

Salmonellas in pigs and animal feeding stuffs in England and Wales and in Denmark

BY A PUBLIC HEALTH LABORATORY SERVICE WORKING GROUP

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SUMMARY

A comparison has been made between the incidence of salmonellas in pigs and feeding stuffs in England and Wales and in Denmark. In Denmark there is veterinary legislation requiring the sterilization of imported and home produced feed ingredients of animal origin. There is no such legislation in England and Wales. In Denmark 0.3% of reesterilized imported meat and bone meal was contaminated with salmonellas. This compared with 23% of meat and bone meal in England and Wales and 20–27% of other ingredients of animal origin. In England and Wales salmonellas were isolated from 7% of caecal samples and 6% of lymph node samples, while in Denmark they were isolated from 3% of caecal samples and 4% of lymph node samples. In England and Wales 25 serotypes were found in both pigs and feeds and these included nearly all the most prevalent human pathogens. In Denmark four of the six serotypes in pigs had been found in reesterilized feed. One notable difference between the two studies was the very wide range of serotypes found in pigs in England and Wales and the narrow range in Denmark. A second was that *Salmonella typhimurium* formed 15% of all *Salmonella* strains isolated from pigs in England and Wales, and 60% of those in Denmark.

It is concluded that sterilization of animal raw ingredients in Denmark has reduced pig infections with types other than *S. typhimurium* that are found in England and Wales, but not with *S. typhimurium*. It is possible that this is because *S. typhimurium* once introduced into pigs is able to establish itself more easily than other serotypes.

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INTRODUCTION

Since 1949, food poisoning reports by medical officers of health and bacteriologists in England and Wales have been collected and the information has been published yearly (Reports, 1950–64; Vernon, 1965–70). Over the past decade the annual number of food poisoning incidents due to salmonellas has fluctuated. Incidents caused by *S. typhimurium* fell sharply between 1959 and 1962. Since then there has been little change. Incidents due to other serotypes fell between 1959 and 1960 and remained fairly constant with minor fluctuations between 1960 and 1966. The 'other serotype' infections rose sharply between 1966 and 1968 when, for the first time, their numbers were greater than those caused by *S. typhimurium*.

A comparison of the number of incidents due to *S. typhimurium* and other serotypes in 1960 and 1968 is shown in Table 1. The relative importance of 'other serotype' incidents is greater now than in the past.

During the last 10 years, a number of serotypes have shown considerable fluctuations in incidence. Between 1959 and 1964, *S. heidelberg* and *S. brandenburg* were prevalent in man. After 1964 *S. panama* and *S. stanley* rose in human incidence and in 1967 the number of infections caused by *S. stanley* was second to those due to *S. typhimurium*. *S. panama* incidents have also continued to rise until in 1968 only *S. typhimurium* caused more infections in man. The prevalence of *S. panama* continues. *S.* 4, 12: d:–, an unnamed monophasic serotype isolated for the first time in England and Wales in 1968, was one of the most frequently found serotypes in 1969. In 1970, *S. agona* appeared and has become common. Pigs appear to be one of the main reservoirs of infection of these serotypes, but they have been found in poultry as well. Common sources of non-human isolations are pigs, pork products, drains of pig farms and bacon factories and pig slaughter areas in abattoirs. They are also isolated from animal feed ingredients.

Untreated animal feed in England and Wales is frequently contaminated with salmonellas. The part played by feeding stuffs in introducing new serotypes is probably significant, but their role in maintaining animal reservoirs of important human pathogens is difficult to assess. Pork and pork products are implicated each year in outbreaks of human salmonellosis. There is also some evidence from reports of salmonella isolations from pigs that they are a major source of salmonellosis in man, and it was thought that feed ingredients might be related to the maintenance of these animal reservoirs and also to the introduction of new serotypes. A com-

Table 1. Comparisons of incidents due to *Salmonella typhimurium* and other salmonella serotypes in England and Wales in 1960 and 1968

	1960*		1968†	
	No. of incidents	% of total incidents	No. of incidents	% of total incidents
<i>S. typhimurium</i>	2907	74	1654	43
Other serotypes	1047	26	2142	57

* Report (1961). † Vernon (1970).

parison of salmonellas isolated from pigs and their feed was arranged. Incidence and range of serotypes was investigated.

During the course of the study in England and Wales the Danish State Veterinary Serum Laboratory and the Institute of Microbiology and Hygiene of the Royal Veterinary and Agricultural University were approached and they agreed to carry out a similar project. There is strict veterinary legislation in Denmark requiring reesterilization of imported feedstuffs of animal origin and sterilization of Danish animal products sent to rendering plants for destruction. A comparison between similar studies in the two countries was, therefore, considered useful in assessing the role of feeding stuffs in salmonella transmission.

MATERIALS

In England and Wales intestinal contents of pigs passing through abattoirs and raw ingredients of feeding stuffs distributed in the same area were examined. Bristol and Cardiff Public Health Laboratories had been monitoring feed ingredients for some years and it seemed reasonable to use the West Country of England and Cardiff in Wales as a suitable geographical unit for survey.

Samples of pig faeces were obtained and examined each week at six abattoirs by the Public Health Laboratories in Bath, Bristol, Cardiff, Exeter and Poole. The Bath laboratory examined pigs from two abattoirs, and the other laboratories from one each. Four of the five laboratories collected specimens by excising the caecum between two ligatures. Samples were placed in suitable containers for transport. In the remaining laboratory (Exeter) caecal contents were aspirated by a wide-bore Pasteur pipette.

Four laboratories also cultured mesenteric glands in addition to caecal faeces. In two, mesenteric glands and caecal samples were removed and examined from the same pigs.

Feeding-stuff ingredients were obtained at Avonmouth from a compounder and examined regularly at Bristol and Cardiff. These ingredients, which are listed in Table 4, include the principal ones of animal origin, that were likely to be salmonella contaminated, used in the manufacture of compounds or concentrates or purchased by farmers for mixing on farms. They include both imported and home produced material.

The only regulations pertaining to feed ingredients manufactured in the U.K. are that meat and bone meal should have certificates stating that the plant is capable of sterilizing this material.

In Denmark pig samples were obtained from two slaughterhouses. Faeces were removed from the caecum in the intestinal cleansing department into plastic beakers and portions were transferred at the abattoir to tetrathionate broth. Lymph nodes (ileo-caecal and part of mesenteric) were removed during meat inspection, cooled immediately to 0–5° C., and cultured the following day in tetrathionate broth. In some cases glands and faeces were collected from the same pig, but this was not a general procedure. These specimens were all examined at the Institute of Microbiology and Hygiene of the Royal Veterinary and Agricultural University.

Examination of feeding stuffs was carried out at the State Veterinary Serum Laboratory in Copenhagen.

Imports of meat and bone meal into Denmark were prohibited in 1933, but re-established after the war. Each shipment had to be accompanied by a document certifying that the consignment had been adequately heated (moist heat at 115–135° C. for at least 1 hr., or dry heat at 140° C. for at least three hours). Between 1949 and 1954, 1357 samples of this heat treated material were examined and 41 (3.6 %) contained salmonellas. Certification did not, therefore, exclude the occasional presence of food poisoning organisms. After March 1954 all imported meat and bone meal had to be reesterilized at a Danish rendering plant in portions of not more than 25 tons.

Fish meal is not imported into Denmark. Danish fish meal is steam heated at 100° C and is reported to be free of salmonellas.

Home produced meat and bone meal has to be moist heat treated at 125° C. for 15 min. In addition there is a rigid separation in Danish rendering plants between the raw materials and the finished product. Plants are constructed so that there is no connexion to allow cross-contamination between the 'clean' and 'unclean' sections. Staff working in the two sections are kept apart by having separate eating, washing and toilet facilities.

There are no regulations requiring treatment of vegetable feed ingredients.

There were two separate series of examinations:

(1) Imported meat and bone meal sampled between 1949 and 1954, i.e. the period before compulsory reesterilization, and from 1954 to 1970, i.e. the period after compulsory reesterilization.

(2) The second series of samples were those actually fed to the pigs whose faeces and lymph glands were being examined. In the area studied, 90 % of all pig feeding stuffs came from two compounders. Both were willing to co-operate.

METHODS

In England and Wales, little alteration was made in techniques usually employed by the Public Health Laboratory Service for salmonella isolation. Selenite F broth, or tetrathionate broth, were used as fluid enrichment media. Incubation was carried out at 37 or 43° C. (Harvey & Thomson 1953) and subcultures were made at 24 hours to deoxycholate citrate agar, brilliant green MacConkey agar, or Wilson and Blair's bismuth sulphite agar. Personal preference naturally determined the method used. British technique was, therefore, less standard than Danish.

In Denmark, 10 g. of pig faeces were transferred to 100 ml. of tetrathionate broth to which 40 mg./l. of the sodium salt of novobiocin had been added. Incubation was carried out at 37° C. and subcultures made at 24 and 48 hr. to brilliant green lactose sucrose phenol red agar plates (B.L.S.F. agar). These were incubated at 37° C. and salmonella-like colonies were picked to triple sugar iron agar slants. Lymph node samples, immersed in boiling water for a few seconds, were cut into pieces by a scalpel and inoculated into tetrathionate broth without novobiocin.

Culture followed as described for faeces. No difficulties were encountered with swarming of colonies. In a few instances 0.02 % of Teepol was added to B.L.S.F. plates to prevent swarming. No improvement in results was obtained.

From each sample of feeding stuff, 10 portions of about 2 g. were cultured individually in approximately 30 ml. of ordinary nutrient broth. After 48 hr. incubation at 37° C., 2 ml. of each broth culture were transferred to about 30 ml. of tetrathionate. The enrichment media were incubated for 24 hr. at 37° C. and subcultured to brilliant green agar plates. After incubation at 37° C. salmonella-like colonies were picked for further identification and serotyping.

RESULTS

England and Wales

Details of samples and isolations were entered on two forms and sent to the Epidemiological Research Laboratory in London, where they were recorded and tabulated. A total of over 8000 pig samples and 1500 feeding stuff samples were examined, the majority in the period November 1968 to January 1970. Sampling has continued in some laboratories after this date.

Table 2 shows results of caecal sampling. Salmonellas were isolated from 7 % of 5637 pigs examined. In Bath, Cardiff, Exeter and Poole, where many specimens were cultured, the incidence varied from 2 to 14%. In Bath, the laboratory recording the highest incidence, there was some degree of bias. More than a third of the samples came from a single farm on which salmonellas were found in feeding stuffs, pen samples and drain swabs.

Table 2 also records the isolations from mesenteric glands. The total incidence was similar to that found in faeces (6 % of 2483) and varied, in different areas, from 2 to 9 %. Two laboratories examined caecal contents and mesenteric glands from the same pigs during part of the investigation (Table 3). Isolation rates from the two sites were similar, but it was unusual for both to be found positive in the same animal. The proportion of pigs containing salmonellas approximately doubled when figures from glands and faeces were combined.

Table 4 provides information on feeding stuff ingredients examined and salmonella isolation rates. Salmonellas were isolated from 20–27 % of feed in-

Table 2. *Salmonella isolations from caecal samples and mesenteric glands of pigs, by five laboratories in England and Wales*

Laboratory	Number of caecal samples		Number of gland samples	
	Tested	Positive	Tested	Positive
Bath	628	88 (14)	—	—
Bristol	67	0	722	34 (5)
Cardiff	1281	115 (9)	281	13 (5)
Exeter	1940	45 (2)	610	14 (2)
Poole	1721	123 (7)	870	78 (9)
Totals	5637	371 (7)	2483	139 (6)

(The figures in parentheses indicate percentages.)

redients of animal origin, with the exception of herring meal of which only 5% of samples were contaminated. They were isolated from 1% of sow nuts – a processed vegetable ingredient. It is noteworthy that 20% of 264 samples of fish pellets contained salmonellas.

The range of serotypes found in the investigation was wide. Thirty-nine serotypes were found from 315 salmonella isolations from 5637 caecal samples, 23 serotypes from 104 salmonella isolations from 2483 gland samples and 52 serotypes from 313 isolations from 1772 samples of feed. Twenty-five serotypes were common to pigs and feed and these included several types prevalent in man. Table 5 lists serotypes found five times or more in each source, in order of frequency. In caecal samples, *S.* 4, 12: d:–, *S. typhimurium* and *S. dublin* were common, while *S. anatum* and *S. senftenberg* were often isolated from feed ingredients. Many other types were infrequently found. *S. dublin* was the commonest serotype found in mesenteric glands where it was found more frequently than in caecal faeces. In contrast, *S.* 4, 12: d:– had the maximum incidence in faeces and was relatively less common in glands.

Tables 6 and 7 contrast the incidence of serotypes common in pigs with those common in feeding stuffs. The tables are self-explanatory. Quantitative similarity of serotype incidence is not evident.

Denmark

Table 8 shows results of the Danish pig study, in which caecal samples and lymph node specimens were examined between May 1969 and July 1970 on 17 sampling days. Salmonellas were isolated from 8 (3%) of caecal faeces and 15 (4%)

Table 3. *Growth of Salmonella from caecal and gland samples taken from the same pigs, England and Wales*

Laboratory	Total pigs tested	Pigs positive in			Total positive
		Caecum and glands	Caecum only	Glands only	
Cardiff	276	3 (1)	13 (5)	10 (4)	26 (9)
Exeter	610	3 (0.5)	11 (2)	11 (2)	25 (4)

(Figures in parentheses are percentages.)

Table 4. *Growth of Salmonella from different ingredients of pig feed in England and Wales*

Raw material	No. samples	No. positive	Positive (%)
Feather meal	99	27	27
Raw materials [unspecified]	138	36	26
Meat and bone meal	704	163	23
Fish meal	31	7	23
Fish pellets	264	53	20
Herring meal	60	3	5
Sow nuts	162	1	1

of lymph nodes. *S. typhimurium* was the serotype most frequently found in lymph node samples.

In the period 1949–54, examinations of consignments of imported animal feed ingredients had shown that the accompanying heat treatment certifications were not proof of absence of salmonellas. Compulsory reesterilization began in March 1954 and 10,782 samples representing about 270,000 tons of imported meat and bone meal were examined between 1954 and 1970. All this material had been reheated in Denmark after importation. Thirty-four samples (0.3%) contained

Table 5. *Salmonella serotypes isolated from different sources, in England and Wales*

Caecum (5637 samples)		Gland (2483 samples)		Feeding stuff (1772 samples)	
Serotype	No. of isolations	Serotype	No. of isolations	Serotype	No. of isolations
<i>S. 4, 12: d:-*</i>	55	<i>S. dublin*</i>	37	<i>S. anatum*</i>	64
<i>S. typhimurium*</i>	51	<i>S. typhimurium*</i>	24	<i>S. senftenberg*</i>	58
<i>S. dublin*</i>	50	<i>S. eimsbuettel*</i>	11	<i>S. livingstone*</i>	29
<i>S. chester</i>	40	<i>S. 4, 12: d:-*</i>	11	<i>S. montevideo*</i>	20
<i>S. panama*</i>	23	<i>S. livingstone*</i>	9	<i>S. oranienburg</i>	20
<i>S. fischerkietz*</i>	23	<i>S. heidelberg*</i>	7	<i>S. reading*</i>	17
<i>S. livingstone*</i>	20	<i>S. rostock</i>	6	<i>S. typhimurium*</i>	15
<i>S. stanley*</i>	12	<i>S. montevideo*</i>	5	<i>S. tennessee*</i>	15
<i>S. bredeney*</i>	10			<i>S. eimsbuettel*</i>	11
<i>S. indiana*</i>	9			<i>S. infantis*</i>	10
<i>S. agama</i>	9			<i>S. 4, 12: d:-*</i>	10
<i>S. heidelberg*</i>	7			<i>S. cubana*</i>	10
<i>S. anatum*</i>	6			<i>S. ruiru</i>	8
<i>S. senftenberg*</i>	5			<i>S. bredeney*</i>	7
<i>S. nagoya</i>	5			<i>S. dublin*</i>	7
<i>S. eimsbuettel*</i>	5			<i>S. thompson*</i>	6
<i>S. montevideo*</i>	5				

* A serotype common to pig and feeding-stuff samples. Only serotypes that have been isolated five or more times are included in this table.

Table 6. *Isolation rates from all sources of some Salmonella serotypes which are common in pig samples (England and Wales)*

Serotype	Number of times specified <i>Salmonella</i> serotypes were grown from		
	Caecum	Glands	Feeding stuffs
All serotypes	371	139	367
<i>S. 4, 12: d:-</i>	55 (15)	11 (8)	10 (3)
<i>S. typhimurium</i>	51 (14)	24 (17)	18 (5)
<i>S. dublin</i>	50 (14)	37 (27)	7 (2)
<i>S. chester</i>	40 (11)	0	0
<i>S. fischerkietz</i>	23 (6)	2 (1)	2 (1)
<i>S. eimsbuettel</i>	5 (1)	11 (8)	10 (3)
<i>S. livingstone</i>	20 (5)	9 (6)	29 (8)

(Figures in parentheses are percentages.)

Table 7. *Isolation rates from caecal samples and glands of some Salmonella serotypes which are common in feeding stuffs (England and Wales)*

Serotype	Number of times specified <i>Salmonella</i> serotypes were grown from		
	Feeding stuffs	Caecum	Glands
All serotypes	367	371	139
<i>S. senftenberg</i>	60 (16)	5 (1)	3 (2)
<i>S. anatum</i>	64 (17)	6 (2)	4 (3)
<i>S. livingstone</i>	29 (8)	20 (5)	8 (6)
<i>S. montevideo</i>	22 (6)	5 (1)	5 (4)
<i>S. oranienburg</i>	20 (5)	0	0
<i>S. reading</i>	17 (5)	1 (1)	1 (1)

(Figures in parentheses are percentages.)

Table 8. *Salmonella isolations from pig samples taken from two abattoirs in Denmark*

Isolations from samples of			
Caecal faeces		Lymph nodes	
Total samples	296	Total samples	359
Total positive	8 (3%)	Total positive	15 (4.2%)
<i>S. typhimurium</i>	2	<i>S. typhimurium</i>	12
<i>S. montevideo</i>	3	<i>S. montevideo</i>	1
<i>S. senftenberg</i>	1	<i>S. dublin</i>	1
<i>S. oranienburg</i>	1	<i>S. bispebjerg</i>	1
<i>S. indiana</i>	1		

Table 9. *Salmonella isolations from imported meat and bone meal in Denmark*

	No. of samples	No. positive
Before compulsory reesterilization 1949-54	1,337	41 (3.6%)
After compulsory reesterilization 1954-70		
Post-sterilization samples	10,782	34 (0.3%)
Pre-sterilization samples	278	47 (16.9%)

Table 10. *Salmonella serotypes isolated more than five times from imported meat and bone meal in Denmark before sterilization, 1949-70*

	Period			Period	
	A	B		A	B
<i>S. cubana</i>	4	10	<i>S. give</i>	2	6
<i>S. anatum</i>	3	9	<i>S. bredeney</i>	3	4
<i>S. typhimurium</i>	6	6	<i>S. montevideo</i>	1	6
<i>S. oranienburg</i>	2	8	<i>S. senftenberg</i>	0	7
<i>S. minnesota</i>	2	7	<i>S. kentucky</i>	0	6

Period A, 1949-54 (Feb.).

Period B, 1954 (Mar.)-1970.

The figures show the number of times each serotype was isolated in each period.

salmonellas. In the same period 278 such specimens were examined before reesterilization. Forty-seven (17%) were positive for salmonellas (Table 9).

Table 10 lists serotypes in frequency order isolated from imported meat and bone meal between 1949 and 1970. This table refers only to samples which had not undergone compulsory reesterilization, while Table 11 shows serotypes isolated from reesterilized imported feed ingredients sampled after 1954.

Table 12 records salmonella isolations from feed samples supplied by the two compounders involved in the Danish study. Two out of 206 samples were positive. One isolation was made from a pelleted mixture, the other from a meal. *S. typhimurium* was the serotype found. It was present in only one of 10 portions of a single sample (see section on Methods).

Comparison of results in England and Wales and in Denmark

From Tables 2, 5 and 8 the results of pig examinations in the two countries can be compared. The percentage isolation rate from caecal samples was 7% in England and 3% in Denmark, that from mesenteric gland samples 6% in England and 4% in Denmark. Forty-three serotypes were isolated in England and seven in Denmark. *S. typhimurium* accounted for 60% of all isolations in Denmark, but only 15% in England.

From Tables 4 and 9 a comparison may be made between the results of examination of imported meat and bone meal in Denmark and those of meat and bone meal examined in two British laboratories, Bristol and Cardiff. In England and Wales the present rate of contamination of 23% compares well with that of Denmark (16.9%) in the period after 1954 before reesterilization was actually carried out. In England and Wales 40 different serotypes were isolated, compared with 32 in Denmark from all unsterilized material from 1949 to 1970.

Table 11. *Salmonella serotypes isolated from reesterilized imported meat and bone meal in Denmark, after 1954*

Serotype	No. of times isolated	Serotype	No. of times isolated
<i>S. senftenberg</i>	7	<i>S. enteritidis</i>	1
<i>S. montevideo</i>	6	<i>S. meleagridis</i>	1
<i>S. typhimurium</i>	5	<i>S. minnesota</i>	1
<i>S. cubana</i>	5	<i>S. oranienburg</i>	1
<i>S. alachua</i>	3	<i>S. pullorum</i>	1
<i>S. enteritidis-danysz</i>	2	<i>S. tennessee</i>	1
<i>S. anatum</i>	1		

Table 12. *Salmonella isolations from feed from compounders supplying two pig abattoirs in Denmark*

Sample	No. of samples	No. positive	Serotype isolated
Feed mixture (pelleted)	88	1 (1%)	<i>S. typhimurium</i>
Feed mixture (meal)	96	1 (1%)	<i>S. typhimurium</i>
Meat and bone meal	11	0	—
Fish meal	11	0	—
Total	206	2 (1%)	

DISCUSSION

In England and Wales 7 % of pig faeces and 6 % of mesenteric glands were positive for salmonellas. Percentages can be misleading unless it is realized that technique has a bearing on results. In Cardiff, in one series studied, inocula of 80 g. of faeces were compared with 0.5 g. Both inocula came from the same pig. The larger inoculum gave an incidence of 10 % of salmonellas, the smaller 5 %. The inoculum of glands used might also have to be taken into account in interpreting percentages. Obviously the figures for the group as a whole are merely averages, and technical variations and their possible influence cannot be evaluated.

When glands and faeces were examined from the same pig, it was rare for both sites to be found positive in the same animal. If isolations from both samples were combined the number of pigs harbouring salmonellas approximately doubled. The animals were symptomless as far as is known.

A recent study of salmonellosis in pigs in Holland identified a higher proportion of positive animals (20–50 %) than was found in the British investigation. Multiple faecal examinations were made during life and after slaughter in the Dutch series and this may well account for the difference (Kampelmacher, Guinée & Keulen, 1965).

In England previous pig investigations were carried out by Scott (1940) who examined mesenteric lymph nodes and spleens of slaughtered pigs at two bacon factories. Thirty-eight isolations were made from 1000 pigs (3.8 %) and eight serotypes were found. In a similar investigation in 1944–5 (Report 1947), 133 (2.5 %) salmonellas were isolated from 5285 pigs, which included 17 different serotypes. From the faeces of 600 healthy pigs, Smith & Buxton (1951) isolated four salmonellas (0.66 % positive) and only two serotypes. A working party of the Public Health Laboratory Service (Report, 1955) examined 5166 samples of lymph nodes and other tissues and an isolation rate of only 0.4 % was obtained with only three serotypes. Smith (1959) isolated 60 salmonellas from 500 pigs (12 %) which included 16 serotypes.

All these surveys have varied in size. They have been carried out in areas differing in size and locality and different sampling methods have been used. In the present survey the isolation rate in the different laboratories varied from 2 to 13 %. The isolation rate from one farm in which the feed was known to be contaminated was 18 %. It is thus clear that there are local factors operating and these may well vary from time to time so that it is difficult to make strict comparisons. However, it can be said that the present study has led to the isolation of many more serotypes in pigs (43) than have previously been isolated (17).

In 1964 Bevan Jones *et al.* showed that there was a relationship between the salmonella isolation rate in mesenteric glands and the length of stay of the pigs in the lairages. A 5–10 % isolation rate of two serotypes was obtained from pigs with a short stay in lairages (less than a week) and up to 90 % in pigs with a long stay (a week to a month). Slaughterhouse regulations of 1966 have forbidden the keeping of animals for longer than 72 hr. The pattern of pig salmonellas found in England and Wales in the present investigation suggested the source of infection. The same salmonella serotype was not usually isolated in succession during the

investigation, unless a number of pigs from the same farm were involved. This indicated that pigs were probably arriving at abattoirs already infected from farm or market and were not contracting salmonellas in the abattoir. Had the reverse been the case, animals from different sources would more likely have been infected with the same serotype on consecutive sampling days. Only one episode of this type occurred, when *S. chester* was isolated on 37 occasions in a single week.

Twenty-five serotypes in British pigs were also found in feed ingredients distributed in the same area. All types of ingredients sampled are fed to pigs. Meat and bone meal was the material most frequently sampled and 23 % of specimens contained salmonellas.

In 1968, over 90 % of meat and bone meal fed to United Kingdom livestock was home produced and nearly all serotypes common to pigs and feed were present in this material. This indicated the existence of a potential cycle of salmonella infection through abattoir and butcher waste products back to animals.

Approximately 75 % of fish meal and herring meal processed by large compounders, such as the firm involved in this study, is imported. Twenty per cent of the imported fish pellets examined from many batches were contaminated. Pelleting is known to minimize salmonella contamination if sufficient heat is employed. The pelleting in this instance was carried out at sea and may be a 'cold' process, i.e. heat below sterilization temperatures is used. Contamination of one batch of South African fish pellets may have occurred after manufacture. Salmonellas could have been introduced in transit to the United Kingdom in ships cleaned with sewage-polluted water. Alternatively, contamination could have occurred in Britain by contact with other contaminated material containing salmonellas, or from contaminated dust from plant environment. This is known to occur in other similar trades.

Approximately half the feather meal used by compounders is home produced and the other half imported.

Sow nuts contain vegetable ingredients (barley, wheat), vitamins and essential trace elements that are fed to pigs.

Of the serotypes isolated from feeding stuffs, *S. typhimurium* was found 15 times and is sixth in order of frequency. It is often suggested that, as this serotype is seldom present in animal feed, transmission by this route is unlikely. Few would deny, however, that *S. typhimurium* is frequently found in animal tissues (Table 5) and that these are sent to rendering plants for conversion into animal feed stuffs. Isolation of *S. typhimurium* from feed ingredients may be difficult. For example, isolation of *S. typhimurium* from bone meal is very much dependent on the technique used (Harvey & Price, 1970).

The 25 salmonellas common to pigs and feed in Britain included nine of the ten most frequently found human pathogens encountered in 1969. The nine serotypes were – *S. typhimurium*, *S. enteritidis*, *S. panama*, *S. heidelberg*, *S. stanley*, *S. 4, 12:d:-*, *S. indiana*, *S. infantis* and *S. dublin*. The first and last serotypes are more commonly isolated from cattle than from other animal sources. *S. virchow* was frequently found in man in 1969, but this serotype is now associated with

poultry and our failure to find it in pigs is not surprising. Most of the other serotypes have been commonly found in poultry as well.

The incidence of salmonella types found both in feed and in pigs shows little quantitative correspondence (Tables 6 and 7). We do not think that this invalidates the potential association. Certain serotypes may survive and multiply differently in different environments and those which readily multiply in the pig intestine may not do so in pig and other animal remains from which animal raw ingredients for feeds are derived. Enterobacteriaceae show differences in this respect and, in examination of bones from India and Pakistan, salmonellas have frequently been isolated from samples *not* containing *E. coli*. This has also been experienced with other dried food materials (Harvey and Price; unpublished).

In the Danish investigation, salmonellas were found in pigs' faeces and mesenteric glands despite sterilization of ingredients of animal origin, but the incidence was less than that in England and Wales – 3 % in faeces and 4 % in glands.

The most striking difference between Denmark and Britain is the narrow range of pig serotypes found in the former country and the wide range in the latter. *S. typhimurium* isolations were 60 % of all isolations in Denmark and only 15 in England. This may be of great significance. During the period 1960–8 *S. typhimurium* accounted for 80 % of all isolations in humans in Denmark. During the same period in England *S. typhimurium* accounted for 60 % of human incidents in man, but this has been getting progressively less and in 1968 *S. typhimurium* accounted for only 43 % of incidents while in the same year it accounted for 64 % of isolations in Denmark.

It is certainly evident that exotic species are not frequently found in pigs or humans in Denmark and this must be due to the heat processing of animal feed. It is likely that the cycle of *S. typhimurium*, the commonest type in pigs and humans in Denmark and other countries, is independent of feed transmission. However, it is still isolated from feeds so that it is also possible that an occasional introduction of *S. typhimurium* in the feed is sufficient to set up a persistent infection in pigs. Intensification of pig farming and animal husbandry in Denmark is similar to that in England except that with the colder climate in Denmark more pen houses are enclosed with greater control of temperature, humidity, lighting and ventilation. It is possible that in these conditions *S. typhimurium* once introduced is able to survive and multiply and so give rise to persistent cross infection.

The salmonella incidence in treated Danish meat and bone meal was 0.3 % as compared with 23 % in the United Kingdom. The finding of salmonellas in treated Danish feed suggests either insufficient heat treatment, or contamination of the treated product by contact with contaminated material, notably vegetable feed materials, which are not sterilized. In Britain it is known that feed pelleting is a rather critical process and it is possible for some failure to occur. Our experience also leads us to suppose that cross contamination from other contaminated materials is a potential danger.

As regards the role of imported feeds in Denmark, Müller (1952, 1957) showed that 'foreign' salmonella species had increased in incidence in animals and were

also found in man. These exotic serotypes were at that time associated with imported meat and bone meal. Since introduction of compulsory feed sterilization, the situation has changed and unusual species are seldom found in animals, but this does not mean that feed is no longer of any importance in Denmark. Vegetable feed ingredients are not sterilized and are known to be occasionally contaminated. In 1958 salmonellas were found in seven of 72 vegetable feed samples examined. It is also noteworthy that of the 7 serotypes that occurred in the Danish pigs, four have been found in reesterilized imported meat and bone meal and three of these – *S. typhimurium*, *S. montevideo* and *S. senftenberg* – are the commonest from this source. *S. typhimurium*, the commonest type in the pigs, was found in two feed samples from the compounders supplying the feed to the pigs. The serotypes found in the pigs are also reported to be the types that have been found in dry rendering plants and feed-stuff factories in the warm, humid summer months.

Calculations made from the incidence of salmonellosis in Danish pigs suggested that five to ten salmonella carriers might pass through the abattoir in an hour. From swabs taken in the slaughter area no evidence of salmonella contamination of the slaughter line was obtained. Numerous examinations of meat products have also been made by laboratories authorized to perform bacterial meat inspection all with negative results. It is felt that improvements in abattoir hygiene that have taken place in Denmark in recent years prevent carcass contamination in spite of the demonstrated carriage of salmonellas by pigs.

In England and Wales, on the other hand, pork sausages and domestic pork products have been found in 1970 to contain *S. typhimurium*, *S. panama*, *S. stanley*, *S. indiana*, *S. 4, 12:d:-* and *S. enteritidis*. There is little doubt, therefore, that in this country pathogens find their way onto surfaces of carcasses and into the meat prepared from infected pigs.

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