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SALMONELLA CONTROL IN SWINE, FOOD SAFETY PERSPECTIVES AND IMPACT ON THE SWINE INDUSTRY

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Summary
Salmonella infections in swine are common in many swine producing countries and are considered important from both a disease and a zoonotic point of view.
Most cases of Salmonella infections are sub clinical and clinical outbreaks of salmonellosis occur mainly due to infections with *S. Choleraesuis* and *S. Typhimurium*. In America and Asia *S. Choleraesuis* is still a major swine disease, while *S. Typhimurium* is now the most frequently isolated serotype among swine in Europe. *S. Typhimurium* has the ability to infect every species of birds and mammals including humans, which makes it an important zoonotic agent.
The effect of antibiotic treatment against Salmonella and possible risk for development of multiresistant strains is addressed. Non-antibiotic measures to reduce the prevalence of Salmonella have been identified in Denmark, 1) use of non heat treated meal feed, mainly barley, 2) fermented liquid feeding, 3) use of weak organic acids in feed or water, 4) all in - all out production with good cleaning and desinfection. Heat-treated pelleted feed has been identified as a major risk factor for active Salmonella infection.
In Denmark, a mandatory nation wide Salmonella Control Programme for swine has been in force since 1995 in order to reduce human salmonellosis. S. Typhimurium is isolated from 70% of the moderate to highly seropositive herds (3% of all herds). Due to characterization of isolates by sero-, phage-, DNA- and antibiotic sensibility typing, it is possible to identify routes of transmission from swine and pork to humans. In 1995, pork was estimated to be responsible for *1,100* cases of human S. Typhimurium-infections in Denmark. During the programme, the estimated numbers have decreased continuously. For year 2000 the estimated number was 166 human S. Typhimurium cases.
The EU zoonosis act is presently under negotiation, it will most likely include Salmonella in swine. In Germany, Ireland and United Kingdom initiatives have been taken to implement national Salmonella control programmes. In the USA surveillance of Salmonella at slaughter carcasses was started in 1995 making it possible for USDA to establish a national salmonella level on swine carcasses. This knowledge is use to define trade regulations for import of swine carcasses, which must meet the US standards.

Key words: Salmonella, swine, pork, Zoonosis, herd intervention, risk factors.

Salmonella in swine

Salmonella infections in swine are common in many swine producing countries and are considered important from both a disease and a zoonotic point of view.
In the US, it is estimated that 30-65% of the swine herds are Salmonella positive, while the level of infected herds in Canada is approximately 25%. In Europe, the herd prevalence's range from practically 0 in Sweden to 10-40% in several central European countries.

Many different serotypes are isolated from swine herds world wide, typically 30-50 serotypes are found every year in the major swine producing countries. However, a few serotypes dominate among the isolates.
In clinical submissions from sick or dead animals, the swine adapted *S. Choleraesuis* and the ubiquitous *S. Typhimurium* are the most frequently isolated types. *S. Typhimurium* has the ability to infect every species of birds and mammals including humans, which makes it an important zoonotic agent.
In America and Asia *S. Choleraesuis* infection is still a major swine disease, while *S. Typhimurium* is now the most frequently isolated serotype among swine in Europe. Several other serotypes like *S. Derby*, *S. Infantis*, and *S. Livingstone* may also cause clinical out break of salmonellosis.
However, most cases of Salmonella infections in swine are sub clinical, and are mainly of interest for zoonotic reasons. The swine get infected mainly by oral uptake of *Salmonella*, and excrete
Salmonella in feces consistently for 1 week to several weeks, followed by a period of weeks to many months with intermittent excretion in feces. A small proportion of the Salmonella infected swine become latent carries of Salmonella bacteria in the gut or internal organs.

Prevention and treatment of Salmonella infections in swine

Clinical salmonellosis can be prevented by use of vaccines. Outbreaks with *S. Choleraesuis* have now for many years been controlled with specific attenuated live *S. Choleraesuis* vaccines, both in Europe and in America. There is only limited experience with prevention of Salmonellosis by use of S. Typhimurium vaccines. In experimental studies, an acceptable good protection has been observed.

In the contrast, live and killed salmonella vaccines have so far not successfully been able to prevent sub clinical salmonella infections in swine. Consequently, vaccines may so far not be used to control sub clinical salmonella infections for food safety reasons.

Clinical salmonellosis typically decline rapidly after systemic antibiotic treatment. However, *Salmonella* have an unpleasant ability to develop antibiotic resistance. The proportion of resistant and multiresistant *Salmonella* isolates rapidly increases, if antibiotics frequently are used in the swine population. Consequently, antibiotic susceptibility testing of *Salmonella* isolates becomes more and more needed, in order to ensure a significant therapeutically effect.
In general antibiotic treatment should not be used to control sub clinical salmonella infections in swine. I several European countries, including Denmark, the use of antibiotic is highly restricted and must not be used to control sub clinical salmonella infection. The reason is due to the fact, that humans become infected with *Salmonella* from meat. Approximately 30% of the registered human cases in Denmark get hospitalised, and 5-10% need systemical antibiotic treatment to control the infection. When more and more Salmonella in livestock develops multi resistance it becomes more difficult for human doctors to treat critical human cases.
In several developed countries, the antibiotic of first choice to treat a critical human salmonellosis is flouroquinolones, e.g. Ciprofloxacin. If Salmonella in swine and pork are multiresistant, including resistance to flouroquinolones, due to uncontrolled use of flouroquinolones in the swine population, it will increase the numbers of non-successful antibiotic treatments in humans.

Non-antibiotic control of Salmonella infections in swine herds

Feedstuffs

Research in Denmark has demonstrated that the use of heat-treated pelleted feed is a major risk factor for a high level of Salmonella in swine herds. It is also well documented that use of meal feed (i.e. not heat-treated and non-pelleted) reduces the prevalence of *Salmonella* among finishers (Table 1).

Table 1.

Meal Feeds vs. Pelleted Feed: Salmonella Presence in Finishers				
	Pellets	Meal	Positive difference	Source
Relative risk	3.01	1	Yes	VeterinærInformation, December 1997

Relative risk	3.33	1	Yes	Report no. 426, National Committee for Pig Production, Denmark
Relative risk	4.90	1	Trend (P=0.051)	Report no. 9608, National Committee for Pig Production, Denmark
Relative risk	1.90	1	Yes	Report no. 385, National Committee for Pig Production, Denmark

The reducing effect of meal feed on *Salmonella* prevalence was also demonstrated in a study of sow units in 69 Danish herds (VeterinærInformation, December 1999).

The effect of changing from a typical compound diet (heat-treated and pelleted) to a diet where part of the grain (typically 25-30%) was not heat-treated and pelleted was examined in Salmonella-positive herds (Dahl et. al. 1999). The change of feed reduced the prevalence of *Salmonella*, significantly (Figure 1).

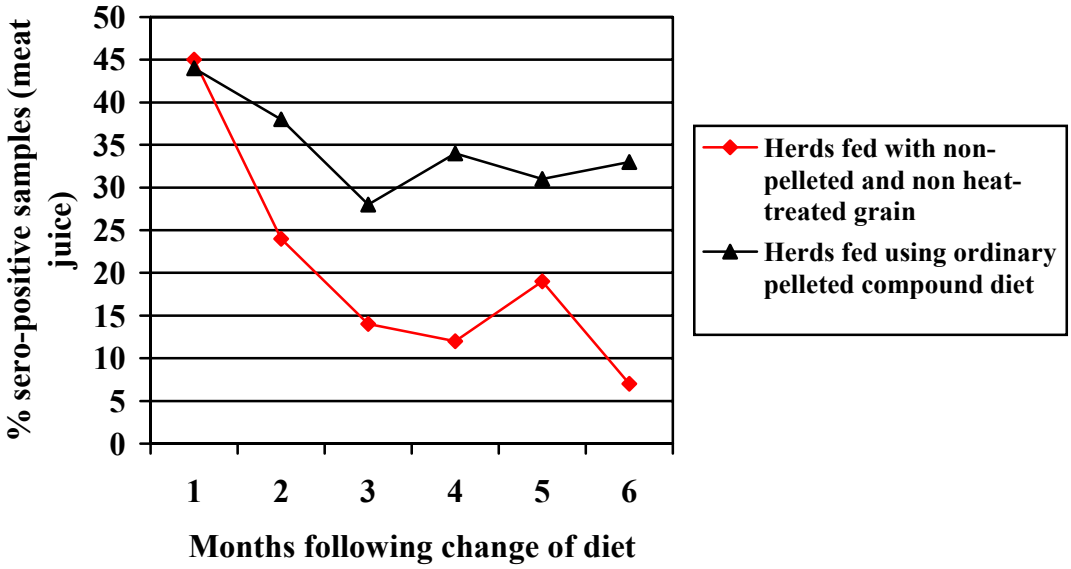


Figure 1. Effect of non-pelleted and not heat-treated feed on salmonella-seroprevalence in finishing herds, (Dahl et al. 1999).

The reason why meal feed has a more beneficial effect than pelleted feed by reducing *salmonella* is presumably because the micro flora and the concentration of organic acids in the gastro-intestinal tract are influenced favourably i.e. poor growth conditions for Salmonella (Report no. 426, National Committee for Pig Production, Denmark). Consequently, a minimum of 25% of the feed mix should be non-pelleted and non heat-treated. This applies to all categories of pigs.

Experiments with non-pelleted and non heat-treated feed have shown that the level of demixing is increased. Experience has also shown that this is difficult to handle at the feed mill and in the feed dispensers on farm. It is also expected that feed conversion will be slightly reduced (Report no. 426, National Committee for Pig Production, Denmark).

Tests with meal feed have also shown that productivity may be reduced, with lower feed conversion than pelleted feed. The tests showed a difference of up to 0.17 FUp/kg growths (Report no. 385 and 475, National Committee for Pig Production, Denmark).

Feed structure
Two tests have shown that coarse grinding can also reduce the prevalence of *Salmonella* (Table 2).

Table 2. Fine vs. Coarse Ground Feeds: Salmonella Prevalence in Finishers

	Fine	Coarse	Positive difference	Source
Relative risk	1.30	1	Trend (P=0.08)	Report no. 385, National Committee for Pig Production, Denmark
Relative risk	3.33	1	Yes	Report no. 426, National Committee for Pig Production, Denmark

Unpublished research has shown that the microbial ecosystem is improved with higher content of coarse ground barley. A high level of lactic acid bacteria, few coliform bacteria, high acid concentration, and low pH in the stomach characterizes a good microbial ecosystem, where *Salmonella* appears to experience poor growing conditions.

Results have also shown that productivity is reduced by use of coarse ground meal feed because of reduced feed conversion.

Grain, which is not heat-treated and not pelleted, should be:

- Ground on a hammer mill with 4 mm screen,
- Grated or rolled

Feed ingredients
Barley has been shown to give intestinal contents firmer consistency compared to wheat, which probably benefits the microbial ecosystem in the gastro-intestinal tract. The effect of the barley/wheat content in pelleted finisher feed on the prevalence of *Salmonella*, gastro-intestinal health and productivity was examined in research now being published. The results show that the majority of positive blood samples were among those pigs that consumed feed in which wheat provided the grain element. It is recommended that at least 25% of the feed mix consist of barley and/or oats (for piglets – at least 15%). If beet pellets are used, the proportion of barley/oats may be reduced in a proportion of 1:1.

Organic acids
Acid has a restrictive influence on *Salmonella* because of a low pH, as *Salmonella* do not grow at pH levels below 4.5. An addition of 0.4% organic acid to the diet through the entire finisher period was shown to reduce the *Salmonella* level immediately before slaughter (Wingstrand et. al. 1997). New results have shown that different acid products (based on lactic acid and/or formic acid and formic acid salt) added in pelleted feed benefit the microbial ecosystem in the gastro-intestinal tract in a similar manner as meal feed (Report no. 489, National Committee for Pig Production, Denmark).
Dutch research has shown that using $2^{0}/_{00}$ organic acids in drinking water in the finishing period reduces Salmonella prevalence (P. J. van der Wolf 2000).

When using dry feed, organic acids or acid salts are added to the feed or the drinking water for piglets and finishers. If acid is added to the feed, at least 0.5% pure organic lactic-/formic acid

or acid salt must be used. The mixture should be adjusted to control the acid concentration in individual products. If acid is added to drinking water, at least 0.2% organic lactic-/formic acid with an acid concentration of minimum 80% should be added. If the acid concentration is lower, the dosage should be adjusted accordingly.

Fermented liquid feed
Research has shown that fermented liquid feed reduces the risk of *Salmonella* (VeterinærInformation, December 1997). Tests have also shown that fermented wet feed with a pH of 4.5 has improved the microbial ecosystem in the pigs' gastro-intestinal tracts measured by prevalence of coliform bacteria compared with dry feed (Hansen et. al. 2000). PH in wet feed should be 4.7 maximum and at optimum under 4.5 pH. It should be controlled weekly in connection with the feeding regime (by the drain pipe in the pen or in the tank after recirculating).

Internal bio security
Good internal bio security is essential to eliminating salmonella problems and minimising the risk of potential problems in future.

Tests have shown that it is possible to obtain salmonella-negative pigs from herds with a high salmonella-level by removal of piglets and by avoiding any contact with salmonella-positive pigs (Dahl et. al. 1997).

The following procedures must be observed:
- All-in / all-out rooms
- Avoiding contact between different age groups (e.g. snouts, manure)
- Continuous use of sick pens and/or destruction of sick pigs
- Avoiding use of the same equipment or tools for different age groups
- Avoiding slurry floods
- Thorough cleaning and disinfection between every batch.
- Manure disposal

External infection control
Good external infection control is vital to prevent spreading of *Salmonella* to neighbouring herds and wild life. Rodents can maintain and possibly propagate salmonella infection in the herd. Humans, dogs, cats, birds, rodents, other animals, and tools can contribute to the spread of infection in the environment and to other herds.

The following procedures must be observed:
- Frequent change of clothes, footwear and hand-washing
- Wild birds and rodents should not have access to the herd
- Total separation between pigs and other animals must be maintained at all times

- Carcasses for destruction must be placed in containers or be properly covered

The Danish Surveillance and Control Program for Salmonella in Swine

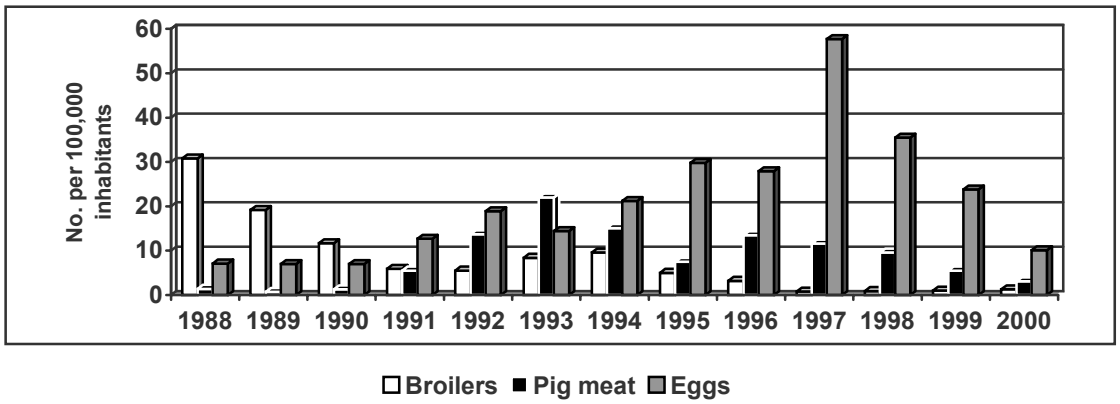
In 1993 the Ministry of Food, Agriculture and Fisheries of Denmark and the Danish Bacon and Meat Council initiated an ambitious programme to eliminate pork as an important source of human salmonellosis. In the beginning of the 1990's pork had become recognized as an increasing important source of human salmonellosis in Denmark (Wegener and Baggesen, 1996).

Estimates from the Danish Zoonosis Centre show that the total number of cases of salmonella poisoning has fallen by 50% since 1997. The number of incidents peaked in 1997 when 5,000 people were taken ill compared to around 2,500 cases in the year 2000. The main sources of salmonella poisoning are food related, both home produced and imported, as well as travel abroad.

Eggs used for fresh consumption have accounted for the majority of the cases during the past few years.

The cases associated with pork peaked in 1993 after which a series of initiatives to reduce salmonella in the pig industry were implemented. Consequently, pork has steadily declined as a source of human salmonellosis since 1996. Results from year 2000 show an incidence of only 3.1 cases per 100.000 inhabitants in Denmark equal to a total number of 160 human cases related to pork (Figure 2).

Figure 2. Sources of human salmonellosis in Denmark, 1988-2000.
Ref: Anonymous, 2001. Annual Report on Zoonoses in Denmark 2000, Ministry of Food, Agriculture and Fisheries



The Danish Salmonella Control Programme in pigs operates at all stages of the production chain and has applied nationally since 1995 (Mousing et al. 1997, Nielsen & Wegener 1997).

By August 1st 2001 a new Salmonella Surveillance and Control Program have been implemented where a number of parameters in the programme have been adjusted.
Basically, the level of salmonella is controlled at various stages:

- Feedstuffs
- Breeder- and multiplier herds
- Weaner producers
- Finisher herds with a production of more than 200 animals per year
- At the slaughterhouse, including special hygienic slaughter of highly infected herds

Feedstuffs
Compounded feedstuffs are heat treated at 81°C to eliminate *Salmonella* bacteria. The national programme requires mandatory Salmonella testing in all plants producing animal feed. The test involves microbiological analysis of compounded feedstuffs, as well as the collection of samples from critical control points during production.
The level of *Salmonella* spp. in final products is low; in year 2000 only 0,3% of the examined samples were test positive.

Breeding and multiplying herds
Each month, all herds are blood sampled and examined for Salmonella antibodies. Based on the level of antibodies a salmonella index is calculated. If the index exceeds 5, pen faecal samples must be taken and examined for the presence of *Salmonella* spp. When the index exceeds 15, as sales ban on breeding pigs is imposed until the index has declined below 15 again.

Weaner producers
If a sow herd sells weaners to a Salmonella level 2 or 3 finishing herd, pen faecal samples must be taken and examined for the presence of *Salmonella* spp.

Finishing herds
Since 1995 all finishing herds producing >100 finishers per year have been tested for Salmonella antibodies. The testing is mandatory and paid by the Danish state. Each month the herds have been divided into 3 levels with respect to the proportion of seropositive samples during the last 3 months, table 3.

- Level 1 — None or very few samples with positive antibodies. Salmonella-index<40.

- Level 2 Medium number of samples with positive antibodies. Salmonella-index40-70.
- Level 3 High number of samples with positive antibodies. Salmonella-index >70.

Table 3. Number of herds in these 3 levels in January 2001.

Level	1	2	3
No. of herds / percentage	14,961 / 96.2	445 / 2.9	139 / 0.9

The herds are also given a salmonella-index, which corresponds to the number of samples with positive antibodies taken in the last three months. The results from the latest month are weighted 3 times the level of the two preceding months. This ensures that recently infected herds implement salmonella reducing measures faster and that Level 3 herds are slaughtered separately while the salmonella load is at its maximum.

Financial penalties

The Danish Bacon and Meat has agreed on a new stricter penalty system. The purpose is to improve the salmonella control as much as possible in herds with positive samples. In practice, an intervention plan in order to reduce the prevalence of salmonella carries this out. The producer and his advisers work out the plan in detail.
The level of financial penalty corresponds to the level to which the herd is assigned:

Penalty (% of the slaughter value):

- Level 1 0%
- Level 2 2%
- Level 3 4%

Level 2 and 3 Herds

The veterinary authorities require that faecal samples are taken in order to identify the salmonella serotype. Herds assigned to level 3 have to be slaughtered under special hygienic precautions. This is done at specially designated slaughterhouses at the end of the day to prevent cross-contamination with other carcasses. Carcasses from level 3 herds also have to be heat-

treated or subject to other special treatment. Slaughterhouses may also randomly test carcasses on the basis of guidelines issued by the Danish Veterinary and Food Services.

Surveillance method for Salmonella in fresh pork

Since 1993 fresh pork has been surveyed for *Salmonella* spp. at the slaughterhouses every month. The prevalence has declined during the last two years (Table 4).
In general, 10-15 different serotypes are isolated from Danish pork. However, *S. Typhimurium* constitutes approximately 60% of the isolates (Table 5).

Table 4. The prevalence of *Salmonell*a spp. in Danish pork, 1996-2000.

Year	1996	1997	1998	1999	2000
% Positive samples	1.2	1.1	1.2	0.9	0.7

Table 5. Distribution of serotypes in Danish pork year 2000

Serotype	%
S. Typhimurium	58.8 %
S. Infantis	7.6 %
S. Derby	4.2 %
Exotic serotypes, n=7	5.9%
Rough isolates	14.3%
Non typable	5.9%

Multiresistant *Salmonella Typhimurium* DT104 has only been detected very rarely (0,002%) in Danish pork.

A new method of Salmonella testing on carcasses was introduced by January 1st 2001; 5 carcasses per slaughter day are swabbed at three defined areas (the sternum, the hind leg near the tail and the jowl) at 100 cm^2 for each sample. The swabbing areas were originally defined by USDA, USA, and are currently used in the USA as the national Salmonella monitoring method on swine carcasses. This method is more sensitive than the one used previously, and the number of positive samples recorded is expected to increase. Preliminary results for 2001 show a prevalence of 1.4-1.8%. This should be regarded as an effect of the improved test sensitivity and not increased Salmonella prevalence as such.

DT104 Herds

Herds infected with multiresistant *Salmonella Typhimurium* DT104 have to follow additional restrictions. The herd is given a Zoonotic Restriction Order. This includes a requirement for a herd intervention plan, restriction on livestock trade, and a requirement for special slurry handling. The herd intervention plan is made to ensure that salmonella reducing measures are im-

plemented in the herd for at least 12 months, and the restriction on livestock trade is to prevent the spread of DT104 infection to other herds.

Hot Water Decontamination

Finishing pigs infected with multiresistant *Salmonella Typhimurium* DT104 may either be slaughtered under special hygienic conditions as with Level 3 herds with subsequent heat-treatment or may be decontaminated with hot water. Decontamination is applied to carcasses after removal of organs. The carcass is showered with 80°C hot water for 14-16 seconds, which produces a significant reduction in the bacterial count on the surface. Five carcasses from each batch are tested to ensure that the process is effective. If *Salmonella* spp. is not detected, the whole batch may be used for fresh consumption.

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