

A study of the distribution of *Salmonella* serovars in an integrated pig company

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Abstract

A total of 3220 faecal samples from 161 pig farms (rearing and finishing units) belonging to an integrated pig enterprise were collected over a period of 18 months. *Salmonella* was found in 630 (19.5%) of the samples. At the farm level, 111 of 161 premises (69%) had at least one *Salmonella*-positive sample. 72.8% of rearing units and 66.6% of finishing units were positive for *Salmonella*; 61.4% of isolates were *S. Typhimurium* (387/630 isolates), and 25% of isolates were *S. Derby* (157/630). *S. Panama*, which was the third most common serovar (4.9% of isolates), is rarely found in pigs or other animals in the UK and appeared to be largely specific to this company, being found in the multiplier herd as well. A total of sixteen serovars were recorded within the study. Many of the serovars were found in breeding and multiplier herds from the company over several years, indicating that they were likely to be persisting or circulating within the integration. This study was carried out before the appearance of monophasic *S. Typhimurium* strains in pigs in the UK, and these were not found in the survey, but are now regularly reported from UK surveillance of pig herds, including from the integration involved in this study. Risk factor analysis suggested that increasing age of the pig farm was associated with increased likelihood of the presence of *Salmonella* and that finishing farms housing less than 500 pigs were associated with lower levels of *S. Typhimurium* than larger finishing farms.

Introduction

It is widely accepted that contaminated pork may be an important source of human salmonellosis, and it has been estimated in various European countries that 10–20% of all cases of salmonellosis in humans are related to the consumption of pork (Borch et al., 1996; Berends et al., 1998; Steinbach & Hartung, 1999; EFSA, 2010). Pork-related outbreaks of non-typhoidal salmonellosis in humans with fatal outcome have been described (Jansen et al., 2007).

Pigs can be infected by several *Salmonella* serovars, and the occurrence of these is also partly geographically determined (Loynachan et al., 2004; Fedorka-Cray et al., 2000). All serovars isolated from pigs are considered to be a potential hazard for public health by the European food safety authority (EFSA) (EFSA, 2006), although it is recognised that most individual serovars, apart from *S. Typhimurium* and possibly *S. Infantis* in some countries, currently play a minor part in human infections.

The occurrence of *Salmonella* serovars in pigs has changed significantly over the past decades. While *Salmonella Choleraesuis* was the predominant serovar in the 1950s and 1960s, it is rarely seen in Western Europe nowadays. Currently, the most commonly isolated non-typhoidal serovars in pigs and pork are *Salmonella Typhimurium*, including variant Copenhagen, and *Salmonella Derby* (Letellier et al., 1999; Davies et al., 2004; Gebreyes et al., 2004; Valdezate et al., 2005; EFSA, 2006; Rostagno et al., 2007). However, during the past five years, reports of monophasic strains of *Salmonella Typhimurium* (*S.* 4,5,12:i:- and *S.* 4,12:i:-) from pigs and humans have become more frequent in various European countries. The public health risk of *Salmonella* infection from consumption of contaminated pork depends on multiple factors including the level of infection in the pig herd (Nollet et al., 2005; Hill et al., 2003).

The aim of this study was to determine the extent of *Salmonella* infection within one big company and to monitor the change in serovars over a period of 14 years. During the initial study, a questionnaire was used aiming at identifying risk factors associated with the presence of *Salmonella*.

The main part of this study was undertaken between 1996 and 1998. Over the following years, several visits were done to multiplier, breeding, rearing and finishing farms belonging to the company, with the last visits to a breeding herd and two finishing herds done in 2010/2011. In addition, reports from voluntary surveillance within the company were assessed. The long period of time over which the study was performed offered a unique opportunity to investigate the persistence, disappearance and introduction of certain *Salmonella* serovars within the company.

Material and Methods

The first part of the study consisted of a survey including 161 farms belonging to the same integrated pig company. Of those, 59 were rearing farms and 102 were finishing farms. No nucleus, multipliers or breeder herds were included in the survey. Each farm was visited once by a veterinary surgeon who interviewed the farmer and completed a questionnaire containing sections on demography, farm structure and management details, herd details, antibiotic therapy and disease security/hygiene measures.

Samples of pooled faeces (10-15g) were collected from pig accommodation at up to twenty locations on the farm. Faeces samples were examined for Salmonella using a standard technique (Davies & Wray, 1997). Briefly; contents of the sample pot were transferred to a honey jar containing 225 ml buffered peptone water and incubated at 37°C for 18hrs +2hrs. 0.2ml of this culture was inoculated into the centre of a DIASALM agar plate and incubated at 41.5°C for 24 and 48 hours. The DIASALM cultures were subcultured to Rambach agar and incubated for 24 ± 4hrs. Suspect colonies of Salmonella were screened using polyvalent O and H antisera and later subjected to full serotyping.

After the initial study had finished, selected breeder and finisher farms belonging to the integration were visited periodically for 3 years, with some additional visits done in 2010/2011. During those visits, the following samples were taken: swabs of wallows and bird droppings and individual faecal samples (at least 60 samples per group). The samples were analysed as above, but during 2010/11 the DIASALM had been replaced by MSRV, to harmonise with EU requirements for primary production testing.

Results

Survey results:

A total of 161 farms were included in the survey and 3220 pooled pen faeces samples were collected and tested for the presence of Salmonella. Of the 3220 samples examined, 630 (19.5%) tested positive for Salmonella spp. *S. Typhimurium* (ST) was found in 387 (12%) of samples making it the most common serovar. A list of the serovars and the frequency with which they were found is shown in table 1. As this part of the study was conducted before the emergence of monophasic strains of *S. Typhimurium* in pigs in the UK, no such isolates were found, but 3 isolates of an aphasic group B strain were identified.

One hundred and eleven farms (111) (68.9%) had at least one Salmonella-positive sample; 43 of those farms were rearing farms and 68 were finishing farms.

The rearing farms in the survey ranged in size from 200 to over 2000 pig places, the mode being 500-800. Forty-three of the 59 (72.8%) rearing units had Salmonella. Of these, 35 (59.3%) were positive for *S. Typhimurium*.

The finishing farms in the survey also ranged in size from less than 200 to over 2,000 pig places. The mode in this case was 800-1,100. Sixty-eight of the 102 (66.6%) finisher farms had Salmonella. Of these 56 (54.9%) were positive for *S. Typhimurium*.

Although Salmonella was isolated from 68.9% of farms, all farms were not affected to the same degree. The proportion of samples positive for Salmonella at the farm level ranged from 5% to 95%.

The length of time for which a farm had been keeping pigs seemed to be a risk factor as such that farms which have had pigs for more than 30 years were more likely to be positive for Salmonella than farms which have had pigs for less than five years.

Over the next three years, a multiplier herd, two breeding herds and five rearing/finishing herds were visited and the same serovars were found as in the survey. While *S. Typhimurium* and *S. Derby* are commonly seen in pigs in the UK, *S. Panama* appeared to be a company-specific serovar which is rarely found in other pig companies.

Visits in 2010/2011:

In 2010/2011, two finishing sites and one breeding site from the company were visited twice each.

The first visit to one finishing herd (outdoor) resulted in a high percentage of Salmonella-positive samples (338/429; 78.8%), with 57.8% of all isolates which were serotyped being *S. London*. The second most prevalent serovar was *S. 4,5,12:i:-* with 24.1% of all serotyped isolates. Other serovars found, but at low numbers, were *S. 4,12:i:-*, *S. Bardo*, *S. Panama* and *S. Newport*. Shortly after the visit, the herd was moved to a different field, and at the second visit, the number of Salmonella-positive samples had dropped significantly to 16.8% (87/518). Of the isolates which were serotyped, 38% were *S. 4,5,12:i:-*, 23.8% were *S. 4,12:i:-*, 21.4% were *S. Typhimurium* and 14.3% were *S. London*. In the other (indoor) finishing herd *S. 4,5,12:i:-* and ST that were identified at the first visit were replaced by *S. Bovismorbificans* and *S. London* at the second visit.

The first visit to the breeding site resulted in a low percentage of Salmonella-positive samples (30/573; 5.2%). Of the 27 serotyped isolates, 10 were *S. London*, 8 were *S. 4,5,12:i:-*, 4 were *S. 4,12:i:-*, 3 were *S. Bardo* (a variant of *S. Newport*) and 2 were *S. Newport*. Similar serovars with the addition of ST, *S. Bovismorbificans* and *S. Panama* were found at the second visit.

Discussion

Results from the survey show, that, in the case of finishing farms, the proportion of farms with *S. Typhimurium* was lower (32.0%) in farms with fewer than 500 pigs than in farms with more than 500 pigs (62.8%). This may be related to the greater diversity of sources of pigs for larger batch farms and suggests that restriction of unit size could be a possible control measure, albeit economically disadvantageous. The length of time that the holding had been in operation as a pig farm was identified as a risk factor, which suggests that long term persistence of Salmonella on farms is an important factor in the epidemiology of infection. Especially in the case of *S. Panama*, which is hardly ever found in the UK apart from this one company, persistence within the company seems to have occurred for more than 14 years. The emergence of monophasic strains of *S. Typhimurium* could be seen within this company and seems to follow a national trend, with more than 7 % of all Salmonella isolates from pigs from the UK being either *S. 4,5,12:i:-* or *S. 4,12:i:-* in 2009, and preliminary data for 2010 supporting this trend. During the two visits in 2010/11 to a finisher herd, a significant shift in serovars was found between the first and the second visit. This might be due to several factors; first, the herd was moved to a different field between visits, which had not been used for pigs for a long time. Therefore, contamination of the soil with Salmonella was presumably very low or even negligible. Even though the pigs supplied to this finishing site came from the same breeding site, different batches of pigs might have different serovars and there was some shift in serovars within the breeding herd at the same time. There might also be a difference in susceptibility of different age groups to different serovars or sequential replacement of serovars according to exposure and partial immunity and clearance.

Conclusion

This study showed that some Salmonella serovars can persist in an integrated pig company over many years. In particular, *S. Panama*, which is a very company-specific serovar, was observed over a period of 14 years as a fairly low proportion of all serotyped isolates.

The emergency of monophasic strains of *S. Typhimurium* (namely *S. 4,5,12:i:-* and *S. 4,12:i:-*) could also be observed and followed a national trend, whereas the prevalence of *S. Typhimurium* went down at the same time.

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Table 1. Salmonella serovars found during the survey and numbers of isolates

Serovar	Number of isolates:
<i>S. Typhimurium</i>	387
<i>S. Derby</i>	157
<i>S. Panama</i>	31
<i>S. Goldcoast</i>	19
<i>S. Reading</i>	14
<i>S. Anatum</i>	4
<i>S. London</i>	5
<i>S. Agona</i>	2
<i>S. Manhattan</i>	2
<i>S. Enteritidis</i>	1
<i>S. Brandenburg</i>	1
<i>S. Bovismorbificans</i>	1
<i>S. Kentucky</i>	1
<i>S. Kimuenza</i>	1
<i>S. Schwarzengrund</i>	1
<i>S. 4,12:-- (untypable)</i>	3