample a minimum of 5 g of fecal material was collected from 5 different locations on the pen floor.

### Bacteriological culture

Bacteriologic culture for *Salmonella* was performed by the Agri-Food Laboratories Branch (AFLB), Food Safety Division of Alberta Agriculture and Rural Development. All samples were refrigerated and cultured within 24 to 48 h of receiving samples and thoroughly mixed prior to culture.

Ten grams of feces were inoculated into 90 mL of buffered peptone water (BPW) and incubated at 35°C for 20 to 24 h. After incubation, 0.1 mL of BPW was inoculated into 10 mL of Rappaport Vassiliadis broth (RV), placed into a 42°C water bath for 30 min, and then incubated at 42°C for 22 to 24 h. Simultaneously, 1 mL of BPW was inoculated into tetrathionate broth (TT) to which 0.2 mL of iodine solution had been added just prior to use, and placed in a 35°C waterbath for 30 min, then a 35°C incubator for 22 to 24 h.

After incubation, 10 µL of RV and TT were streaked onto XLT4 and Rambach (RAM) selective agar plates and incubated at 35°C for 18 to 24 h, then read. Plates without significant growth of suspect colonies were re-incubated and read after an additional 24 h. At the same time, 0.3 mL of TT (0.1 mL to each of 3 sites) was inoculated onto a modified semi-solid Rappaport-Vassiliadis (MSRV) plate and incubated at 42°C for 20 to 24 h. The halos of growth that occurred on the MSRV plates were streaked to XLT4 and RAM plates and incubated at 35°C for 24 h. Negative plates were re-incubated and read again at 48 and 72 h. Suspect colonies were screened using triple sugar iron agar slants, urea agar slants and lysine iron agar slants, and plated to a blood agar plate and MacConkey plate to check for purity then tested with *Salmonella* Poly O and Poly O1 antisera agglutination (Denka Seiken Company, Tokyo, Japan). Unusual or atypical reacting suspect colonies were further tested using Vitek GNI or API-20E (bioMerieux Vitek, Hazelwood, Missouri, USA). All isolates were frozen at -70°C then forwarded to the Laboratory for Foodborne Zoonoses, Public Health Agency of Canada, Guelph, Ontario, for confirmation by serotyping.

### Serotyping and phagotyping

One isolate per each *Salmonella*-positive sample, or 2 isolates if they were morphologically distinct, was sent for serotyping and phagotyping at the OIE Reference Laboratory for Salmonellosis, Laboratory for Foodborne Zoonoses, Public Health Agency of Canada, Guelph, Ontario. The serotyping and phagotyping techniques followed standard procedures and have been previously reported (5).

### Data collection

During sampling, the primary investigator observed and recorded pen and pig information; sex and age of each individual pig sampled; number of pigs in pen, area and pig density; floor and wall type, and cleanliness of each pen; feed type and feeding method; nose-to-nose contact between pigs through pen separations, and feces characteristics. A list of variables and their distribution is shown in Tables I and II.

### Statistical analysis

The pig and pen were *Salmonella*-positive if the fecal sample collected from that pig or pen tested positive. Descriptive statistics were summarized and statistical models were developed using a commercial software program (Stata/SE v9.2; StataCorp, College Station, Texas, USA). Generalized linear mixed models, with a random intercept to account for clustering of individual and pen samples within herd, were used to: 1) examine the difference in *Salmonella* shedding among production phases and the associations between pen-level variables and *Salmonella* shedding; 2) estimate the proportion of variance of *Salmonella* shedding attributable to each production level; 3) compare *Salmonella* recovery from pooled fecal versus individual samples collected from grow-finish pigs; and 4) describe the differences in serovar-specific prevalence among the various production phases. All models used a logit link function, binomial distribution, and an exchangeable correlation structure.

**Table I. Distribution of the categorical variables in each production phase that were considered as possible risk factors for the occurrence of *Salmonella* in 10 farrow-to-finish pig farms from Alberta and Saskatchewan**

Variable	Levels of response	Distribution		
		Sows (%)	Nursery (%)	Grow-finish
Sex	gilt(s)	na	34	38
	barrow(s)	na	34	37
	mixed pen	na	32	24
	sow(s)	100	na	na
Fecal score	runny	0	0	3
	normal	93	99	96
	hard/dry	7	1	1
Fed pelleted feed	yes	30	51	29
	no	70	49	71
Fed wet feed	yes	44	78	83
	no	56	22	17
Fed on floor	yes	20	0	8
	no	80	100	92
Pen cleanliness	clean	56	76	25
	slightly wet/dirty	24	23	49
	moderately wet/dirty	16	0	9
	very wet/dirty	4	0	16
Pen floor type	full slatted	25	94	16
	part slatted	66	6	80
	not slatted	9	0	4
Concrete floor	yes	79	4	91
	no	21	96	9
Concrete walls	yes	9	4	54
	no	91	96	46
Nose-to-nose contact between pens	yes	79	53	80
	no	21	47	20
Production phase	sows	100	0	0
	nursery/weaners	0	100	0
	grow-finishers	0	0	100

Risk factor analysis was limited to pooled fecal samples to minimize potential bias introduced by different sampling strategies among production phases and because individual samples were only available from 2 of 3 phases. In the first step, the unconditional association between each potential risk factor and whether or not the pooled fecal culture was positive for *Salmonella* was evaluated. All variables with an unconditional *P*-value of  $\leq 0.20$  were evaluated for inclusion in a multivariable model using a manual forward-stepwise process. Risk factors were defined as confounders if removing or adding the factor changed the effect estimate by more than 20% (11). Variables with  $P \leq 0.05$  were considered statistically significant. Biologically reasonable first-order interaction terms were examined

**Table II. Distribution of the continuous variables in each production phase that were considered as possible risk factors for the occurrence of *Salmonella* in 10 farrow-to-finish pig farms from Alberta and Saskatchewan**

Variable	Production phase	Mean	Median	Minimum	Maximum	s	n
	Age in weeks <sup>a</sup>	Nursery	6.6	7	3	11	2
	Grow-finish	16.6	16	8	27	4.7	255
Number of pigs in pen <sup>b</sup>	Sows	5.3	6	1	10	2.7	39
	Nursery	18.8	18	6	70	9.8	255
	Grow-finish	16.4	14	3	120	12.9	295
Pig density (m <sup>2</sup> per pig) <sup>b</sup>	Sows	2.3	2.25	0.75	4.3	1.1	39
	Nursery	0.25	0.25	0.1	0.53	0.08	255
	Grow-finish	0.85	0.74	0.15	3.12	0.47	295

s — standard deviation.

<sup>a</sup> Observations on age were not recorded for breeding females.

<sup>b</sup> Data from pigs in pens only; does not include observations from sows in gestation/farrowing crates ( $n = 161$ ).

**Table III. Proportion of all fecal samples positive for *Salmonella* based on bacterial culture for each phase of pig production in 10 farrow-to-finish herds in Alberta and Saskatchewan**

Farm	Total number of samples	Overall	Number positive				
			Sows (pooled)	Sows (ind)	Nursery (pooled)	Grow-finish (pooled)	Grow-finish (ind)
1	120	77	17/20	10/10	17/30	21/30	12/30
2	96	54	14/20	5/10	1/16	17/25	17/25
3	120	95	17/20	5/10	22/30	28/30	23/30
4	120	2	1/21	0/9	0/30	1/30	0/30
5	120	34	13/20	5/10	2/30	11/30	3/30
6	120	46	12/20	3/10	5/30	15/30	11/30
7	119	18	5/19	2/10	9/30	2/30	0/30
8	104	58	17/20	7/10	13/14	15/30	6/30
9	108	11	5/20	0/10	2/19	3/30	1/29
10	116	12	1/20	1/10	10/26	0/30	0/30
All farms	1143	407/1143	102/200	38/99	81/255	113/295	73/294

where > 1 significant risk factor was identified in the final main effects model. Statically significant interaction terms were included in the final model.

To estimate the proportion of variance in *Salmonella* shedding attributable to production phase, a 3-level model was developed, including a random effect for production phase nested within farm. Using pooled samples only, a model with intercept as the only fixed term (null model) was fitted to compute the proportion of the overall variance in *Salmonella* shedding accounted for at the level of production phase and then farm. The proportion of variance that was accounted for by differences between herds was estimated as:

$$\rho_h = \frac{\sigma_h^2}{\sigma_h^2 + \sigma_p^2 + \sigma_\varepsilon^2} \quad \text{[Equation 1]}$$

where:  $\sigma_h^2$  was the herd-level variance, and  $\sigma_p^2$  was the production phase variance estimated from the null model; and  $\sigma_\varepsilon^2$  was the sampling variance estimated according to the latent variable method

(12). Likewise, the proportion of variance that was accounted for by differences between production phases was estimated as:

$$\rho_p = \frac{\sigma_p^2}{\sigma_h^2 + \sigma_p^2 + \sigma_\varepsilon^2} \quad \text{[Equation 2]}$$

To evaluate *Salmonella* recovery from different sampling procedures, the odds of obtaining a *Salmonella* positive sample from a pooled fecal sample were compared to the odds of obtaining a positive culture from an individual sample. The unconditional association between sampling strategy and whether or not the fecal sample was *Salmonella*-positive was evaluated in a model with a random intercept for herd. This analysis was restricted to samples from grow-finish pigs as this was the only production area where both pooled and individual samples were collected from the same pen.

Both pooled and individual samples were used collectively to estimate differences in serovar-specific prevalence among the different phases of production. A positive outcome was the presence

**Table IV. Unconditional associations between predictor variables and the occurrence of *Salmonella* positive pooled fecal samples from pens on 10 farrow-to-finish pig farms from Alberta and Saskatchewan**

Variable	Levels of response	$\beta$ (coefficient) <sup>a</sup>	95% CI ( $\beta$ )	P-value
Sex	<i>overall</i>			0.018
	gilt(s)	-0.85	-1.34 to -0.36	0.001
	barrow(s)	-0.55	-1.03 to -0.67	0.026
	mixed pen	-1.65	-2.39 to -0.92	0.001
	sow(s)	Reference		
Fecal score	<i>overall</i>			0.071
	normal	-1.48	-3.28 to 0.31	0.11
	hard/dry	-0.40	-2.53 to 1.74	0.72
	runny	Reference		
Fed pelleted feed	yes	0.95	0.19 to 1.71	0.014
	no	Reference		
Fed wet feed	yes	0.57	0.09 to 1.06	0.020
	no	Reference		
Fed on floor	yes	0.57	0.08 to 1.24	0.087
	no	Reference		
Pen cleanliness	<i>overall</i>			0.008
	slightly wet/dirty	0.34	-0.06 to 0.74	0.10
	moderately wet/dirty	0.19	-0.52 to 0.90	0.60
	very wet/dirty	-1.17	-2.11 to 0.22	0.016
	clean	Reference		
Pen floor type	<i>overall</i>			0.46
	part slatted	0.65	0.28 to 1.03	0.001
	not slatted	0.15	-1.21 to 1.51	0.83
	full slatted	Reference		
Concrete floor	yes	0.77	0.40 to 1.14	0.000
	no	Reference		
Nose-to-nose contact between pens	yes	0.67	0.16 to 1.18	0.009
	no	Reference		
Production phase	<i>overall</i>			0.089
	nursery/weaners	-0.73	-1.17 to -0.29	0.001
	grow-finishers	-1.10	-1.56 to -0.63	0.000
	sows	Reference		
Number of pigs in pen	-0.05	-0.07 to -0.03	0.000	Number of pigs in pen
Pig density	0.67	0.28 to 1.06	0.001	Pig density

95% CI — 95% confidence interval.

<sup>a</sup> Log odds ratio from random-effects logistic regression model.

of a specific serovar; any other serovar, or any *Salmonella*-negative sample, was considered a negative outcome. For each of the 5 most prevalent serovars, the association between production phase and whether or not the fecal sample was positive for each of these 5 serovars was investigated. All models were adjusted for sampling strategy (pooled versus individual samples) by including this variable as a fixed effect in each model.

## Results

### Farm description

Farm size ranged from 130 to 2070 breeding females (mean 531, median 333) and the number of pigs produced for slaughter by each farm ranged from 1100 to 27 000 animals annually (mean 8332,

**Table V. Final multivariable regression model for associations between predictor variables and pen *Salmonella* status on 10 farrow-to-finish pig farms from Alberta and Saskatchewan**

Variable	$\beta$ (coefficient) <sup>a</sup>	95% CI ( $\beta$ )	P-value
Fed pelleted feed			
Yes	2.1	1.18 to 3.03	0.000
No	Reference		
Nose-to-nose contact			
Yes	0.81	0.24 to 1.37	0.005
No	Reference		
Production phase			
Nursery	-1.4	-1.91 to -0.88	0.000
Grow-finish	-0.84	-1.30 to -0.88	0.000
Sows	Reference		
Grow-finish	0.56	0.09 to 1.02	0.019
Sows	1.40	1.91 to 0.88	0.000
Nursery	Reference		

95% CI — 95% confidence interval.

<sup>a</sup> Log odds ratio from random-effects logistic regression model.

**Table VI. *Salmonella* serovars isolated from 10 farrow-to-finish pig farms in Alberta and Saskatchewan, grouped according to production phase**

Serovar	Sows, pooled	Sows, individual	Nursery, pooled	Grow-finish, pooled	Grow-finish, individual	Total
S. Derby	20	12	6	48	33	119
S. Typhimurium var. Copenhagen	23	6	24	19	9	81
S. Putten	12	4	7	14	12	49
S. Infantis	8	3	4	7	6	28
S. Mbandaka	0	0	14	8	4	26
S. Give	8	8	1	1	1	19
S. Anatum	5	2	3	5	2	17
S. Ohio	0	0	3	1	0	4
S. Rubislaw	2	1	0	1	0	4
S. Livingstone var. 14+	1	0	3	0	0	4
S. Typhimurium	0	0	0	0	3	3
S. Worthington	3	0	0	0	0	3
S. Give var. 15+	0	0	1	1	0	2
S. Enteritidis	1	0	0	1	0	2
S. Ohio var. 14+	1	0	1	0	0	2
S. Brandenburg	2	0	0	0	0	2
S. Lexington var. 15+	0	0	0	1	0	1
S. Heidelberg	0	0	1	0	0	1
S. Kentucky	1	0	0	0	0	1
Untypeable	15	2	15	10	8	50

median 4300). Three herds primarily produced breeding stock but finished the barrows and cull gilts. Seven herds produced hogs for slaughter only.

### ***Salmonella* prevalence (both pooled and individual samples)**

*Salmonella* was isolated from all 10 study farms. Based on total numbers of positive samples, prevalence within presumed-negative

herds ranged from 20% to 56%, while prevalence within presumed-positive herds ranged from 2% to 79%. There were 407/1143 (36%) positive fecal samples across all production phases (Table III). Four farms accounted for 70% (284/407) of all positive samples (Table III). The highest proportion was found in the breeding sows, with 38% (38/99), and 51% (102/200) of individual and pooled samples, respectively, positive for *Salmonella*. In the grow-finish population, 25% (73/294) of the individual samples and 38% (113/295)

**Table VII. *Salmonella* phage types isolated from 10 farrow-to-finish pig farms in Alberta and Saskatchewan**

Serovar	Phage type	Number of isolates	% of isolates
S. Typhimurium var. Copenhagen	UT5	30	34.5%
	21	16	18.4%
	104	13	14.9%
	22	5	5.7%
	208 var	5	5.7%
	135	4	4.6%
	146a var	3	3.4%
	208	1	1.1%
	142 var	1	1.1%
	Untypeable	2	2.3%
S. Typhimurium	UT3	1	1.1%
	27	2	2.3%
S. Enteritidis	U276	1	1.1%
	11b	1	1.1%
S. Heidelberg	20a	1	1.1%
	10	1	1.1%
<i>Total</i>		87	100%

of the pooled pen samples tested positive. In the nursery, 32% (81/255) of all pooled pen samples were positive. The occurrence of *Salmonella* positive samples varied significantly among all production phases for the pooled samples ( $P < 0.001$ ) and between the breeding sows and grow-finish population for the individual samples ( $P = 0.002$ ).

### Risk factors for shedding *Salmonella* (pooled sample results)

Risk factor variables that were unconditionally associated ( $P \leq 0.20$ ) with *Salmonella* shedding in the pooled samples are summarized in Table IV. Several management factors were specific and uniform to the breeding herd on the farms studied; for example, all breeding females were, naturally, “sex = female,” and most breeding females were housed in gestation stalls or farrowing crates. The variable “sex” was therefore perfectly correlated with “production phase — sows” and the variables “number of pigs in pen,” and “pig density” were also found to be highly correlated with this production phase. Consequently, these 3 variables were not included in the initial model. A second model was developed to assess the significance of these variables in nursery and grow-finish pigs only.

Only the variables “fed pelleted feed,” “production phase,” and “nose-to-nose contact” were found statistically significant ( $P \leq 0.05$ ) in either model; thus, the estimates are reported for a single model including these 3 variables and applied to data from all production phases (Table V). In this model, the odds of a positive pooled *Salmonella* culture remained different across the different production phases (Table V). Sows were 2.3 (CI<sub>OR</sub> 1.5, 3.7) times more likely to shed *Salmonella* than grow-finish pigs, and 4.0 (CI<sub>OR</sub> 2.4, 6.8) times more likely to shed than nursery pigs; grow-finishers were 1.7 (CI<sub>OR</sub> 1.1, 2.8) times more likely to shed *Salmonella* than nursery pigs. Pooled samples from pens that

received pelleted feed were 8.2 (CI<sub>OR</sub> 3.2, 20.6) times more likely to be *Salmonella*-positive than samples from pens with non-pelleted feed (Table V). Pens allowing for nose-to-nose contact among pigs were 2.2 (CI<sub>OR</sub> 1.3, 4.0) times more likely to be *Salmonella*-positive than pens without such contact (Table V).

### Variance component estimation (pooled fecal samples)

The estimates of variance in the occurrence of *Salmonella* positive pooled fecal samples at the herd and production phase levels were 2.24 [standard error ( $S_{\bar{x}}$ ) = 1.31] and 1.34 ( $S_{\bar{x}}$ , 0.57), respectively. Using the latent variable method (12), the proportion of variance residing at the herd level [Equation 1] was 33%, while 20% of total variance was due to production phase [Equation 2].

### *Salmonella* recovery from pooled versus individual samples

Overall, *Salmonella* was isolated from 38% (113/295) of pooled grow-finish samples and 25% (73/294) of individual samples. The odds of *Salmonella* recovery from grow-finishers were 2.9 times (CI<sub>OR</sub> 1.8, 4.5;  $P < 0.001$ ) higher from pooled than individual samples. In sows, 51% 102/200 of pooled samples and 38% (38/99) of individual samples were *Salmonella*-positive; however, no statistical test for differences between sampling strategies was done for this production phase as paired pooled and individual samples were not collected from the same pen or animals.

### *Salmonella* serovar and phage type distribution (both pooled and individual samples)

The serovar prevalence for each production phase is shown in Table VI. Nineteen distinct serovars were identified. Multiple serovars (2 to 8 per farm) were detected on all but 1 farm. Fewer serovars were detected in individual samples (7 and 8 typed serovars, for sows and grow-finish, respectively) than in pooled samples (13, 12, and 12 typed serovars, for sows, grow-finish, and nursery, respectively). The 5 most common serovars were *S. Derby* (28.5%), *S. Typhimurium* var. Copenhagen (19.4%), *S. Putten* (11.7%), *S. Infantis* (6.7%), and *S. Mbandaka* (6.2%) (Table VI). Phage typing results for all *S. Typhimurium*, *S. Typhimurium* var. Copenhagen, *S. Enteritidis*, and *S. Heidelberg* isolates are presented in Table VII. On the 7 farms where these serovars were found, the number of phagetypes isolated per farm ranged from 1 to 6, with multiple phagetypes found on 4 farms. *Salmonella* Typhimurium PT104 was detected on 2 farms, and on both these farms this was the only phagetype present.

The serovar distributions in various production phases were compared for the 5 most prevalent serovars, with the exception of *S. Mbandaka*. Since this serovar was not isolated from the breeding herd, this comparison was limited to nursery pigs and grow-finishers production phases. In an analysis adjusted for sample type (pooled versus individual), significant differences in serovar distribution were found between production phases; these pair-wise contrasts are presented in Table VIII.

**Table VIII. Differences in *Salmonella* serovar distribution between production phases on 10 farrow-to-finish pig farms in Alberta and Saskatchewan**

Serovar	Contrast	OR	CI <sub>ORlower</sub>	CI <sub>ORupper</sub>
S. Derby	grow-finish versus nursery	10.2	4.2	24.9
	grow-finish versus sows	1.5	0.9	2.5
	sows versus nursery	6.7	2.6	16.9
S. Infantis	sows versus nursery	3.1	0.9	10.8
S. Putten	sows versus nursery	3.2	1.2	9.0
S. Typhimurium var. Copenhagen	nursery versus grow-finish	3.0	1.4	6.4
	sows versus grow-finish	3.3	1.7	6.4
S. Mbandaka	nursery versus grow-finish	4.4	1.7	11.3

## Discussion

Existing research on the epidemiology of *Salmonella* in pigs has focused primarily on finishing pigs due to their proximity to the consumer. Still, pigs of other ages can play an important role in the maintenance and dissemination of *Salmonella* on-farm, as well as contribute to food safety issues themselves. In this study we investigated the epidemiology of *Salmonella* throughout all levels of swine production and reported on production phase level factors which could potentially influence the *Salmonella* status of pigs, an important contribution to future surveillance and control efforts for *Salmonella* in western Canada.

Three herds included in this study were initially presumed to be free of *Salmonella*; however, all 10 herds were ultimately found to be *Salmonella*-positive. Although only *S. Typhimurium* and *S. Choleraesuis* commonly cause clinical salmonellosis in pigs, infection by other serovars causes prolonged carrier states and intermittent shedding (13). Even when *S. Typhimurium* and *S. Choleraesuis* are present within a herd, infection may remain primarily sub-clinical without outbreaks of clinical salmonellosis. In these cases, and in the absence of regular testing, the presence of *Salmonella* goes unsuspected and undetected. Our observations then emphasize clinical history is not an accurate indicator of herd *Salmonella* status.

The current study parallels previous studies that sows were more at risk for shedding *Salmonella* than both nursery and grow-finish pigs (8,14–17). Cull sows are usually shipped to slaughter immediately after weaning, when increased shedding has been observed (16). Additionally, transport and lairage practices may contribute to increased shedding of *Salmonella* by sows immediately prior to slaughter (18,19). For these reasons, potential control efforts should be placed on this population both on-farm and at slaughter to reduce the on-farm *Salmonella* reservoir as well as minimize potential food safety risks.

The use of pelleted feed and nose-to-nose pig contact through pens were 2 other significant risk factors detected in this study. Other researchers, both in Canada and elsewhere, have also reported strong associations between the use of pelleted feed and farm *Salmonella* status (15,20–22). Other research groups reported that the use of acidifying rations reduced the prevalence of *Salmonella* in market-age pigs (23), and that pelleted feed decreased stomach acidity in the pig compared with coarser feed (24) or increased mucin secretion, contributing to the survival of ingested *Salmonella* and colonization of the pig (25). Efforts to reduce *Salmonella* at the farm level could

incorporate acidification of water or rations or changing feed to coarser-grind rations. Nose-to-nose contact between pigs through pens is a less likely target for intervention, since this is a feature inherent to barn design and unlikely to be easily changed. However, consideration of the possibility of transmission of *Salmonella* and other important pathogens between pens and production units should be taken into consideration when designing and building new barns.

Approximately 1/3 of the estimated variance of *Salmonella* shedding resided at the farm level, suggesting that farm-level factors may exert the greatest influence on the outcome (26). Others have reported farm type as a significant risk factor for *Salmonella* shedding (27), which further supports the premise that farm-level management factors significantly impact the *Salmonella* status of pigs. Within farms, 20% of the variance of *Salmonella* shedding was attributable to production phase, suggesting that production phase specific factors might also be important and concurs with our finding that production phase is a significant risk factor when included as a fixed effect in the regression model. However, previous studies investigating risk factors for *Salmonella* have focused primarily on finishing pigs and little information regarding risk factors for pigs of other ages is available. Further research into production phase level factors, which could potentially influence the *Salmonella* status of pigs, is required.

One-time sampling of individual pig feces (compared with repeated or pooled samples) has been identified, among other reasons, for poor sensitivity of *Salmonella* culture (28). Similarly, in our study, more positives were found in pooled pen samples than from individual pigs. Furthermore, more positive farms were identified when sampling pigs from all production phases. Consequently, the use of pooled pen samples, from all phases of pig production, is recommended as a more reliable means of accurately of establishing the prevalence of *Salmonella* in swine herds.

The observed distribution in *Salmonella* serovars was similar to other findings within Canada (3,5,9,29) and the United States (30,31), except for one notable exception. *Salmonella* Putten, a serovar that has not been reported by any of these studies, was the 3rd most common serovar in the study, and was found only in 3 farms in Saskatchewan; these farms also accounted for over 80% of all untypeable isolates. Taken together, this is suggestive of either possible geographical differences in serovar distribution in pigs in western Canada or other common factors that contributed to the transmission of specific serovars between these herds. Molecular methods, such as those used to document transmission of *S. Typhimurium* DT 104 between



geographically related herds in Denmark (32), would be necessary to further investigate this observation and further our understanding of the spread of *Salmonella* within and between herds.

Significant differences were observed in serovar prevalence between production phases. Surveillance efforts which focus solely on finisher pigs, either on-farm or at slaughter, would not have detected the full range of serovars present on these farms. As other researchers have noted, an understanding of serovar type and distribution is important because certain serological tests, such as the Danish-mix enzyme-linked immunosorbent assay (ELISA), detect antibodies against serogroups B, C1, and D1 only (5). Serological response to serovars such as *S. Mbandaka*, *S. Anatum*, or *S. Putten*, would not have been detected by this ELISA. The changes in serovar distribution as pigs progress through the production cycle presents a challenge to *Salmonella* surveillance and control efforts that use serological tools only; cost-effective complementary bacteriologic testing of samples from all levels of pig production is necessary for accurate evaluation of *Salmonella* status in swine herds.

In this study, only 3 significant risk factors were identified, possibly because the study did not have sufficient power to detect other significant risk factors due to the small number of studied farms. The main study limitation was the use of purposeful selection of farms, which was necessary to meet the objectives of a concurrent study. For these reasons, no conclusions based on this study should be made regarding *Salmonella* prevalence in western Canadian swine farms in general. This study does, however, indicate that the breeding herd plays an important role in the persistence of *Salmonella* infection within pig herds, as suggested by other researchers (8,16). Molecular fingerprinting methods are needed to confirm clonal spread of *Salmonella* from sows to other production phases within these herds. In summary, the study herein has contributed to future surveillance and control efforts by providing important insight into the on-farm epidemiology of *Salmonella* in western Canada.

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