# An environmental survey of bakehouses and abattoirs for salmonellae

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#### INTRODUCTION

Outbreaks of salmonella infection are frequently associated with bakehouses, and almost every sizeable outbreak of paratyphoid fever in this country in recent years has been spread by bakers' confectionery. It is now known that certain ingredients used in the bakery trade harbour salmonellae, including *Salmonella paratyphi B*. Outbreaks of salmonella food poisoning are not so frequently associated with butchers' meat, but during the course of the investigations reported here evidence was obtained that some of the less dramatic, but widespread, outbreaks of disease in human beings might be spread by this product. Salmonellae are commonly found in animals, and all unsterilized animal products are likely to be contaminated with these organisms from time to time.

The surveys reported here were begun after an extensive outbreak of paratyphoid B fever, associated with bakers' confectionery in South Wales, because it was then apparent that infection was not carried into the bakehouses by the bakers but was introduced in infected materials (Culley, 1953; Thomson, 1953). Since this investigation was begun, imported egg products (Newell, 1955; Smith & Hobbs, 1955; Newell, Hobbs & Wallace, 1955) have been shown frequently to harbour salmonellae, including S. paratyphi B. The surveys were continued, however, in order to get some evidence of frequency of infection of bakehouses by salmonellae in the absence of any outbreak of disease in human beings, as it is of great importance, in the study of epidemic disease, to understand what happens during interepidemic periods. The surveys later produced evidence of infection of the staff in the bakeries and of illness in the general public apparently spread from abattoirs, but this did not alter the fact that it was the primary intention of the investigation to gain insight into the spread of salmonella infection without waiting for the occurrence of clinical human illness.

#### MATERIALS AND METHODS

Samples were taken, using the Moore's gauze swab technique (Moore, 1948). In bakehouses, samples were taken from the sullage drains which received the washings of the bakehouse floors and also from the drains receiving the excreta of the staff. In the bakehouse chosen for this investigation, the two drainage systems were separate, and as both were water trapped, there appeared to be no likelihood of cross-contamination by means of rats. It was, therefore, assumed that infection of ingredients and of staff were separately reflected in the isolations of salmonellae from each type of drain. Samples were usually taken at weekly intervals. In addition, specimens were examined from time to time from the staff wash-hand basins and from the surface of several articles of bakehouse machinery (a preliminary account of some of this work has already been published (Harvey, 1957)).

The abattoir samples were taken from five drains. The first drained blood from pigs, the second blood from cattle and the third the contents of pig gut. The fourth drain sampled blood from cattle and sheep and the fifth drain collected the contents of the beef gut processing department. It will be seen from Table 2 that the number of samples examined from points IV and V were relatively small.

Ta	bl	e	l.	Isolations	of	' salmonellae	from	a singl	le ba	kery	1955-56
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	Number of tin	nes isolated
F bakery	loor drains in preparation rooms	Bakery staff sewage
Total swabs examined	111	93
Total swabs positive	31	15
Serotype isolated:		
Salmonella typhimurium	8	8
S. aberdeen	12	1
S. thompson	9	0
S. paratyphi B	2	2
S. kentucky	0	2
$S. \ sundsvall$	1	1
S. ness-ziona	0	1
S. newington	1	0
Total serotypes isolated	6	6

The laboratory technique was a simple one gradually developed from past experience (Harvey & Thomson, 1953; Harvey & Phillips, 1955; Harvey, 1956; Harvey, 1957). The swabs arrived at the laboratory in wide-mouthed screw-capped jars and were never removed from them. Single-strength selenite F broth containing a final concentration of 1 in  $10^6$  brilliant green was poured on to each swab which was then compressed several times with a sterile glass rod to express its fluid into the enrichment medium. The swab, in its surrounding enrichment medium, was incubated at  $43^\circ$  C. in a water-bath, and subcultures were made at 24, 48, 72 and 96 hr. on to brilliant green MacConkey agar (Wilson & Darling, 1918) and on to de Loureiro's (1942) modification of Wilson and Blair's bismuth sulphite agar. These plates were incubated at  $37^\circ$  C. and were examined at 24 and 48 hr. Suspicious colonies were picked and examined by direct slide agglutination with polyvalent O salmonella serum and polyvalent H specific and non-specific salmonella serum. Colonies giving positive results with the polyvalent sera were then further investigated. No screening media were used, and in almost every case salmonella colonies could be identified accurately by naked-eye examination checked by slide agglutination. The prolonged incubation of the enrichment media was found to be valuable, not only in the separation of salmonellae from nonpathogens, but also in the separation of one salmonella serotype from another. It was for instance, sometimes found that one serotype predominated at the 24 hr. subculture while a different serotype was dominant in a later subculture (Harvey, 1957).

Table 2.	Drains from which salmonellae were isolated in
	Glamorgan abattoirs 1957–59

				Cattle/		
	Pig	Cattle	Pig gut	sheep	Beef gut	
	slaughter	slaughter	processing	slaughter	processing	
Specimen	drain	drain	drain	drain	drain	Total
Number examined	102	75	81	9	7	<b>274</b>
Number positive	37	35	31	4	4	111
Serotypes:						(40.5%)
Salmonella typhimuriu	m 13	17	15			45
S. dublin	5	9	5		1	20
S. derby	1	2	1		-	4
S. meleagridis	3	3	1			7
S. anatum	2		5			7
S. thompson	2	1	3		1	7
S. bovis-morbificans	2			_	_	2
S. muenchen	4					4
S. senftenberg	2					2
S. enteritidis (jena)		2			_	2
S. enteritidis (danysz)		<b>2</b>	1			3
S. enteritidis (chaco)		1				1
S. kiambu	1					1
S. kentucky	2			_		2
S. abony	1					1
S. weltevreden	1					1
S. paratyphi B				1	_	1
S. heidelberg				2		2
S. agama				1		1
S. london					1	1
S. infantis			_		1	1
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#### RESULTS

#### Bakehouse survey

As was expected, salmonellae were frequently found in the bakehouse (Table 1). Of the 111 samples taken from the bakehouse floor drains receiving spilt raw materials, 31 were positive, the three common types being S. typhimurium, S. aberdeen and S. thompson, which were found on eight, twelve and nine occasions respectively. It will be noted also that S. paratyphi B (phage-type 1) was found on two occasions.

Of the 93 specimens taken from the drains receiving the sewage of the staff, fifteen were found positive. S. thompson was never isolated and S. aberdeen only once. S. typhimurium was found on eight occasions and S. paratyphi B on two occasions.

It was by no means difficult to find samonellae inside the bakehouse. In addition to their occurrence in the floor gullies, they were found in three out of 42 samples from the out-flow water from staff wash-hand basins and from several swabs which had been wiped over the surface of the bakehouse machinery. S. typhimurium and S. aberdeen were cultured from wash basins and S. thompson and S. virchow from bakehouse machinery surfaces. Routine cleansing of a cake mixer did not prevent us from isolating S. thompson from its surface subsequently.

It is clear, therefore, that the bakehouse investigated was contaminated with many types of salmonellae. There is no reason to believe that the conditions at the bakery examined or the materials employed were significantly different from other bakeries.

It is suggested, therefore, that salmonellae, including S. paratyphi B, are likely to be very frequently present in bakehouses. Certain kinds of bakers' confectionery are ideal culture media for salmonellae and the potential risk to health would appear to be great. Nevertheless, having regard to the theoretical risk it must be admitted that outbreaks of disease are surprisingly rare. In the area served by this laboratory there is a population of nearly one million with, presumably, several hundreds of bakehouses, yet, in a period of 5 years, there were only two outbreaks of salmonella food poisoning undoubtedly spread by bakers' confectionery and of these one was a relatively minor affair. Neither of these two outbreaks was associated with the bakehouse in the survey; one of the outbreaks is the subject of a separate communication (Harvey, Price, Davis & Morley-Davies, 1961).

#### Abattoir survey

Many samples taken from gullies of the abattoirs were found positive, and 21 different types of salmonellae were isolated, as shown in Table 2. Seventeen of these serotypes occur in animal feedingstuffs. Almost 41 % of all samples were positive. The commonest serotypes found were *S. typhimurium* and *S. dublin*. The occurrence of *S. typhimurium* was of particular interest because, for the first 20 months of the survey, this organism was never isolated from abattoir swabs, and in the second 16 months it was the type most frequently found. *S. typhimurium* was first found in the gullies of the abattoirs in September 1958, in the 21st month of the survey, as shown in Table 3. The frequent isolation of salmonellae from abattoirs was less surprising than the frequent isolation from bakehouses. It was thought, however, that the occurence of salmonellae as a contaminant of butchers' meat would carry less risk to the human population than similar contamination of bakers' confectionery. We had later to modify this opinion.

In September 1958, when S. typhimurium was first isolated from the abattoir, after failure to culture this serotype over a period of 20 months, there was a slight rise in the number of human infections due to S. typhimurium in Glamorgan. From January to August, a total of 14 human cases had been identified. In September there were eight cases, and in October and November the increase in human infections due to this serotype was even greater (56 in October and 20 in November). These cases were widely scattered with none of the usual vehicles to link them together. It was found that the phage-type of S. typhimurium isolated

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from the abattoir gulley in October and November and of the strains isolated from the majority of the human infections during the same 2 months, was identical— 1a var. 3. This phage-type was not isolated once from abattoir swabs during the next 12 months, although many other phage-types of *S. typhimurium* were isolated from this source.

At the end of 1958 we were informed by the Veterinary Investigation Department in Cardiff that cases of infection due to S. typhimurium were occurring in calves on certain farms in Glamorgan with some deaths in the animals infected.

<b>Month</b> isolated	Cultures from diseased animals*	Abattoir swab cultures	Human infection cultures
1958:			
October	la var. l; la var. 3	la var. 3	la var. l; la var. 3
November	la var. 3	la var. 3	1a var. 3; 1a
December	la var. 3		la var. 3
1959:			
Januarv	Untypable		la var. 3
February	· · · · ·	Untypable	la var. 3
March	•	Untypable	Untypable
April		1b	Untypable
May		la; untypable	1a; 24
June	•	2b; 1a var.2	1a; untypable U 29
July	la var. 2	la var. 2; lb; U15	U 29; 32 24
August	•	la var. $2; 1a;$ U 30; untvpabl	la; la var. l e 32
September	1	2a; 2b; 2c; 1b	1; 1a var. 3; 2b; 2c; 1a; 2; 1a var 1
October	2	l var. 5; U15; untypable	2; 1a val. 1 U59; 1 var. 5; 2c
November			l var. 5
December			1a; 2c

## Table 4. Phage-types of Salmonella typhimurium isolatedeach month 1958–59

\* From Veterinary Investigation Department.

Strains of the serotype were obtained, and on three out of five of the infected farms the phage-type responsible for the illness was 1a var. 3. This phage-type was isolated from calves in October, November and December 1958, but never subsequently (Table 4).

Routine investigation of the human cases of illness, which were scattered over several local authority areas, failed to incriminate any single foodstuff, bakehouse or store. There was, therefore, the suspicion that some incidents of S. *typhimurium* infection were caused by eating infected local butcher's meat. The case for this was later strengthened by analogy with observations on illness due to other salmonella serotypes. There was, for instance, a tendency for human cases due to S. *enteritidis* 

(jena), S. derby, S. heidelberg, S. meleagridis, S. agama and S. muenchen to occur in the local population in the same quarter of the year as corresponding samples from the abattoirs were found positive for these serotypes.

#### Human and animal strains compared

S. typhimurium had not been isolated for over a year prior to September 1958 and was regularly isolated from abattoir swabs during 1959. It seemed profitable, therefore, to compare the phage-types of these strains with those of cultures isolated from human subjects during the same period. Prior to the use of phagetyping, it was almost impossible to trace the origin of sporadic human infections with S. tuphimurium. The phage-typing scheme for this organism, introduced by Felix & Callow (1943, 1951) and Felix (1956), was extended by Callow (1959), and the scale on which the typing of S. typhimurium of human and animal origin is carried out has recently been greatly increased. As the result of this work it is now possible to compare the distribution of phage-types in man with that concurrently prevalent in livestock (Anderson, 1960). It has been shown, for example, that types occurring in cattle can be related to outbreaks of human infection. Such studies have shown that diffusely scattered, apparently sporadic cases of S. typhimurium infection, can be connected with each other and with the sources of their infection. It was thus hoped that, by parallel observations on abattoir and human isolations, some light might be shed on what proportion of such seemingly sporadic infections might be presumed to be due to local contaminated butcher meat products. Representative cultures of S. typhimurium from human infections and from abattoir swabs were, therefore, sent to the Central Enteric Reference Laboratory, Colindale, for phage-typing from October 1958, until the end of 1959. A few strains provided by the Veterinary Investigation Department were included. The results are given in Tables 4 and 5.

It will be seen that the phage-types of S. typhimurium isolated from the slaughter house swabs corresponded in type, and not infrequently in timing, with the phage-type isolated both from animals and from human infections. It was noted that of the 30 typable human strains of S. typhimurium, 23 (76.7%) belonged to phage-types found in the abattoir environment, or in local farm animals. If we exclude strains isolated from definite outbreaks, i.e. phage-type 1a var. 3 from October 1958 to February 1959 and phage-type 2c in December 1959, the remaining 25 typable cultures represent sporadic incidents. Of these, 18 (72%) are once more types found in the abattoir swabs and in the faeces of local farm animals. The simplest explanation of this finding is that many of the sporadic incidents are associated with eating infected local meat products.

It is the experience of most laboratories that, apart from dramatic outbreaks of food poisoning from one small identifiable source, there occur, from time to time, incidents of disease not apparently coming from one source, but of a more diffuse nature. It is suggested that these might be associated with contaminated butchers' meat.

It has been established very recently (Hobbs & Wilson, 1959) that imported carcass meat can be contaminated with salmonellae. It has been shown also that

fresh pork sausages can harbour salmonellae (Galton, Lowery & Hardy, 1954). In order to see how frequently fresh meat in the abattoir described here was contaminated by salmonellae a limited number of specimens of pork sausage, pig bile, pig blood, pig spleen, pig liver, cattle spleen, cattle liver and cattle blood were examined. The results of these specimens are given in Table 6. In all 215 specimens were examined with only one positive.

### Table 5. Distribution of phage-types of Salmonella typhimurium in diseased animals and abattoir drains in Glamorgan in 1958–59

Phage-type	Pig faeces	Cattle faeces	Pig slaughter drain	Cattle slaughter drain
1	•	+	•	
1 var. 5	•		+	+
la var. l		+		•
la var. 2	+	+	+	+
la var. 3	•	+	+	+
1b				+
2	+			
2a	•	•	+	•
<b>2</b> b	•		+	+
2c	•	•	+	•
U15		•	+	+
<b>U 30</b>	•	•	+	
Untypable	•	+	+	+

Table 6. Miscellaneous specimens of animal origin examined for salmonellae

Specimen	Total samples	Number positive	Serotypes isolated
Pig spleen	22	0	
Pig liver	20	0	•
Pig blood	7	0	•
Pig bile	30	0	
Pork sausage	84	1	S. poona
-			S. reading
Ox spleen	21	0	•
Ox liver	21	0	
Ox blood	10	0	•

#### DISCUSSION

Samples taken from the floor gullies of bakehouses showed 31/111 (27.9%) to be positive for salmonellae, and having regard to the potential risk it is surprising that outbreaks of food poisoning carried by bakers' confectionery are not more common. Yet, in a period of 5 years, in the county of Glamorgan, we have encountered only two incidents of food poisoning due to salmonellae which can definitely be said to have been caused by eating bakers' confectionery. Both of these occurred in 1959.

We have found the sewer-swab technique an effective means of surveying the environment of food premises for salmonellae. In the abattoir it was more effective in the demonstration of the presence of salmonellae in the environment than was the examination of meat specimens. This is probably due to the size of sample represented by a single sewer swab. This sampling technique also probably accounted for some of the more unusual serotypes isolated.

The discovery of S. paratyphi B (phage untypable) in an abattoir drain, not subject to human contamination, raises the question of occasional incidents of paratyphoid B fever carried by meat. This question has been discussed previously (Editorial note, Medical Officer, 1951; Nicol, 1956; McCann & Cross, 1956; Bernstein, 1958).

The occurrence of salmonellae in abattoirs was less surprising than their isolation from a bakehouse environment. It was not known, however, what part was played by infection of butchers' meat in the dissemination of salmonellae amongst human beings. A close inspection of the results obtained in abattoirs and the occurrence of human disease produced circumstantial evidence that some of the less dramatic incidents of salmonella infection are indeed spread by butchers' meat. A similar conclusion was reached by McDonagh & Smith (1958). It was thought that rats played little or no part in these findings. Over 200 rats caught outside the abattoir were examined with entirely negative results. It is not, therefore, probable that salmonellae were introduced into the abattoir by rats from outside. We would consider that our isolations of salmonellae from the slaughter house drains reflected infection introduced by farm animals entering the abattoir for slaughter.

The observation that 16 % of specimens from bakery staff sewage were positive was of interest. This figure reflected the degree of potential infection of personnel in a trade dealing with raw materials very frequently contaminated with salmonellae. This point has recently been raised (Leading article, Lancet, 1958). Our results, however, led us to the conclusion that bakery staff did not excrete salmonellae for a prolonged period, as the staff sewers were only very intermittently positive. This observation was important, as it suggested that only relatively small numbers of salmonellae were being excreted into the sewers and contrasts with results previously obtained when dealing with human cases of frank disease (Harvey & Phillips, 1955). The suggestion that the personnel of certain trades including bakeries should be regularly examined for excretion of salmonellae does not, therefore, promise to be a profitable method for the discovery or control of infection. The danger in the bakehouse arises from the entry of infected materials and not primarily from the staff. There is indeed evidence that bakers themselves acquire infection from these materials. McCullough & Eisele (1951a, b), by human feeding experiments with salmonellae, suggested that the infection rate and the duration of excretion was a function of dosage, small doses of the pathogens giving rise to short periods of excretion. It seems possible that the type of infection that occurs in bakery staff, who are probably subjected to frequent contact with salmonellae in small dosage, is of this order. Newell, McClarin, Murdock, Mac-Donald & Hutchinson (1959) have drawn a similar analogy in pigs excreting salmonellae. It was interesting to contrast the serotypes found in bakeries with those discovered in abattoirs. The only type commonly found in both environments was S. typhimurium. This was not surprising, as this serotype, more than

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any other species of salmonella, is a universal pathogen. It is common to egg products, farm animals and animal feedingstuffs. The remaining bakery serotypes were mostly those found in Chinese frozen egg, a material in daily use in the bakery during the major part of the survey. S. kentucky and S. ness-ziona have not been found in Chinese egg and their origin may, therefore, be different. It is possible, that other contaminated raw materials, apart from egg, introduce salmonellae into bakeries.

The contamination of the bakery environment was easy to account for by the amount of egg visible on floors, tables, etc. Some of the staff were covered in egg from hand to elbow, so that the isolations from wash-hand basins were understandable.

The wide range of salmonella species found in the abattoir would lend support to the hypothesis that farm animals become infected by means of animal feedingstuffs. These materials contain a very wide range of serotypes and 17 out of 21 species found in the abattoir are known to occur in animal foods, or in the raw material incorporated in them.

We feel that the surveys described above serve to illustrate certain local and remote aspects of the salmonella problem. Unfortunately, the solution of this problem is as yet far from being reached.

#### SUMMARY

1. Moore's gauze swabs have, in our hands, been shown to be a reliable instrument for the survey of salmonella infection in the environment of two food premises.

2. A large bakery was demonstrated to be regularly contaminated with salmonellae. Thirty-one floor gully swabs out of 111 (27.9%) were positive for salmonellae. Little spread of infection to the general public was experienced.

3. Staff infection with salmonellae was shown to occur in this bakery. Fifteen positive specimens of staff sewage out of 93 were obtained  $(16\cdot1\%)$ . There was little evidence of prolonged contamination of the sewers with salmonellae. Staff infections were probably, therefore, of short duration. The staff probably became infected from contaminated goods in the bakehouse. Rat contamination of sewers was excluded by water trapping.

4. It is not thought that regular examination of staff excrete would be a profitable means of discovering carriers or of preventing contamination of the products. The frequency with which such examinations would have to be performed to discover short-term excreters would be administratively impossible.

5. Of 274 swabs from gullies in abattoirs, 111 were positive for salmonellae (40.5%). Twenty-one different serotypes were isolated. Seventeen of these sero-types are known to occur in animal feedingstuffs.

6. Parallel examination of abattoir swabs and excreta from human infections for S. typhimurium, coupled with phage-typing of the strains, provided information of the geographically local nature of much sporadic illness due to this serotype. Of a total of 30 typable strains, 23 belonged to phage-types found in the abattoirs, or on local farms. Sporadic incidents were represented by 25 of these cultures, 18 of which corresponded in phage-type to those found in the abattoirs or in local farm animals. The phage-types of S. typhimurium isolated from the slaughter houses not infrequently corresponded in timing with the strains isolated from human infections.

We are indebted to Dr E. S. Anderson and the Staff of the Central Enteric Reference Laboratory and Bureau, Colindale, for phage-typing the cultures of *S. typhimurium*, which made it possible to expose the similarity in type distribution of cultures of human origin and those isolated from abattoirs and livestock.

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