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Herd-level risk factors for subclinical *Salmonella* infection in European finishing-pig herds

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Abstract

Our objective was to find herd factors associated with pigs testing seropositive for Salmonella. Data were collected from 359 finishing-pig herds in Germany, Denmark, Greece, The Netherlands and Sweden, between 1996 and 1998. Pigs fed non-pelleted feed (dry or wet) had 2- and 2.5-times lower odds of seropositivity, compared to pigs fed pelleted feed. The protective effect of non-pelleted feed over pelleted feed may be ascribed to the structure and composition. Also, pigs that were given whey (to drink or as the liquid part of the diet) had 2.6-times lower odds to test seropositive than pigs not getting whey. Pigs produced in batches in herds with hygienic-lock facilities had >3-times lower odds for testing seropositive compared to pigs in herds where only one or neither factor was present. In herds where the caretaker(s) washed hands consistently before tending to the animals, pigs had 1.5-times lower odds of seropositivity than pigs in herds where the caretaker did not. Pigs which were able to have snout contact with pigs in neighbouring pens (because pen separations were either open or too low) had 1.7-times higher odds to test seropositive compared to pigs for which such contact was prevented. Pigs in herds recruiting from more than three supplier herds had three-times higher

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odds to test seropositive than pigs in herds which breed their own replacement stock or recruit from a maximum of three supplier herds.

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1. Introduction

Over the last decade, pork has gained recognition as a source of human salmonellosis (Lo Fo Wong et al., 2002). Salmonella has been identified in all stages of pork production. This means that efforts to decrease the Salmonella burden on society could be targeted at various stages of the production chain. An increasing number of European countries are focussing on the primary pork production phase. One of the greatest challenges of this approach lies in identifying effective intervention and control measures that can be taken at the herd level. Therefore, management strategies and production processes that can increase the risk of introduction and transmission of Salmonella need to be investigated.

Our objective was to identify herd factors associated with the detection of antibodies against Salmonella O-antigens in finishing pigs (henceforth referred to as "seropositivity"). The investigation was part of an international research programme, mainly sponsored by the European Commission, entitled 'Salmonella in Pork' or SALINPORK (Lo Fo Wong and Hald, 2000). A rather unique property of this study was that the same protocol was used in Germany, Denmark, Greece, The Netherlands and Sweden—allowing for comparison across countries and over-all modelling of Salmonella epidemiology. By combining data from all the participating countries, the statistical power of the study was increased. The country-specific risk-factor analyses are published elsewhere (von Altrock et al., 2000; Stege et al., 2001; van der Wolf et al., 2001a; Leontides et al., 2002).

2. Materials and methods

2.1. Collection and analysis of blood samples

Between October 1996 and June 1998, standardised seroprevalence studies were performed of *Salmonella* in finishing-pig herds in Germany, Greece and Sweden, based on a Danish seroprevalence study which was ongoing at the time (Stege et al., 2000a). Serological profiles were made of finishing-pig herds to determine their subclinical *Salmonella* status (Lo Fo Wong et al., 2001a). The results from these studies were used as the herd-status outcome in this study. In addition to data from these three countries, data from the Danish study and from a closely related study in The Netherlands (van der Wolf et al., 2001b) were made available.

Sample sizes were subject to practical and financial constraints. However, minimum sample sizes were required from participating countries. Herd prevalences could be estimated, with a serological test with a sensitivity of 95% (Nielsen et al., 1995) and 95% confidence with an accepted error of $\pm 13\%$, by testing 57 herds (Martin et al., 1987). Fifty blood

samples are sufficient to detect at least one (subclinically) infected animal with 95% confidence when the expected within-herd seroprevalence is \geq 6% (Cannon and Roe, 1982), as well as to estimate the within-herd seroprevalence with an accepted error of \pm 14% when the seroprevalence is expected to be 50%. Thus, each country agreed on selecting at least 60 finishing-pig herds and collecting 50 blood samples from animals close to or at slaughter from each herd.

Danish, German, Greek and Swedish blood samples were analysed for antibodies against *Salmonella O*-antigens with the Danish *Salmonella* mix-ELISA (Nielsen et al., 1995) and the Dutch samples with the Dutch *Salmonella* mix-ELISA (van der Heijden et al., 1998). Both mix-ELISA tests are based on the combination of *Salmonella O*-antigens 1, 4, 5, 6, 7 and 12. The mix-ELISA should detect *Salmonella* infections with serotypes belonging to serogroups B, C1 and D1. Results are expressed in optical-density percentage (OD%) of a known positive control, where samples with OD% > 10 were considered positive. At this level, Nielsen et al. (1995) reported the (experimental) test sensitivity to be 95%. Based on collaborative diagnostic work on the same batch of serum samples, the Danish and Dutch mix-ELISA were intercalibrated so that serological results obtained by these tests could be compared (i.e. Dutch OD% were divided by 2.4).

2.2. Collection of questionnaire data

A standardised risk-factor questionnaire was developed, based on work done by Stege et al. (2001) (in English, available upon request from corresponding the author), including 72 questions on herd size and type, housing conditions, management practice, feeding practice, hygiene practice, cleaning and disinfection procedures, health and disease prevention, and production parameters (Table 1). Fifty-eight percent of the questions (42) were closed (e.g. yes/no, always/frequently/seldom/never or pre-set options), 32% were semi-closed (e.g. information on days, number of days or animals, frequencies of certain procedures) and 10% were open-ended (e.g. listing product names, diseases, descriptions). Definitions of facilities and practices were discussed and standardised at a workshop for study participants. In this context, hygienic-lock facilities were defined as a changing room (i.e. locker) in connection to the pig houses where caretakers can take hygienic precautions before entering the herd. After consensus on the contents and interpretation of the questionnaire was achieved among the participating researchers, a data-entry program was developed in Epi Info 6.03 (Dean et al., 1990) and distributed, to ensure standardised data recording and data management. In total, 370 herds were involved in this study. All herds were visited where herd owners were presented with the risk-factor questionnaire in a personal interview. The interviewers had no prior knowledge of the Salmonella status of the herds at the time of questioning.

2.3. Data validation

The following procedures were applied to increase data quality in this study:

• *Standardise procedure:* the purpose and interpretation of all questions was discussed with the interviewers in an international workshop.

Table 1 Summary of factors that were covered by the risk factor survey on seropositivity for *Salmonella* conducted in 359 finishing-pig herds in Germany, Denmark, Greece, The Netherlands and Sweden between 1996 and 1998

Subject	Factors Type and number of animals present (sows, boars, gilts, growers, finishers)			
Herd type				
Housing	Number and size of accommodations, type of separation between units, type of pen separations, possibility for snout contact between pens, type of floor, partitions close-fitted to floor, quarantine facilities, hygienic-lock facilities			
Management practice	Age and weight at transfer to finishing unit, frequency of transfer, mean number per transfer, transfer through occupied pig house sections, mean age at slaughter, frequency of delivering to slaughterhouse, mean batch size per delivery, pigs moved to occupied pens, recruitment of animals, number of suppliers, frequency of purchasing animals, mean number of animals per purchase, placing back of sick or slow growing animals, culling of breeding animals, all in/all out production			
Feeding practice	Pelleted or non-pelleted feed, wet or dry feed, purchased or home-mixed feed, add acid to feed or drinking water, add whey, change in feeding and if so when, feed supplier, ad libitum or restricted feeding, structure of feed, pH of wet feed, use of straw litter, feeding with pro-biotics, feeding with growth promoters, other feed additives, Cu-content of feed, crude protein content in feed			
Hygiene practice	Changing clothes, changing footwear, washing hands			
Cleaning and disinfection procedures	Manure-free cleaning, cleaning between batches, disinfection after cleaning, type of disinfectant, pens empty after cleaning			
Health and disease prevention	Certified free and from which diseases, previous detection of <i>Salmonella</i> , diarrhoea problems last 3 months, dose and duration of antibiotic treatment, health status of suppliers			
Production parameters	Mean daily weight gain, mean slaughter weight			

- First-hand check: the interviewers were to check some of the factors while going through the pig houses, using a standard check sheet (in English, available from corresponding author upon request).
- Cross referral: questions relating to the same (herd) factors were cross-checked for consistency.
- Data-entry check: the data-entry program contained a check-file, which for a number of questions only accepted predetermined options or ranges.

Finally, unlikely answers were identified (e.g. misinterpretation, error in data entry) and confirmed with the producer in question.

2.4. Statistical method; logistic-regression model

Serological results were combined with the questionnaire information. The outcome variable was defined as the number of seropositive samples out of the number of samples

taken per herd (i.e. p/n). Continuous explanatory variables were tested for a linear relationship with the outcome. If this assumption was violated, transformation or categorisation in roughly equal sized groups was attempted. For categorical variables with more than two levels, statistical difference between levels could be tested using the contrast statement in PROC GENMOD in SAS (SAS Institute, 1996). If no significant difference was found between successive ordinal levels, these were collapsed into one.

For the screening process of candidate variables for multivariable modelling, a tri-variable approach was used, as suggested by Martin (1997). The two variables hypothesised to be important for the final model were country of origin (COUNTRY) and feed type (FEED). The factor for country was believed to contain a complex of factors, most of which were likely not measured (e.g. certain country-specific traditions or attitudes, infrastructure) which could influence the association between the herd factors of interest (i.e. those factors which could be manipulated for intervention and control purposes) and seropositivity. FEED was forced into the model from the beginning because other studies found that the type of feed used had an influence on the *Salmonella* status of pigs (e.g. Stege et al., 1997; Dahl, 1997; Dahl et al., 1999; Jørgensen et al., 1999). Three factors in the questionnaire addressed feed (i.e. wet versus dry (WETORDRY); home-mixed versus purchased (HOMEMIX); pelleted versus non-pelleted (PELLET)). These were checked for collinearity by calculating Spearman correlation coefficients. The factors HOMEMIX and PELLET were highly correlated ($\rho = 0.82$). FEED was constructed from WETORDRY and PELLET as a three-level parameter (wet, pelleted feed was not observed).

All variables in the risk-factor questionnaire were screened by logistic regression, using PROC GENMOD in SAS (SAS Institute, 1996), together with these two factors. A significance level of 0.25 (likelihood-ratio χ^2) was used as the screening criterion, based on work by Mickey and Greenland (1989) who showed that a more-traditional level (such as P < 0.05) often fails to identify variables known to be important.

All variables with P < 0.25 in the tri-variable models then were offered to the full model. This model was reduced by backwards elimination (Hosmer and Lemeshow, 1989), until the remaining parameter estimates had a significance level of approximately ≤ 0.15 . Two-factor interactions were created between all remaining variables and offered to the model. A significant interaction between two dichotomous variables was recoded as one new variable with four levels. Then, backwards elimination was continued to reduce the number of non-significant terms. When only significant variables (P < 0.05) were left in the reduced model, a stepwise forward selection process was performed, i.e. previously deleted variables were offered to the final model one at the time. This was done to ensure that any of the variables excluded earlier during the backward-elimination process but adding significantly to the final model were included.

Based on the intra-class correlation coefficients found by van der Wolf et al. (2001b) and Lo Fo Wong et al. (2001a), we expected that pigs within herds were more alike than pigs from different herds, due to the infectious nature of salmonellosis and exposure to common herd factors. This would lead to a variation in excess to that assumed by the binomial variance. Overdispersion in the data, because of clustering at the herd level, was adjusted for by allowing an overdispersion factor to inflate the variance. The overdispersion factor (i.e. scale parameter) was estimated based on Pearson's χ^2 -statistic (P scale option, PROC GENMOD, SAS Institute, 1996).

Table 2
Risk factors selected for multivariable analysis by tri-variable screening analyses (with factors COUNTRY and FEED) for association with seropositivity for *Salmonella* in 359 finishing-pig herds in Germany, Denmark, Greece, The Netherlands and Sweden (1996–1998)

Definition of factor	Level	Number of herds	
Adding acid to water or feed ^a	Feed	33	
ridding held to water or rece	Water	6	
	No	320	
Feeding home-mixed or purchased feedb	Home-mixed	165	
1 couling nome maked of purewasses some	Purchased	194	
Provide whey to pigs (to drink or as liquid part of feed) ^c	Yes	36	
Trovide whey to pigo (to diffin of all 1/4-12 pins)	No	227	
Batch production (all in/all out)	Yes	171	
Daten production (an insun out)	No	188	
Hygienic-lock facilities	Yes	219	
Trygiome-lock lacinities	No	140	
Herd declared free from any diseases ^c	Yes	180	
Tiere declared from any diseases	No	82	
Washing hands consistently ^c	Yes	124	
Trushing hards completely	No	175	
Quarantine facilities present	Yes	59	
•	No	300	
Snout contact between pens	Yes	324	
Broth bornaon bornaon pene	No	35	
Number of pig suppliers used	>3	30	
14mmoor or hig suppliers asset	0-3	329	

^a One indication as 'both water and feed' was recoded as 'feed'.

3. Results

Twelve explanatory variables (including COUNTRY and FEED) were considered for analysis in the full model (Table 2). Eleven herds were excluded from the final analysis because of missing data for one or more variables in the final model. The final model is based on results from 19,330 serological tests from 359 pig herds: 60 from Germany, 96 from Denmark, 49 from Greece, 94 from The Netherlands and 60 from Sweden. The median sample size per herd was 50 (Q3 – Q1 = 8), with a minimum of 32 and a maximum of 136. In 272 herds (76%), one or more seroreactors were found. The median within-herd seroprevalence was 8% (Q3 – Q1 = 22%). Fig. 1 shows the over-all frequency distribution of the within-herd seroprevalence of all herds that were included in the final model, at cut-off OD% > 10.

^b Home-mixed feed tested in bivariate model with COUNTRY.

^c Information not obtained from all herds. Missing observations were kept in the analysis as separate levels. None of the levels coded 'missing' were significantly associated with the outcome.

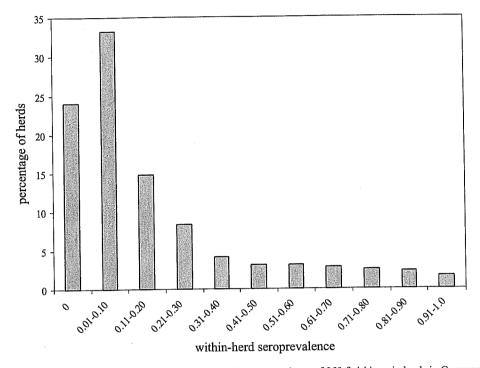


Fig. 1. Frequency distribution of within-herd *Salmonella* seroprevalence of 359 finishing-pig herds in Germany, Denmark, Greece, The Netherlands and Sweden between 1996 and 1998 (cut-off: OD% > 10).

Factors which were discarded from modelling already at the screening stage included: restricted feeding versus ad libitum; addition of antibiotic growth promoters to the feed; addition of feed additives other than antibiotic growth promoters; open versus closed pen separations; the presence of slatted floors; the use of straw; changing boots and clothes when entering the herd; the use of probiotics; frequency and number of pigs purchased; whether the supplier herd is declared free of any diseases; average daily weight gain; slaughter age and weight; pig-house cleaning procedures.

COUNTRY contributed significantly to the final model (P < 0.0001), thereby accounting for between-country differences in the final model. Because of differences in how well the study population is representative for each country in our study, direct comparison between countries might be invalid. Therefore, COUNTRY is reported anonymously. Six factors (one a recoded interaction between two factors) were associated with seropositivity for Salmonella in finishing pigs (Table 3). Risk factors associated with higher odds of seropositivity were: pelleted, dry feed; snout contact with pigs from adjacent pens; not both producing pigs in batches and having hygienic-lock facilities (interaction); recruited pigs from more than three supplier herds; not giving whey; and the caretakers not washing hands consistently before tending to the animals. The factor home-mixed feed versus purchased feed was not offered to the model to avoid multicollinearity with the factor FEED.

Table 3 Final multivariable model of herd factors associated with seropositivity for Salmonella in 359 finishing-pig herds in Germany, Denmark, Greece, The Netherlands and Sweden between 1996 and 1998, at sample cut-off OD% > 10

Variable	Parameter	b	S.E. (b)	OR	CI95 ^a	P value
Country ^b	Α	2.01	0.42	7.43	3.40, 17.48	< 0.0001
	В	0.47	0.52	1.59	0.60, 4.58	
	C	0.30	0.39	1.35	0.63, 2.96	
	D	0.04	0.45	1.04	0.44, 2.55	
	E	0	0	1	_	
Type of feed	Non-pelleted and dry	-0.56	0.24	0.57°	0.36, 0.90	0.0007
	Non-pelleted and wet	-0.87	0.26	0.42^{c}	0.25, 0.68	
	Pelleted and dry	0	0	1	_	
Snout contact between pens	Yes	0.51	0.26	1.66	1.01, 2.85	0.05
	No	0	0	1	_	
Batch production and hygienic-lock facilities	Both	-1.30	0.33	0.27	0.14, 0.51	<0.0001
	One or neither	0	0	1	- '	
Number of supplier herds	>3	1.19	0.37	3.28	1.60, 6.79	0.001
	0–3	0	0	1	-	
Use of whey	Yes	-0.93	0.42	0.39	0.16, 0.86	0.02
	No	0	0	1	_	
Washing hands consistently	Yes	-0.40	0.20	0.67	0.45, 1.00	0.05
	No	0	0	1	-	

Model statistics: intercept, -2.56; model deviance, 5440.6; d.f., 347; scale parameter, 3.95.

4. Discussion

4.1. The risk factors

The purpose of the pelleting- and heat-treatment process is to reduce Salmonella contamination in compound feed. Although Salmonella seldom is isolated from pelleted (heat-treated) feed at the feed mills (Anonymous, 1999), feed can become re-contaminated with Salmonella during transport (Fedorka-Cray et al., 1997), storage or in the feeding system at the farm. This is supported by the fact that Salmonella could be isolated from pelleted feed sampled from the feeding outlet in pig pens (Lo Fo Wong et al., 2001b).

Jørgensen et al. (1999) found that the population of lactic-acid bacteria in the stomach contents was smallest in pigs receiving pelleted feed, intermediate in pigs fed a non-pelleted heat-treated grain, and largest in pigs receiving meal. Based on this, they hypothesised that non-pelleted feed results in a microbiological ecosystem that provides *Salmonella* with poor growing conditions compared to pelleted feed. In the same study by Jørgensen et al. (1999), no association was found between (the degree of) heat treatment and seropositivity in pigs suggesting that the protective effect of non-pelleted feed compared to pelleted feed

^a Likelihood-ratio based 95% confidence limits.

^b Countries listed in random order.

 $^{^{\}rm c}$ No significant difference between non-pelleted FEED levels (P=0.30).

might (in part) be explained by differences in the coarseness of the feed (Kjeldsen and Dahl, 1999). Coarsely ground grain might not digest as well as more-finely ground pelleted feed; therefore, some part of the carbohydrates of coarsely ground grain will ferment in the large intestine. This results in the forming of volatile fatty acids, creating a hostile environment for *Salmonella* (low pH, organic acids, competitive exclusion, etc.) (Reid et al., 1996, 1998; Silvi et al., 1999; Reid and Hillman, 1999). Dahl et al. (1999) found a significant protective effect of feeding pelleted feed where non-heat treated wheat or barley was added to the feed after pelleting compared to conventional pelleted feed.

Even though the two feed-related factors investigated in this study (wet/dry and pelleted/ non-pelleted) were combined into one variable, their effect could be estimated separately by testing the appropriate contrast between levels of the new variable. Although the protective effect of non-pelleted, wet feed appeared stronger than the effect of non-pelleted, dry feed, no significant difference could be shown between these two types of feed (P = 0.30). However, a decrease in risk when using wet feed compared to dry feed has been shown in other studies (e.g. Bager, 1994; Stege et al., 1997; Dahl, 1997; Beloeil et al., 1999; van der Wolf et al., 2001a). Several explanations have been offered, including that during a natural fermentation process in wet feed, the pH is lowered due to the production of lactic acid and acetic acid by lactic-acid producing bacteria and the growth of yeasts (which inhibits growth of Salmonella on the feed) (van Winsen et al., 2001) or at least reduces the numbers to beneath the detection limit. It is likely that the protective effect of whey found in this study is based on the same principle. The protective effect of organic acids in whey and fermented by-products against (subclinical) Salmonella infections has been discussed in a number of papers and is considered as a possible intervention method at the herd level (van Schie, 1987; van Winsen et al., 2001; Dahl, 1997; van der Wolf et al., 2001c). No association between adding acid to either water or feed and seropositivity could be found in this study. This association is difficult to test in risk-factor studies because organic acids commonly are used both as a protective and therapeutic measure against E. coli and possibly Salmonella infections. However, in targeted studies, a beneficial effect has been demonstrated of the administration of organic acids in feed (Wingstrand et al., 1997; van Winsen et al., 2001) and water (van der Wolf et al., 2001c) (reduced Salmonella prevalence in pigs).

Faecal-oral transmission is the most-likely mode of transmission of virulent Salmonellae (Schwartz, 1999). Therefore, we hypothesised that preventing spread of manure between pens would prevent spread of infection. Dahl et al. (1996) showed that closed pen separations posed a barrier which prevented faecal contact between adjacent pens and thereby spread of infection. However, we did not find open pen separations to be significant. A factor added to the questionnaire for cross-checking purposes was 'the possibility for pigs from different pens to have snout contact'. Twelve farmers indicated that, though their pens had closed separations, pigs still were able to have snout contact (most likely by standing on their hind legs and interacting with pigs in the adjacent pen(s)). Snout contact was significantly associated with seropositivity.

The purpose of a hygienic-lock facility is to prevent the introduction of pathogens into the herd through the caretaker(s) and visitors. This room is supposed to be used to change clothes and footwear and possibly has sanitary facilities such as a sink, a toilet or a shower. Batch production commonly is accepted as an important aspect of good farming practice.

Producing pigs according to the all in/all out principle might not prevent an introduction of an infection into the herd, but it can help prevent cross-contamination between batches and allows for cleaning and disinfection between batches. However, few studies have been able to demonstrate any benefit from general hygienic measures or batch production over continuous production (e.g. Stege et al., 2001). Our findings support the general belief in a beneficial effect of batch production only in combination with the presence of hygienic-lock facilities. However, this does not explain why a protective effect of consistently washing hands could be demonstrated—but other hygienic parameters (such as changing clothes and/or footwear when entering the herd) did not contribute significantly to the model. No interaction or multicollinearity between these factors and the other hygienic parameters in the study was found. It might be that pathogens are easier spread via hands than clothes or footwear. On the other hand, it is also possible that what is measured here, is a level of awareness of pig producers and a certain mentality towards hygiene. The latter also was suggested by Funk et al. (2001), who found that the presence of a toilet on site constituted a protective effect on the Salmonella status of the herd.

Finishing-pig herds are not closed systems. Even integrated herds are subject to introduction of feed or even replacement stock. We found no beneficial effect of managing an integrated or closed herd. Some of the potentially beneficial effect of being an integrated herd might have been captured in the analysis by the factor 'number of supplier herds' but after removing this factor from the analysis, there was no association of managing a closed herd with seropositivity. We found that recruiting pigs from more than three supplier herds was a risk factor which makes biological sense (the more herds supply animals to one herd, the larger the probability of introducing an infection into the herd through one of these contacts). That the critical number of supplier herds seemed to be three might be because in The Netherlands, certified pig herds were restricted to have a maximum of three supplier herds (at the time this study was performed).

Herd size is a complex factor that also includes all related factors (such as the frequency of introduction of animals, the number of supplier herds, the size of the production area, the number of units, and a larger supply of feed and water). That an increased herd size imposes an increased risk of *Salmonella* infection only few studies have confirmed (Dahl, 1997; Carstensen and Christensen, 1998) and we also found no association. However, an increase in herd size does not necessarily mean an increase in pig density at the pen level. In most countries, pen density is restricted by legislation and larger herds basically are comprised of more epidemiological units. In fact, larger operations might have the resources necessary for the implementation of effective biosecurity measures and good manufacturing practice. That van der Wolf et al. (2001a) found an increased risk for *Salmonella* in small-to-moderate-size herds (<800 finishers) compared to large herds would support this.

We found no association where other studies had: e.g. slatted flooring (Davies, 1998), an increased risk with use of antibiotics (Berends et al., 1996), a positive effect of declaration of freedom of disease other than *Salmonella* (SPF-status versus conventional; Dahl, 1997), disinfection after cleaning between batches (Stege et al., 1997; van der Wolf et al., 2001a) and the use of straw (in poultry; Bhatia et al., 1979). It is not clear why these associations were not found in our study or in some others (e.g. Baum et al., 1998; Funk et al., 2001). Perhaps, some of the effects are masked or accounted for by other variables in the study.

However, test sensitivity and specificity, primary and secondary sample size, and modelling strategy all can influence the final model (Lo Fo Wong and Dahl, 2001).

4.2. Sensitivity and specificity issues

The Danish mix-ELISA test (used for samples from Germany, Denmark, Greece and Sweden) was developed to detect most of Salmonella serotypes prevalent in Denmark (Nielsen et al., 1995; Mousing et al., 1997). Perhaps other countries have different spectra of Salmonella serotypes or cross-reacting bacteria. In the SALINPORK project on sera originating from Sweden (Wiuff et al., 2001), serological results did not correlate to bacteriological findings (e.g. Anonymous, 1995; Wahlström et al., 1997) (although the reactions might have been caused by low-level Salmonella infections). In non-endemic countries, herd-management factors otherwise identified as risk factors are not significant simply because Salmonella is absent. However, excluding Swedish data from our study resulted in a final model with only slightly stronger associations between the factors found and seropositivity and led to the exclusion of 'the number of supplier herds' as a risk factor (P = 0.13). So, we argue that by including the Swedish data we were conservative. Also, including the country of origin in the analysis should have accounted for differences in test performance and unmeasured country-specific management characteristics. Finally, the same analysis procedure as presented here was performed using a test cut-off of 40 OD% (Lo Fo Wong et al., 2000); this resulted in the same final model—only with stronger associations.

5. Conclusions

Allowing pigs from adjacent pens to have snout contact and purchasing pigs from more than three suppliers were associated with increased odds of seropositivity for *Salmonella*. Feeding non-pelleted feed, a combination of batch production and the presence of hygienic-lock facilities, the use of whey and washing hands consistently when tending to the animals were protective.

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