

Sources of salmonellas in market swine*

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INTRODUCTION

Pigs at slaughter and pig products are frequently infected with *Salmonella* and are therefore a possible source of salmonella infection in man (Felsenfeld, Young & Yoshimura, 1950; Galton, Lowery & Hardy, 1954; Wilson *et al.* 1961; Williams, 1965). If this contamination of human food is to be controlled the sources of pig infections need to be ascertained and described, as terminal treatment of these products prior to retail sale is impracticable at this time.

Several investigators have reported an increase in the proportions of pigs found to be excreting salmonellas at the farm and in the holding pens before slaughter, and in the infection rate after slaughter. Galton *et al.* (1954) recovered salmonellas from 7.2, 15 and 51% of pigs sampled in these three locations. American Meat Institute researchers reported 2.7% of pigs on the farm, 94% of faecal specimens in the abattoir pens, and 43% of pigs at slaughter to have salmonellas (Leistner *et al.* 1961). McDonagh & Smith (1958) reported 2.9% of pigs tested excreting salmonellas in holding pens, and 13% infected after slaughter. These investigations were done in different areas, by different investigators using different methods. They were of cross-sectional design, comparing unlike pig populations, often at different times and places.

While it seems probable that most farm infections are the result of salmonella contamination of feed, and that new infections in the marketing process and in the holding pens are due to indirect spread (from pig via the environment to pig) or possibly direct spread from pig to pig, this has not been clearly demonstrated. At the time our investigations were begun we were aware of only one longitudinal study in the literature. Shotts, Martin & Galton (1962) demonstrated a build-up of swine infections by comparing salmonella excretion of cull sows at a sale barn, in abattoir pens after transport, and after slaughter as demonstrated by rectal swabs. They did not state whether the trucks contained salmonellas that could account for this build-up. Further, they did not report the serotypes recovered from the holding environment or the pigs after slaughter in such a way that the data could be compared, and the importance of the holding pen assessed.

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Two investigations had previously shown that salmonella serotypes in feed could infect swine and be excreted by them. Newell *et al.* (1959) traced infected pigs at slaughter to their farm of origin and found many of the same serotypes in feed ingredients being used on the farms and in pigs being fattened on the farm. In 1960, Smith fed salmonella-free piglets on a known contaminated feed and recovered some of the feed source serotypes from their faeces during life and from their lymph nodes after slaughter. When salmonella-free rations were fed to the remaining animals their excretion stopped.

In this article we describe a longitudinal study conducted on one farm and in one slaughterhouse in Louisiana, where, by changing some of the variables and by the use of serotyping and phage-typing methods, we have attempted to assess the importance of different salmonella sources.

The purpose of our research was twofold. The first was to examine the effect of using pig feed with a decreased level of salmonella contamination in a farm operation. The second was to determine the risk of salmonella infection for each lot of pigs from their environment after leaving the farm and before slaughter. To accomplish these aims six feeding lots of comparable pigs were examined before and after transport, and after slaughter. Samples were also taken from the feed, truck, and abattoir environment before the pigs' exposure to them.

FACILITIES

The pigs used in this experiment were made available by a group in Louisiana growing sugar cane, and raising pigs and feeder calves. Pigs were born and raised to market weight on this well-managed farm. They spent their entire life on concrete. Pens were covered and well drained. They were cleaned daily. No rodent signs were observed in the feeding or storage areas.

Pigs were transported to slaughter in the farm's double-decked open truck with the exception of one lot of eighty that was sent in an open single-decked truck that had previously been used for cattle and not cleaned.

Before the introduction of special feed (low-level salmonella contamination) to the farm all pig feeding, feed mixing, and feed transporting equipment was cleaned with live steam and disinfected with O-syl (Lehn and Fink Products Corp., Bloomfield, New Jersey) at a 1½% strength.

The cooperating slaughterhouse was under city inspection only. Sanitation was good in the covered holding pen area, on the kill floor, and in the coolers. When caecal swabs were collected at the time of evisceration, the individual intestines were examined grossly for signs of enteritis. None was observed. Furthermore, no carcasses or carcass parts from study pigs were condemned during the time of these investigations. Carcass sides were washed with cold water, rapidly flowing but not under pressure, and placed directly in the chill room.

FEEDING MATERIALS

The pigs were first fed, after weaning, on a commercial pelleted ration which included meat and bone meal, fish meal, condensed fish solubles, and animal fat.

It was medicated with 50 g. of oxytetracycline per ton. This feed was never sampled.

When pigs reached 50 lb. they were fed on cracked corn and the regular finishing supplement meal made by the same company producing the weaner ration. This contained the same animal origin ingredients as the weaner ration with the exception of animal fat. It, too, was medicated with 50 g. of oxytetracycline per ton.

The special feed used for the study was specially prepared with no animal origin ingredients in it. It was medicated with 80 g. of oxytetracycline per ton. Before the manufacture of each lot of special feed the mill operator 'cleaned' the system by running two tons of cracked corn through it. No attempt was made to assess the usefulness of this measure in the reduction of the contamination level of the special feeds.

SAMPLING METHODS

Pigs and environment

A rectal swab was taken from the pigs in each lot while they were still on the farm, before loading and transport to slaughter. The six lots of pigs were designated as lot I, IIA, IIB, IIC, IIIA, and IIIB to correspond with the last lot of feed they received just before slaughter. Following the 4 hr. of transport the pigs were unloaded into abattoir pens. The first three lots (I, IIA, IIB) were allowed a 1½ hr. period to settle down before rectal swabs were collected from them. The last three lots (IIC, IIIA, IIIB) were swabbed immediately on arrival. In addition to the swabs taken on arrival, lots I, IIB, IIIA, and IIIB had rectal swabs taken after an overnight holding period to determine a salmonella build-up. This was just before slaughter.

The truck was sampled before and after transport for lots I, IIB and IIIA, but only after transport for lots IIC and IIIB. The truck used for lot IIA pigs was not sampled, either before or after. Pens and their watering troughs were sampled before the entry of each lot of pigs by rubbing floors, gates, fences, and the inside of the trough with sterile swabs.

After slaughter the caecum was incised with a sterile knife and a swab was introduced in such a way that it did not touch the outside of the caecum.

Carcasses were sampled in the chill room by rubbing the swab over the outside and inside of the carcass (both halves) covering the maximum area possible.

All samplings of pigs, environment, and carcasses were done with sterile cotton swabs. These were immediately introduced into fresh tetrathionate for transport and enrichment.

Feed

Feed ingredient samples were collected through the weighing scale inspection door at the time of feed manufacture. They were put in sterile plastic bags until cultured. They were transported and held at ambient temperature if not examined immediately.

Table 1. Number of isolations from study feeds by type and source

Lot no. ...	Feed 1 (regular)		Feed 2A (special)	Feed 2B (special)	Feed 2C (special)	Feed 3A (regular)		Feed 3B (regular)		
	M	S				M	S	M	S	M
No. of subsamples ...		29	16	15	21		25		15	
Percentage positive ...		48	6	13	10		36		40	
Source of salmonella type ...			S	S	S	M	S	M	S*	F*
<i>anatum</i>	1	2	.	.	.	1
<i>cubana</i>	1	1	2	.	.	.
<i>derby</i>	1	1	.	.
<i>illinois</i>	2
<i>infantis</i>	1
<i>kentucky</i>
<i>livingstone</i>	2	.	.	1	1
<i>manhattan</i>	1
<i>mississippi</i>
<i>newington</i>	.	.	.	1	.	.	.	1	.	.
<i>newport</i>	2
<i>oranienburg</i>
<i>poona</i>	1
<i>senftenberg</i>	2
<i>simsbury</i>	2
<i>tennessee</i>	.	.	1	.	.	.	4	.	.	.
<i>typhimurium copenhagen 3A</i>	1
Totals	12	2	2	2	3	3	6	3	6	3

M, meat meal; S, soybean meal; F, fish meal.
 * Not sampled—same bin in use as in 3A.

LABORATORY METHODS

Cultural methods were similar to those of Galton (1962). Faecal and environmental samples were placed in tetrathionate broth with 0.001% brilliant green added, and incubated at 37.5° C. for 18–24 hr. A loopful of broth was streaked onto brilliant green agar (BGS) (Difco) with sulphadiazine added. Feed subsamples of 15 or 30 g. were placed in 50–100 ml. of tetrathionate (Difco) with 3–6 ml. of 1/1000 tergitol added and incubated at 37.5° C. for 72 hr. BGS plates were streaked at 24 and 72 hr. All salmonella strains were screened with a Salmonella H. Antisera Spicer-Edwards Set (Difco) and representative cultures were serotyped in the laboratory of the Louisiana State Board of Health.

All *Salmonella typhimurium* strains were typed by bacteriophage by one of us (Williams) at the Communicable Disease Centre, Atlanta, Georgia in 1963. An incomplete battery of thirteen phages was available and while it was not possible to fit all of the strains to the nomenclature used in that laboratory many consistent patterns were found in each lot of pigs and the strains from their environment. These were useful in demonstrating relationships between strains.

RESULTS

Feed

Three different types of supplement were used. These were designated 1, 2 and 3, and consisted of one, three and two batches respectively. Each batch was manufactured at a different time and differed in the variety of salmonella serotypes recovered and probably the number of salmonella organisms present. The distribution of serotype is shown in Table 1. Feed 1 and 3 were from the same source.

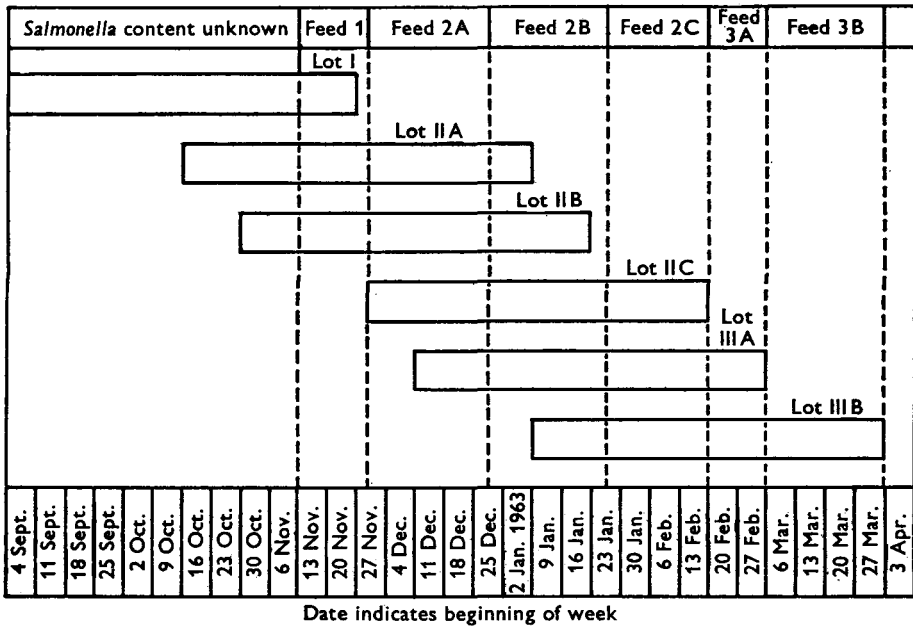


Fig. 1. Feeds consumed by pigs sampled on farm G (11–12-week finishing period). Each lot of pigs described by bars. Changes in feed are indicated by dotted lines.

Forty-one per cent of the subsamples of this regular feed were positive for a salmonella. The same figure for the special feed was 10%. The period of use of these various feeds is shown in Fig. 1 (Williams & Newell, 1967).

Environment and pig samples

A total of 276 rectal swabs were taken from these six lots of pigs while they were still on the farm. In no case was a salmonella isolated from these samples. The additional results of the follow-through sampling of each lot of pigs is shown in Table 2. The greatest number of salmonellas were recovered from lot I pigs and their associated environment after they left the farm. The next highest group was lot IIB pigs. The least number of recoveries came from lot IIIB pigs.

Table 2. *Salmonella* recoveries from swine and the environment they were exposed to in marketing

Source of recovered salmonellas	Pig lot or experiment no.					
	I	IIA	IIB	IIC	IIIA	IIIB
Environmental samples						
Truck before loading	1/10*	.	0/10	.	0/12	.
Truck after transport	3/10	.	1/10	0/10	0/6	3/10
Abattoir pen swabs	12/15	3/8	5/15	0/12	0/10	0/10
Abattoir watering trough swabs	5/5	0/3	2/5	0/5	0/6	0/4
Live pig samples						
Farm rectal swabs	0/50	0/40	0/48	0/40	0/50	0/48
Arrival rectal swabs	26/50	0/40	19/48	0/40	4/50	0/48
Holding rectal swabs†	3/50	—	2/48	—	0/49	0/48
Samples after slaughter						
Caecal swabs	26/49	9/50	17/48	10/50	6/50	1/48
Carcass swabs	4/39	.	8/40	3/40	.	.

* Number of positives over number of samples.

† After 12–19 hr. in abattoir pens.

The results of the sampling of four lots of pigs after the overnight holding period were not as expected. When the pigs were positive on arrival there was a decrease in recovered salmonellas the following morning. This change in excretion has been discussed in a previous paper (Williams & Newell, 1967).

Complex relationships between the salmonella types from the environment and feed and those subsequently recovered in pigs occurred only in lots I and IIB. The abattoir pen and trough water were both contaminated before the entrance of the pigs and these pigs had a 1½ hr. settling-down period before rectal swabbing. This period was apparently long enough to permit passage and excretion of environment types. The results by serotype recovered from lot I pigs or their environment are shown in Table 3. Similar results from IIB pigs have been summarized in Table 4. Lot IIA pigs were the only additional lot exposed to a known contaminated abattoir environment. The abattoir pen contained *S. typhimurium* of phage types 1a and 2a. One isolation of phage type 1a was made from the caecal swabs collected from this lot of pigs.

The results of the prospective sampling of feed and environment were used to assign pig or pig product isolations to one of three sources, food, environment (abattoir), and unknown. Two problems arose in making these determinations which may have affected the accuracy of assignment. The first was the number of

Table 3. *Salmonella* serotypes associated with lot I pigs after leaving the farm

Serotype	Truck before	Truck after	Pen water	Pen	Arrival swab	Caecal swab	Carcass swab
<i>anatum</i> *	.	.	×	×	×	×	×
<i>archevaleta</i>	.	.	×
<i>blockley</i>	.	.	×
<i>bredeney</i>	×
<i>cerro</i>	×	×	.
<i>cubana</i> *
<i>derby</i> *	×	.
<i>livingstone</i> *	×	×	.	.	×	×	×
<i>montevideo</i>	×	.
<i>oranienburg</i> *	×	.	.
<i>rubislaw</i>	×	.
<i>san diego</i>	.	.	×
<i>saint paul</i>	×	.
<i>senftenberg</i> *	.	×
<i>typhimurium</i> 2 var.	.	.	×	×	×	×	.
<i>typhimurium</i> NTAP	×	.	.

* Serotypes found in feed ingredients fed to these pigs.

Table 4. *Salmonella* serotype associated with lot IIB pigs after leaving the farm

Serotype	Truck before	Truck after	Pen water	Pen	Arrival swab	Cecal swab	Carcass swab
<i>typhimurium</i> 1a	.	×	.	.	×	×	.
<i>typhimurium</i> 3a var.	.	.	×	×	×	.	.
<i>typhimurium</i> NTAP	.	.	.	×	×	×	×
<i>typhimurium cope</i>	×	×
1 var.	×	×
<i>derby</i> *	×	.
<i>muenchen</i>	×	.
<i>simsbury</i>	×	.

* Serotype found in feed ingredients fed to these pigs.

S. typhimurium not typed by available phage (NTAP) that could have been a homogeneous or a heterogeneous group. The second was the presence of *S. anatum* in the feed that lot I pigs consumed and also in the abattoir pen that they were confined to. With nothing similar to a phage typing system to solve this problem the *S. anatum* isolates from lot I rectal and caecal swabs were arbitrarily divided equally into the feed and environment source lists. The results of salmonella recovery by source are summarized in Table 5 and Figs. 2 and 3. Findings from lot I indicate that the contaminated abattoir environment contributed the greatest number of positives of both rectal and caecal swabs. In lot IIB pigs the carry-over

of environmental types to the kill floor was not so marked as in lot I pigs according to these methods of ascertainment. When only the pen was contaminated in lot IIA no arrival rectal swabs contained a salmonella from any source even though

Table 5. *Percentage recovery of salmonella from rectal and caecal swab samples by most probable origin of salmonella type*

	Swabs	No. of samples	No. positive	Positive (%)	Positive feed type (%)	Positive environment type (%)	Origin unknown (%)
Lot I	Rectal	50	36	72	40*	54*	5.5
	Caecal	49	26	53	33*	60*	8
Lot IIA	Rectal	40	0	0	0	0	0
	Caecal	50	9	18	44	11	44
Lot IIB	Rectal	48	19	40	21	68	11
	Caecal	48	17	35	38	26	35
Lot IIC	Rectal	40	0	0	0	0	0
	Caecal	50	10	20	35	0	65
Lot IIIA	Rectal	50	4	8	25	0	75
	Caecal	50	6	12	17	0	83
Lot IIIB	Rectal	48	0	0	0	0	0
	Caecal	48	1	2	0	0	100

* *S. anatum* found in feed and environment—strains from these samples were arbitrarily divided equally into feed and environment source lists.

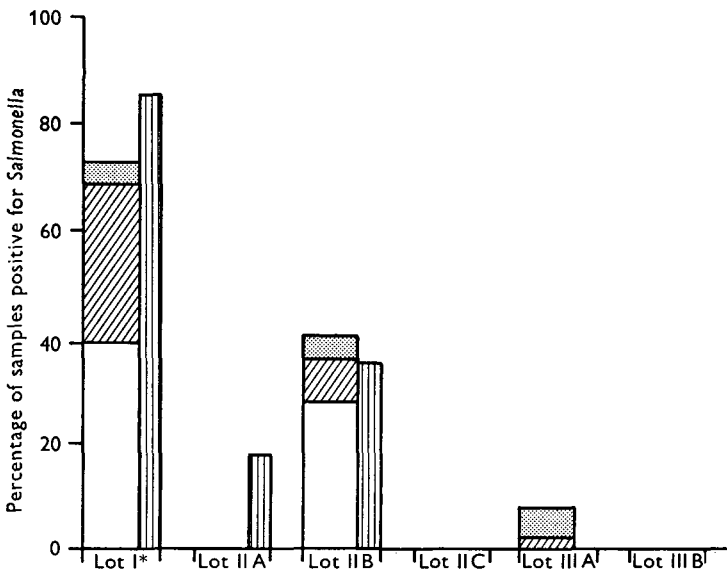


Fig. 2. *Salmonella* recoveries from rectal swab samples taken on pigs' arrival at the plant with proportions of isolations from different sources. (The percentage of environmental positive swabs are shown for comparison.) **S. anatum* found in feed and environment. Plates from which this type only was picked were divided equally between these two sources. □ Environment source. ▨ Source unknown. ▩ Feed source. ▪ % of environmental swabs positive.

this group was allowed the settling period; however, following an overnight stay in the pen 11% of the caecal swab recoveries were environmental source types.

Feed source salmonellas were isolated from rectal swabs from lots I, IIB, and IIIA in percentages varying from 21 to 40%. Caecal swab positives attributed to feed sources were recovered in all but the last lot of pigs. The highest of these percentages was 44.

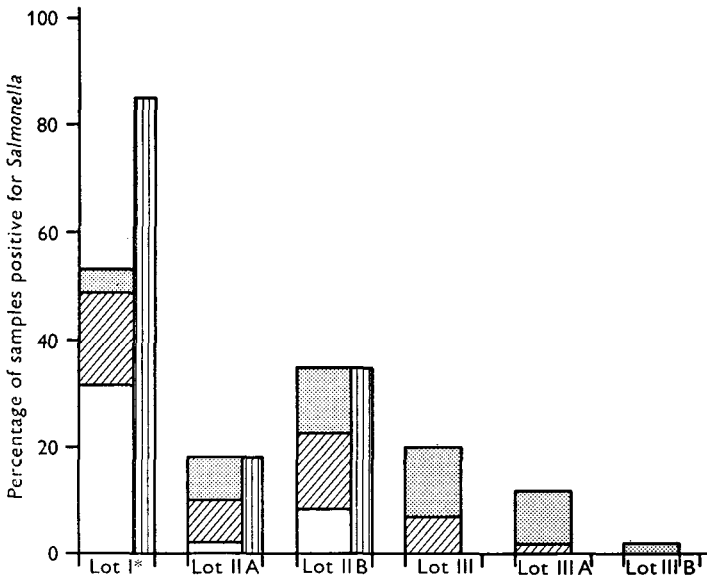


Fig. 3. *Salmonella* recoveries from caecal swab samples with proportions of isolations from different sources (The percentage of environmental positive swabs are shown for comparison.) **S. anatum* found in feed and environment. Plates from which this type only was picked were divided equally between these two sources. □ Environment source. ▨ Source unknown. ▩ Feed source. ▤ % of environmental swabs positive.

Table 6. Recovery of salmonellas from carcass swabs and the probable source of these isolations

Date of collection	Carcass samples			Probable origin of carcass isolations		
	No. of samples	No. positive	Positive (%)	Feed	Environment	Unknown
(Lot I) 27 Nov. 62	39	4	10	2 (3)*	1 (2)*	0
(Lot IIB) 29 Jan. 63	40	8	20	0	6	2
(Lot IIC) 19 Feb. 63)	40	3	8	1	0	2

* *S. anatum* was recovered from both feed and environment. Isolates could be from either source.

Carcass swabs

During the course of these investigations carcass swabs were taken from lots I, IIB, and IIC. The results of these samplings are shown in Tables 2 and 6. An effort was again made to appraise the probable source or origin of these strains. Feed and the packing plant environment appeared to play equal roles here, though the numbers are very small.

DISCUSSION

The isolation of only seventeen salmonella serotypes from the feed ingredient samples collected in this study must be considered the minimum number of salmonellas present. The number and amount of samples were small and probably not representative. The commercial situations and the limited resources of this investigation restricted the sampling procedures. The very extensive investigation of salmonellas in fish meal by Jacobs *et al.* (1963) indicates the possible deficiencies of our estimate of salmonella content of feed. These workers failed to isolate salmonellas from five 10 g. samples from each of seven bags of fish meal; however, when the entire contents were examined six of the bags yielded salmonellas.

Eight of the seventeen serotypes found in the feed were later isolated from one or more of the rectal or caecal swabs. Only one of the eight (*S. anatum*) was also isolated from the packing plant environment. Our findings were in agreement with Smith (1960) and the earlier work of Newell *et al.* (1959) that indicated that either host selection factors or the infecting dose of salmonella may influence the risk of a pig becoming infected or excreting a salmonella serotype when consuming contaminated feed. Seven serotypes were found in pigs that were not isolated from the six lots of feed or from the abattoir environment. Their source was unknown. The work of Smith (1960) also indicated that the pig, possibly acting as a sort of biological filter, could activate and excrete serotypes from the feed that had not been demonstrated by extensive laboratory examinations. Some of the seven unknown source types may have actually been present in feed ingredients. We would also recall that the first three lots of pigs consumed feed of unknown salmonella content for from 2 to 10 weeks before going on to a sampled feed.

An explanation of why no salmonellas were found in rectal swabs taken on the farm and yet were present after transport has been presented and discussed elsewhere (Williams & Newell, 1967). This change was probably related to stress-induced excretion. The rarity of salmonella excretion on the farm was emphasized in prior experiments using the same farm and the same methods we have described for this study. One pen lot of pigs was taken off antibiotic feeds and rectal-swabbed three times in 72 hr. No salmonellas were isolated from these specimens. At another time five pen lots of pigs (203 total, including just weaned and young pigs) were sampled and only one specimen was positive for a rare serotype (L. P. Williams & K. W. Newell, unpublished data). It was identical with the stock culture being used to check media at the beginning of this study and may have been an error.

The speed with which salmonella serotypes from the pen watering trough were

excreted by entering pigs (1½ hr exposure) was an unexpected finding. In a British study (Green & Jewell, 1965) food passage rates in pigs were shown to range 24–48 hr. in pigs fed on a standard ration and from 14 to 48 hr. in pigs given senna as a purgative. These animals were not stressed by transit, however, and passage time was measured by the feeding and recovery of polystyrene markers, not by passage of bacteria. There may well be a great difference between food passage rates through farm pigs and the passage of water through market pigs after transport and accompanying stresses.

In two recent controlled studies conducted in the Netherlands, the authors demonstrated that heat-decontaminated feed ingredients and pelleted feeds either prevented or decreased salmonella recoveries from test animals after slaughter (Edel *et al.* 1966; Kampelmacher, Guinée & van Keulen, 1965). We chose exclusion of animal origin products as our method of decreasing the salmonella content of feed because it was the most acceptable to the farm management. They feared the results of heat-treating the supplement as it was not known how it would affect palatability and weight gain. These fears may have been well founded. In the study using heat-decontaminated meals (Kampelmacher *et al.* 1965) it was observed that pigs fed on the decontaminated meal did not have a daily weight gain equal to that of control pigs (510 g. *vs.* 550). Our farm observed no difference in the length of fattening time during the special feeding period. The special feed was also cheaper than the feed regularly used on the farm.

Salmonella recovery rates from both rectal and caecal samples were less in all pigs that consumed the special feed. This was most marked in lots IIC, IIIA and IIIB, that is in pigs that never consumed a known highly contaminated feed until they were well developed or came in contact with a contaminated abattoir environment. It is interesting to note the decrease in caecal swab positives from 10 of 50, to 6 of 50, to a final low of 1 of 48. In experiments with chicks Milner & Shaffer (1952) demonstrated that older chicks were more refractory than young ones to induced *S. typhimurium* infection. If these findings are applicable to young pigs, and if the pelleted weaner ration was salmonella-free, they may account for the unexpected low recovery rate in lots IIIA and IIIB even though these lots consumed regular feed before slaughter.

The pigs fed on special feed for a longer time excreted fewer salmonellas and fewer feed type salmonellas. The majority of the last three lots of pigs did not excrete salmonellas at all on arrival at the plant. They went through a clean plant environment and left it clean for the next group of animals destined to occupy the same pens.

This study demonstrates that there is a build-up of salmonellas at the slaughterhouse and that some of these serotypes come from a contaminated environment. But some of the salmonella excretion and infection found at this time (and later by caecal swab after slaughter) is related to the feed consumed at the farm. These infections may persist, may be the source of infection to other pigs, may be related to abnormal excretion due to stress and other factors, and may in the end contaminate carcass sides. If this is the pattern, there is a chain from animal feed to the kitchen of man.

SUMMARY

A commercial swine fattening ration containing animal origin ingredients was shown to be related to the salmonella excretion of market pigs being sent to slaughter from a well-managed farm. When similar animals from this farm were fed a ration of much lower salmonella content, due to exclusion of ingredients of animal origin, their excretion declined as measured by both rectal and caecal swabs. When subsequent lots of pigs consumed a pelleted weaner ration and the special supplement only, before being exposed to a supplement containing the usual level of contamination, they seemed to be refractory to infection. Probably, this was a function of age at first exposure.

With prospective methods it was possible to show that salmonellas from the abattoir environment could infect pigs, that they would excrete them within a very short time, and that organisms from this source could be demonstrated at slaughter and shown to be a cause of carcass contamination.

While these findings support the view that the build-up of salmonellas in pigs is by contact with a contaminated environment, they indicate that the primary source of the contamination is most probably the salmonella-excreting pig which has consumed contaminated feed ingredients on its farm of origin.

If the great majority of pigs went to slaughter salmonella-free they would not serve as a source of infection to other pigs being sent to slaughter. Their intestinal contents could not contaminate carcass sides in the slaughtering process. This should help to prevent the contamination with salmonellas of the food preparation area of both homes and commercial eating establishments.

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