BACTERIOLOGICAL AND SEROLOGICAL CHARACTERISATION OF SLAUGHTER PIGS FROM 25 SEROLOGICALLY IDENTIFIED "SALMONELLA HIGH RISK" HERDS

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Danish finishing herds are routinely screened for antibodies to Salmonella in random samples of meat juice from slaughter pigs. The herds are categorized by the seroprevalence of samples from the preceding three months into three infection levels (1, 2 and 3). Herds are allocated to level 3 ("Salmonella high risk" herds) at seroprevalences exceeding 33-50% depending on herd size (larger herds lower limit) (Mousing et al., in press).

In level 3 herds, special hygienic precautions are practised at slaughter. The precautions are mandatory and comprises separate transportation to slaughterhouse, altered slaughter procedure, bacteriological examination of carcasses and heat treatment or slating of highly contaminated carcasses.

Large expenses and troublesome coordination are associated with these precautions and led us to characterise level 3 herds bacteriologically and serologically and investigate the predictive value of bacteriological and/or serological parameters - in-herd or on carcasses - for a differentiation between level 3 herds in which slaughter pigs are actually culture negative ("historical" seroreaction) respectively culture positive for Salmonella.

MATERIALS AND METHODS

Sampling:

Herd: 25 level-3 herds (deliverance >=1000 pigs/year, 13 to abattoir X and 12 to abattoir Y).
Pen samples: In each herd 30 pools of faecal material (25g each) collected from pens housing slaughter pigs (70kg bw - slaughter weight).
During the test slaughter period (the following 2 months), 40 pigs from each herd (10 pigs every second week) were examined.
From each pig were collected: Faecal samples: Rectal contents (22g/sample). Pharyngeal swab: Superficial swabbing of the pharyngeal surface. Carcass swab: Swabbing 1400cm² of inner thigh and ventral abdominal cut-edge.
Meat juice samples: Screening samples from the test slaughter period, from the preceding 3 months and individual samples from the 40 test-slaughter carcasses.

Examinations:

Bacteriological examination: Pen samples, faeces, pharyngeal swabs were examined for Salmonella by initial non-selective pre-enrichment, selective enrichment in RV-broth, plating on MSRV agar plates, biochemical verification of Salmonella, serotyping and finally phage-typing of S. Typhimurium-isolates. For carcass-swabs an automatized ELISA-method

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was used involving non-selective pre-enrichment and selective enrichment.

*Antibodies to Salmonella in meat juice:* Indirect mix-ELISA (antigen: O-types 1,4,5,6,7, and 12) (Nielsen et al., submitted).

*Statistical significances* were set at p<0.05.

**RESULTS**

*Salmonella enterica* was isolated from pen samples in 21/25 level-3 herds. The proportion of culture positive pen samples is shown in figure 1. 35% of all pen samples were culture positive. *S. Typhimurium* was isolated from 19/21 culture positive herds (95.7% of the isolates), 66.4% of the *S. Typhimurium* isolates were DT12. Only 3 herds had more than one sero-/phage type.

Twenty-two out of 25 herds were culture positive in faecal samples from the carcasses, overall 32% of the faeces samples were culture positive, and sero- and phage types differing from isolates from pen samples were found in pigs from 15 herds.

No herds were culture negative in pharyngeal swabs, and only one herd was culture negative in carcass swabs. Compared to pen- and faecal samples the proportion of *S. Typhimurium* was reduced in pharyngeal and carcass swabs, and sero- and phage types differing from isolates from pen samples was found in pigs from 17 herds.

Figure 1. Proportion of culture positive pen samples/herd in 25 serologically identified *Salmonella* "high risk" herds.

Interactions between the individual results from bacteriological examination of faeces, pharynx and carcass (*S. Typhimurium, other Salmonella* or culture negative) and the serological result (high response or not) were compared on individual level (2-factor interactions shown in figure 2). The interaction between bacteriological results from pharynx and faeces was far the strongest. The interaction between seropositivity and bacteriological result from carcass was not significant. All parameter estimates were >1.

Figure 2. Significance-probabilities for all 2-factor interactions between bacteriological result of faeces, pharynx- and carcass-swab and seropositivity.

A positive and statistically significant correlation was found between % culture positive pen samples in a herd and % culture positive carcass swabs from the same group of pigs. The
correlation declined with increasing interval between sampling and test slaughter. A positive but not statistically significant correlation between the seroprevalence in varying periods prior to test slaughter and % faeces culture positive at first sampling was demonstrated. Similar positive but not significant correlations were found between % culture positive faecal samples at first test slaughter and at 2nd, 3rd or 4th test slaughter.

The bacteriological findings in faeces, pharynx and carcasses were compared two by two. Observations of culture negative in first sample combined with culture positive in second sample and observations with different Salmonella sero- and phage types in the samples were considered indicative for cross-contamination. The number of “cross-contaminations” on the abattoirs was compared in table 1. Only the proportion of cross-contaminations between faeces and pharynx differed significantly between abattoirs.

Table 1. Comparison of Salmonella findings on each abattoir indicating cross-contamination (culture negative to culture positive or different serotypes isolated from the samples)

<table>
<thead>
<tr>
<th>Pair of observations</th>
<th>No. “cross-contamination” observations/No. observations</th>
<th>Total</th>
<th>Abattoir X</th>
<th>Abattoir Y</th>
<th>Significance Level</th>
</tr>
</thead>
<tbody>
<tr>
<td>Faeces-Carcasses</td>
<td>136/881 (15%)</td>
<td>67/458 (15%)</td>
<td>69/423 (16%)</td>
<td></td>
<td>0.69</td>
</tr>
<tr>
<td>Faeces-Pharynx</td>
<td>252/706 (36%)</td>
<td>76/337 (23%)</td>
<td>176/369 (48%)</td>
<td></td>
<td>3.6x10^-6</td>
</tr>
<tr>
<td>Pharynx-Carcasses</td>
<td>101/842 (12%)</td>
<td>62/449 (14%)</td>
<td>39/393 (10%)</td>
<td></td>
<td>0.26</td>
</tr>
</tbody>
</table>

DISCUSSION AND CONCLUSION

The majority (21/25) of serologically identified “Salmonella high risk herds” were actually shedding Salmonella with S. Typhimurium DT12 as the dominating type. For safe prediction of the actual bacteriological status of slaughter pigs only bacteriological examination of faecal samples from slaughter pigs in the herd could be recommended. Strong indications of cross contamination of pharynx and carcasses excluded these as suitable measures.

Very strong interactions between bacteriological results from faeces and pharynx and between pharynx and carcass were found. In combination with significant differences between abattoirs in proportion of observations indicating faeces-pharynx cross-contamination led us to propose, that contamination of pharynx with faeces from other herds during transport or in pens at the abattoir might contribute significantly to contamination of the carcass. The abattoirs in this investigation both used bung-bag for prevention of direct faeces-to-carcass contamination at evisceration.

REFERENCES
