

## The detection of a carrier of multiple phage-types of *Salmonella paratyphi B*

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### INTRODUCTION

Provisional figures for the total notifications of typhoid and paratyphoid fever throughout Scotland in 1957 and 1958 are given as 71 and 125. (Reports, 1957, 1958). The infection is now generally considered infrequent and the chance of water-borne infection remote. Against this background it seems worth while recording the following events not only for their academic and epidemiological interest but also as a cautionary tale.

### NARRATIVE, INCLUDING EPIDEMIOLOGICAL AND BACTERIOLOGICAL STUDIES

In July 1954, an Edinburgh boy, aged 13 years, (case I), was found to have a *Salmonella paratyphi B* infection, the organism (phage-type Taunton) being isolated from his faeces. Examination of his home contacts proved entirely negative, but some 2 weeks previously the boy had been at a camp about 15 miles from Edinburgh. There had been much paddling and bathing in the river beside the camp (see map) and the boy admitted that, contrary to instructions, he had drunk the river water on at least one occasion.

The camp drinking-water supply (a spring under the bank of the river) was examined and found to be of good quality and free from pathogenic organisms; water from a small tributary entering the river beside the camp and which received the sewage effluent from a nearby village, S, was also found to be free from pathogens; but *Salm. paratyphi B* was isolated on two occasions from samples of the river water taken about 100 and 200 yards upstream between the camp and A, a hamlet of fifteen houses on the river banks. One of these strains belonged to phage-type Taunton; the phage-type of the other strain was not determined, but it seemed obvious that the river water was the vehicle of the boy's type Taunton infection.

The river was a typical lowland stream with its origin in a bog alongside a main

railway line, and close to a signal box and railway cottages. Thereafter it flowed some 3 miles across a grassy moor where it received drainage from a mansion house, and from three farms and their workers' cottages; thence under a trunk road to A about  $\frac{1}{2}$  mile distant. Of the fifteen houses in A, five were condemned at the time of this investigation although one of these was still permanently occupied by an old lady (Mrs X) who did not wish to move, and another was used as a holiday house. Of the remainder, eight were permanently occupied and two were used as holiday cottages. Eight of the fifteen houses had water-borne sanitation to septic tanks, but the outflow of the effluents to the river was not easy to locate. The river, at that point a shallow, stony stream about 10 ft. in width was crossed by a road bridge; after which for a short distance the banks were low, and then high, rocky and wooded and difficult of access. Upstream and adjacent to the road bridge the largest house was a one-time inn, standing almost on the water's edge. Across the bridge from the inn and on the other side of the road was a row

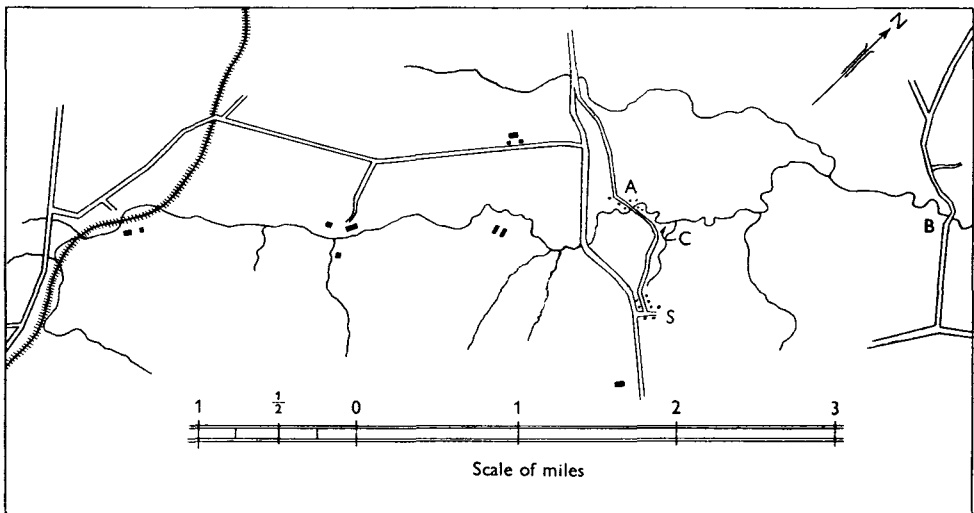


Fig. 1. Sketch-map of stream from source to 'B'. A, hamlet; B, bridge; C, camp site; S, village.

of houses; nearest the river were two small semi-detached cottages without water and inside sanitation, the two condemned but still occupied houses already referred to. The sanitary accommodation for these two cottages consisted of privies situated only a few yards from the bank of the stream. Downstream from the bridge at A, the steep banks extended for about  $\frac{1}{4}$  mile until the river again reached flat country near the site of the camp and received the small tributary containing the effluent from S. Thence it flowed through grassy country, was joined by two streams with a farm on the banks of each, and continued on its winding course for a mile through uninhabited country, under sparse trees, to a road bridge at B.

The isolation of *Salm. paratyphi B* from the river between the camp site and A pointed to the presence of a carrier living upstream. All the residents at A were questioned but no suggestive history of enteric fever was elicited. Extensive

sampling of the river and tributaries upstream was also carried out, but with negative results; and after some months of fruitless search, the matter was allowed to lapse. Pollution was assumed to have been caused by an itinerant, or possibly by the discharge from a train toilet at any of the several points where the railway crosses or is adjacent to the river, as shown on the map.

Nothing further of interest came to our notice until some 4 years later, when in June 1958, *Salm. paratyphi B* was isolated from the faeces of a farm worker, aged 68 years (case II). He had a typical though mild illness, was admitted to hospital and made an uneventful recovery.

This man lived with his sister in a modernized cottage about  $\frac{1}{2}$  mile uphill from the bridge at B. He had 'never been ill in his life', never been abroad and seldom went far from home. Examination of his sister's faeces, urine and blood gave entirely negative results; the household water supply was of good quality and found free from pathogenic organisms; but with the knowledge that in 1954 this same stream had been polluted, water from the river taken near the bridge at B was immediately examined and *Salm. paratyphi B* isolated. The man denied having drunk any river water, and there seemed to be no link with the river until it was discovered that every week he cleaned the intake grating to a hydraulic ram which pumped spring water from river level to supply his own and other houses. The ram was housed near the bridge at B and the intake was about  $\frac{1}{4}$  mile upstream. A rake was supplied to clear away the leaves and debris from the intake, but in summer it was doubtless easier to use one's hands. It seemed then that the immediate source of this man's infection was obvious until it was found that his strain of *Salm. paratyphi B* belonged to phage-type Dundee, and that from the water at B to type 3aI var. 4 (see Table 1).

Table 1. *Isolations of Salm. paratyphi B phage-types*

Source	1958							
	1954 Aug.	June	July	Aug.	Sept.	Oct.	Nov.	Dec.
Three cases	T	D	. T	. .	. .	. .	. .	. .
River water at B	.	4	. T	T .	. .	T T	4 D	. .
							D 1	
River water at A (upstream from B)	T	.	D .	. .	T 4	. .	. .	T 4 4 D D
Carriers: Mrs X	.	.	. 4	. .	. .	4 4	4 .	. 4 T D
Mrs Y	.	.	. T	. .	. .	. .	. .	. .

Phage types: T = Taunton; D = Dundee; 1 = 3aI var. 1.; 4 = 3aI var. 4.

Samples of water were then taken at appropriate points upstream from B, from the two tributaries just before they joined the main stream, and from the river

some yards below the bridge at A. Nothing of interest was found in the samples from the two tributaries, but *Salm. paratyphi B*, phage-type Dundee, was isolated from the sample taken downstream from the bridge at A. Apparently, then, two carriers must be sought, one to account for the Dundee infection of case II, and another for the pollution of the river with type 3aI var. 4, as distinct from the source of the type Taunton infection in 1954.

Early in the investigation, the method used in examining water was to put the whole of a 250 ml. sample through a Seitz filter, cut up the filter pad, and place one-half in 'Selenite F' medium and the other in tetrathionate broth. After 24 hr. incubation, subcultures were made on sucrose-desoxycholate-citrate agar and Wilson and Blair's bismuth sulphite medium. If necessary, subcultures were again made from the primary cultures after 72 hr. incubation. Incubation at 37° C. was found satisfactory. Later, the swab technique recommended by Moore (1948, 1950) for the examination of sewage was used. Swabs were made from sixteen layers of stockinet such as is sold for domestic purposes; and plates inoculated from the primary selenite cultures, undiluted, and diluted 1 in 10, and 1 in 100.

At this stage, in the course of interviewing all the twenty-nine inhabitants of A, a suggestive history was obtained from the old lady, Mrs X, aged 81 years, who lived in one of the condemned cottages near the bridge. She had lived there since 1948, and previously some 20 miles away. She was almost stone-deaf and her memory very poor, and at the time of the inquiry her sister-in-law, Mrs Y, aged 87 years but less deaf, was paying her a long visit. Mrs Y had lived at A until 1956 when her cottage had been condemned and had 'never been ill' herself, but, mainly owing to her prompting, Mrs X was reminded of having been 13 weeks in the district isolation hospital in 1938 or 1939 with 'paratyphoid'. Unfortunately no hospital records are available which might have confirmed and added to this information. *Salm. paratyphi B* was isolated from the faeces of both Mrs X and Mrs Y. No relevant history was obtained from anybody else at A; and a specimen of urine and faeces obtained from each individual proved negative on examination.

It was now discovered that Mrs X habitually disposed of the contents of her privy pail by quite simply emptying it into the river only some 10 yards distant. Steps were at once taken to put a stop to this. Mrs X was told that urine and faeces must on no account be put into the river and her chamber pot must be emptied into the privy pail. Owing to her age and infirmity special arrangements were made to ensure the satisfactory disposal of the contents. Moreover, after some initial failures a bacteriologically satisfactory method of disinfecting the excreta was arrived at, and examinations made at intervals.

Meanwhile further interest was added when it was found that the strains isolated from Mrs X and Mrs Y belonged to different phage-types: phage-type 3aI var. 4 had been isolated from Mrs X and Taunton from Mrs Y. Mrs Y had been resident in A in 1954 and could therefore have been the source of the Taunton infection (case I). This seemed to us highly probable as she also had had a dry closet. On the other hand, as the strain isolated from Mrs X belonged to phage-type 3aI var. 4, and it is generally accepted that for practical purposes a carrier excretes one phage-type

only, we were led to assume that Mrs X could not be the source of the Dundee infection (case II). The carrier of phage-type Dundee had therefore still to be discovered (see Table 1).

About this time (16 July) a boy, aged 16 (case III) who lived within 6 miles of the bridge at B, but in a different drainage area, and who had become ill about the beginning of July was found to have a *Salm. paratyphi B* (phage-type Taunton) infection. Examination of his home contacts proved completely negative, and it was only on very close questioning that he was discovered to have been fishing in the river, a popular trout stream, near the bridge at B about 10 days before he became ill, and to have drunk the water there. This seemed sufficient to account for his infection as phage-type Taunton was recovered from the water both at B and A at this time, with Mrs Y as the apparent source of this phage-type.

The search for the Dundee carrier went on. Swabs were placed in the drains of the houses in A upstream from where the Dundee strain had been procured which was 10 yards downstream from Mrs X's house. The effluent from village S was also examined. In addition, we reluctantly turned our attention to the upper reaches of the river. Swabs were placed at points below the entry of each tributary receiving the drainage from dwelling houses and farm buildings and also in the drains themselves. The 111 people living in the railway cottages, mansion house and farms were interrogated, their ready co-operation obtained, and a sample of faeces and urine procured from everyone. A specimen of blood for Widal test was taken from the only person whose history was at all suggestive of enteric fever. All this, however, was without any positive result. Meanwhile, *Salm. paratyphi B* of either phage-type Taunton or 3aI var. 4 was being isolated with monotonous regularity from the river below A and from the inlet to the ram at B. Measurements indicated that the volume of water passing the bridge at B was 60% greater than at A; but in spite of this degree of dilution conditions must have favoured the survival of paratyphoid bacilli, and it was necessary to warn the Scout Master in charge of a camp near B that the water was dangerously polluted. Mrs Y, the known Taunton carrier, had left the neighbourhood on 24 August. Mrs X denied putting any urine or faeces into the river. (Her co-operation could only be won by the expenditure of much tact and time.) It was only several weeks later that our suspicions were justified and Mrs X was discovered to be faithfully emptying her chamber-pot into the privy pail, but immediately thereafter rinsing it in the river. It seemed that this must be the explanation of the continued pollution of the river.

Up to this time then (October), we had obtained no evidence of pollution of the river above Mrs X's house in spite of our intensive investigations, and everything pointed to the source of pollution being within the few yards below the bridge. Could it be that Mrs X was excreting not one phage-type but three?

It was now becoming difficult to get the old lady's co-operation, so in an effort to ascertain how many phage-types were present in the water, another two samples were taken by swabs suspended at the ram intake at B. Five single colonies were picked from primary platings of the first, and three from the second. *Four* phage types were obtained from these: Taunton, 3aI var. 4, Dundee and 3aI var. 1.

When at last further specimens of faeces and urine were procured from Mrs X, eight

single colony cultures were made from primary platings, and phage-types Taunton and 3aI var. 4 were found. Mrs X was therefore presumed to be carrying more than one type; but it seemed advisable to eliminate any possibility of the Taunton strains (i.e. those of Mrs Y's type) having been introduced by the remote chance of contamination of Mrs X's utensils during Mrs Y's stay in the house 2 months previously; and after much staff work and delay, specimens of urine and faeces were procured from Mrs X which could be guaranteed uncontaminated by any other source of *Salm. paratyphi B*. Twenty-three single colony cultures were obtained from primary platings. Of these eight belonged to phage-type 3aI var. 4, six to Dundee, and seven to Taunton (two were rough and therefore untypable). All three types were obtained from both urine and faeces. Mrs X was therefore carrying at least three phage-types of *Salm. paratyphi B*; she might well have been the source of the Taunton infection in case I and case III, and we had found the Dundee carrier and the source of infection in case II.

The situation was difficult from both a human and public health aspect. Any treatment short of cholecystectomy would probably have been unsuccessful even if Mrs X could have been persuaded to go to hospital; but to leave such a prolific carrier so dangerously near a stream in such primitive conditions would demand impossibly strict and continued surveillance. The situation was resolved by obtaining Mrs X's consent to remove her to a modern house in a nearby village. It seemed, however, both unnecessary and unkind to demand further faecal specimens on the chance of finding phage-type 3aI var. 1 and on these grounds too no blood was ever procured for a Widal test. However, as no *Salm. paratyphi B* was isolated from continued samplings of the water at A and B after Mrs X ceased to live at A, it seems highly probable that she was the carrier of type 3aI var. 1 also.

#### DISCUSSION

There can be little doubt that for one reason or another pockets where poor housing conditions still exist are to be found in many rural districts throughout Britain, though it may be difficult to believe that the conditions found in Mrs X's house should persist after a 100 years of sanitary reform. The 'harmless' old lady continuing at her own request to live in her condemned cottage with her primitive sanitation and method of disposal of excreta seems at least a century out of date.

The comparatively negligible incidence of enteric fever in Great Britain is, after all, of quite recent date. In 1931 the total notifications in Scotland were 737 and the average for 1921-30, 535 (Report, 1931). In 1958 the notifications totalled 125. The average of the number of notifications given for England and Wales for the 5 years 1934-38 is 1785 as against 789 for 1952-56 (Reports, 1934-38; 1952-56). In the area under discussion, records going back to 1923 indicate a regular incidence of enteric fever with occasional sharp outbreaks up to 1940, and sporadic cases since then. It seems then that there must still be an appreciable number of chronic carriers, and that they are likely to be relatively more numerous amongst the elderly, the very section of the community who, in rural areas, tend to inhabit old insanitary houses. The risk from these carriers seems to be surprisingly latent



until circumstances combine to create a dangerous situation as they threatened to do at A. The prolific carrier, the privy so conveniently near the river, the excellent camp sites, a good trout stream in which conditions favoured the survival of enteric bacilli, and finally the intake to the ram which was in effect a bacterial filter all contributed to make the district a potentially epidemic area.

A carrier of multiple phage-types is of great academic interest. That Mrs X could have acquired at least three different infections during her stay in hospital or elsewhere seems to us improbable but must remain a matter for conjecture; and it seems more likely that the multiple phage-types she was excreting were due to type conversion *in vivo*. This is the subject of a separate study by Bernstein (1960) (see p. 201).

Apart from this, the phenomenon was of immediate practical importance for it indirectly added to the labour of our survey and led to many time-consuming field and laboratory investigations which in the event proved unnecessary. Since the work of Craigie & Yen (1938*a, b*) on *Salm. typhi* phage-types, and of Felix & Callow (1943, 1951) on *Salm. paratyphi B*, phage-typing has proved an important tool in tracing the path of these infections; and the recent work of Callow (1959) has provided a detailed scheme for the phage-typing of *Salm. typhi-murium*. Hitherto it has been found for all practical purposes that each excreter produces one phage-type only. We had isolated organisms of three and finally four, apparently distinct and unrelated phage-types from the river water and expected to find a different human source for each of these types. It was only by a long process of elimination that we were driven to consider Mrs X as a possible carrier of multiple types, and finally succeeded in incriminating her. Our findings show that in searching for the source of cases or contamination due to different phage-types of *Salm. paratyphi B* it may save much time and trouble to remember that a carrier can be an active excreter of more than one type, and to take steps to exclude this possibility without delay.

#### SUMMARY

Polluted water from a stream caused three cases of infection with *Salm. paratyphi B* belonging to two phage-types. These phage-types and two others were isolated from the water.

One woman, aged 81, with a history of enteric fever 20 years previously was discovered to be a prolific carrier of at least three of these phage-types, and to be the source of the pollution.

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