THE INCIDENCE OF ORGANISMS OF THE SALMONELLA GROUP IN WILD RATS AND MICE IN LIVERPOOL

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In a few instances outbreaks of food poisoning have been traced with a high degree of probability to rodents (Savage & White, 1925; Salthe & Krumwiede, 1924; Jones & Wright, 1936), but in the main the idea that such outbreaks may be due to contamination of food with the excreta of rats and mice is based on the fact that organisms of the Salmonella group have frequently been found in the faeces or organs of these animals. Savage & Read (1913–14) made cultures from the spleen, liver, heart blood and intestinal contents of forty-one rats after enrichment in liquid media and isolated five strains of *Bacterium enteritidis* (Gaertner). Savage (1918) obtained four strains of what he regarded as "para-Gaertner" which failed to ferment dulcitol and one strain of Gaertner's bacillus from forty-eight rats by direct inoculation from the spleen and heart blood on to MacConkey's medium. Savage & White (1922) from the spleen, liver, heart blood and intestinal contents of ninety-six rats recovered six strains of Gaertner's bacillus. Kerrin (1928) isolated eleven strains of *Bact. enteritidis* (Gaertner) from 100 rats. His observations were made on the liver and spleen with preliminary enrichment in brilliant green peptone water and subsequent plating on MacConkey's medium. All these British observations agreed in indicating that Gaertner's bacillus was not uncommonly found in wild rats, but in a much more extended series reported from America by Meyer & Matsumura (1927) twenty-eight strains of *Bact. enteritidis* (Gaertner) and thirty of *Bact. typhi murium* (*Bact. aertrycke*) were isolated from 775 rats. These workers inoculated large portions of liver, spleen and colon into broth containing brilliant green and gentian violet and subcultured on brilliant green eosin agar. Verder (1927), who examined 114 rats in another part of America, records the isolation of five strains of Gaertner's bacillus and one of *Bact. aertrycke.*

Present investigation

The present investigation was undertaken in order to determine whether a more extended series of observations might not reveal the presence of members of the Salmonella group which had not hitherto been found in rats in Great Britain, and also whether the incidence of these organisms showed any seasonal variations.

The rats and mice were taken from those which were sent daily to the City

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Laboratories, Liverpool, for routine examination for plague. About 70% of them were of the large brown variety, probably *Rattus norvegicus*, and 30% of the small black variety, probably *R. rattus*. Most of the brown rats were trapped in the city, while the black ones came from the port and docks. About 10% of the rats showed macroscopic lesions in liver or spleen or both, but no special correlation was noticed between these lesions and proved Salmonella infections. A piece of liver, half the spleen and a piece of gut (mostly ileum) with its contents were removed for examination. Sterile instruments were used for opening each animal and a separate set for cutting the organs. One set of instruments was used for the three organs, and so the probability of accidentally carrying the infection from one organ to the other could not be excluded. Work commenced with the liver and ended with the intestine. The parts were removed directly to tubes of tetrathionate broth, incubated at 37° C. for 18 hr. and then plated on brilliant green eosin agar (Jones, 1936). The plates were incubated at 37° C. for 24 hr. and suspicious colonies inoculated into tubes of ordinary broth for further investigation. All media were frequently controlled throughout the work with known strains of *B. coli* and of the Salmonella group.

All the strains isolated were examined morphologically and biologically in the usual manner to see that they fell properly into the Salmonella group, but the final identification was made on the result of agglutination tests which were in the main done with Oxford standard agglutinating sera. For the confirmation of identification in certain cases and for the identification of the Thompson strain, I am indebted to Dr W. M. Scott of the Ministry of Health Laboratory. Most strains were readily identifiable on isolation, but in some cases repeated subculture was necessary to bring up the H agglutination or dispose of a degree of auto-agglutinability exhibited by the original culture.

The investigation extended over a period of about nine months, from January to September 1936. Altogether 750 rats were examined, from fifty-five of which eighty-nine strains of organisms were isolated which belonged to the Salmonella group. These were identified as follows (Table I).

Table I. *Organisms of Salmonella group isolated from* 750 *rats*

Organism	Strains	Rats positive	% rats positive
Bact. enteritidis (Gaertner)	45	24	3·2
Bact. aertrycke	40	27	3·6
Bact. newport	3	3	0·4
Bact. thompson	1	1	0·13
Total	89	55	7·33

The general results are very similar to those reported from California by Meyer & Matsumura, whose technique was very like that adopted in this investigation. They obtained fifty-eight positive results from 775 rats (7·5%), which agrees very closely with my figures of fifty-five positive results from 750 rats (7·3%). Furthermore, Meyer & Matsumura isolated *Bact. enteritidis*

(twenty-eight strains) and *Bact. aertrycke* (thirty strains) in about equal numbers, a finding which is again very close to the numbers recorded in Table I (*Bact. enteritidis* 24, *Bact. aertrycke* 27). They too, unlike other workers, were able to isolate these organisms from the intestine in nine cases. The source in the present series is indicated in Table II.

Table II. *Sources of organisms isolated from rats*

	Isolated from						
Organism	L., S. and I.	L. and S.	L. and I.	S. and I.	L. only	S. only	I. only
Bact. enteritidis (Gaertner)	5	8	1	1	4	3	2
Bact. aertrycke	4	4	.	1	11	6	1
Bact. newport	.	.	.	.	1	1	1
Bact. thompson	.	.	.	.	.	.	1

L., S., I. = liver, spleen and intestine respectively.

It will be seen that while liver and spleen provided the majority of the strains the organisms were isolated from the intestinal contents only in five cases, and from these as well as from another organ in twelve instances. We attribute the success with the intestinal contents to the use of tetrathionate broth and brilliant green eosin agar. It should be noted that from one rat *Bact. aertrycke* was isolated from the spleen and *Bact. enteritidis* from the intestine. This was the only instance of double infection which was encountered.

The finding of *Bact. newport* (three times) and *Bact. thompson* (once) in rats appears to be a new observation. According to Topley & Wilson (1936) the former has previously been isolated from a dog, whereas the latter appears not to have been recorded from animal sources other than man. It may be of interest to record that *Bact. thompson* has been twice isolated from patients in Liverpool in the past two years.

Jones & Wright were able to isolate *Bact. aertrycke* from a mouse, and from mouse faeces in dried milk, in the course of their study of a case of food poisoning. I have examined 125 mice in the same way as recorded for rats but have been unable to isolate from them any organisms of the Salmonella group. These animals were all examined in the summer of 1936, and in the light of what is recorded below this fact may have considerably affected the findings.

The rats examined in this study were obtained from widely scattered sources in the city and port. There seemed to be no clear evidence of any definite localization of the infection in particular areas. Unfortunately the incidence in the different species of rats was not adequately investigated.

SEASONAL INCIDENCE OF SALMONELLA INFECTION IN RATS

Little attention has previously been paid to the question of seasonal incidence of this type of infection in rats. This investigation spread over a period of nine months, from January to September 1936. In each three-month period 250 rats were examined and the results have differed very much in the various periods (Table III).

Table III. *Seasonal incidence of Salmonella infections in rats*

	Jan. to March	April to June	July to Sept.
Animals examined	250	250	250
Animals infected	44	10	1
% infected	17·6	4	0·4
Infected with *Bact. enteritidis*	16	7	1
,, *Bact. aertrycke*	26	1	0
,, *Bact. newport*	2	1	0
,, *Bact. thompson*	1	0	0

It is evident that the distribution throughout the year was extremely irregular, and that examination of a smaller or less widely dispersed sample might have given very different results. As mentioned above, the examinations of mice were all done in the July to September period, and the results may well be just as misleading as would those of the rats for the same period as an indication of the general prevalence of such infections. We have no evidence on which to base an explanation of this distribution of infections. It may, indeed, be mere chance and represent the findings for this particular sample of rats in this particular year. It is, however, possible that it may be an expression of factors connected with the rat population which affect the dissemination of these infections. In any case it does not accord with the seasonal distribution of cases of food poisoning in man.

Conclusions

1. Organisms of the Salmonella group were isolated from fifty-five out of 750 wild rats trapped in Liverpool (7·3%).

2. The organisms isolated were *Bact. enteritidis* (Gaertner) (24 rats), *Bact. aertrycke* (27), *Bact. newport* (3), and *Bact. thompson* (1).

3. The incidence of these infections was much higher in the winter months (17·6%) than in the spring (4%) or summer (0·4%).

4. No organisms of the group were isolated from 125 mice examined in the summer period (July to September).

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