

SPORADIC INFECTIONS IN ABERDEEN DUE TO FOOD-POISONING ORGANISMS OF THE SALMONELLA GROUP

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THE elucidation of the antigenic structure and consequent classification of the various members of the Salmonella group has given a fresh impetus to the study of infections caused by these organisms. Their serological classification has been intensively examined, particularly by White (1926) in this country and by Kauffmann (1929, 1930) in Germany. Lovell (1932 *a, b*) has correlated the findings of the various workers and has summarised the existing knowledge on the subject. At present the Salmonella group includes some thirty-five members, certain of which cause disease in man only, others, so far as is known, in animals only, and many in both man and animals.

SCOPE OF WORK.

During the four-year period 1929–32 an intensive study of all cases of Salmonella infections has been undertaken. In the present paper the results of this inquiry are presented, but infections due to *S. typhosus* and *S. paratyphosus* B are not included. The discovery of many of these cases was, to some extent, accidental, since a diagnosis of bacillary dysentery had been suggested by the symptoms of the illness. Again, certain cases of continued fever have been found to be undoubtedly due to members of the paratyphoid group other than *S. paratyphosus* B and only by a complete analysis of the antigenic structure could the causative organism be completely identified.

METHODS.

Through the good offices of Dr Wm Scott of the Pathological Laboratory of the Ministry of Health various type cultures of organisms of the Salmonella group were obtained and also a supply of various sera. From these cultures and others obtained from the National Collection agglutinating sera were prepared for the "H" antigens of the smooth monophasic types and of the specific and group phases of smooth diphasic types. Furthermore, "O" antigens were prepared and, from rabbits immunised with these, "O" sera were obtained. The rabbits were immunised with "H" antigens until trial bleedings showed the sera to have titres ranging from 1/12,800 to 1/51,200 or more. Rabbits immunised with "O" antigens did not respond so readily as those immunised with "H" antigens and accordingly were killed when

the serum obtained was found to have a titre of 1/3200 to 1/6400. The animals were then anaesthetised and fully bled, the sera after separation being preserved with 50 per cent. pure glycerine.

For routine diagnosis all samples of faeces, urine and vomit were plated on McConkey's bile salt neutral red lactose agar. Non-lactose fermenting colonies were picked into tubes of medium containing lactose, glucose, mannitol, dulcitol, broth and semi-solid agar. If, after an incubation period of 24 hours, the morphological, staining, and sugar reactions were correct and if no indole was produced then the organism was subjected to various agglutination tests. For the purpose of preliminary tests one polyvalent serum containing equal quantities of the sera prepared for the monophasic types (with the exception of *S. typhosus* and European *S. suispestifer*), a second containing equal parts of all sera prepared for specific diphasic types, a third for the European *suispestifer* type to cover all group phases, and a fourth for the specific phase of *S. paratyphosus* B were employed. If agglutination was obtained with any of these sera further steps were then taken to identify the organism.

Agglutination tests were next carried out with "O" agglutinating sera, and when agglutination occurred the serum or sera producing agglutination were absorbed with the strain under observation and thereafter tested for agglutination against the homologous antigen or antigens. This procedure enabled one to limit the "H" agglutination and absorption tests to a *Salmonella* sub-group with a particular somatic antigen. Thereafter, agglutination tests were carried out with the "H" sera of the somatic group employing, in the case of the diphasic types, an emulsion of the specific phase of the organism. Absorption tests were similarly carried out with an emulsion in the specific phase. In the case of the Thompson type the procedure described by Scott (1926) was employed to convert the group to the specific phase of the organism before proceeding with further agglutination and absorption tests. Furthermore, in order to identify more completely the monophasic group European *suispestifer* strains, agglutinating sera were prepared and reciprocal absorption tests carried out.

In addition to the serological tests the biochemical reactions of the various strains in media containing lactose, glucose, mannitol, maltose, dulcitol, xylose, inositol, arabinose, rhamnose, lead acetate agar, and litmus milk were examined.

RESULTS.

Incidence of the various Salmonella types.

The results of the classification of the various strains are presented in Table I, and are compared with the findings by Savage (1932) for England, by Seligmann and Clauberg (1932) and Boecker and Silberstein (1932) for Germany. Savage in his broad review of food poisoning points out that the investigation of the organisms from 121 outbreaks in England during the

period 1919-31 was mainly carried out by Dr W. Scott at the Ministry of Health Pathological Laboratory, and somewhat similar findings are given by Newman (1932). During the four-year period in Aberdeen only 46 cases of infection were encountered, but these 46 cases were distributed over 30 separate outbreaks. Five types of *Salmonella* organisms were encountered; the majority of the infections (20) were due to *S. aertrycke*, next in frequency came infections—17—due to *S. thompson*, thirdly, six infections due to *S. dublin*, and finally, only single cases of infection due to *S. suipestifer* European and American types. In one instance it was not possible to identify completely a strain since the organism recurred in the group phase; its "O" antigen was identical with the "O" antigen of *S. paratyphosus* C sub-group, and it is probable that it was a *thompson* type in the group form.

Table I.

Group	Type	Savage outbreaks 1919-31	Seligmann and Clauberg cases	Boecker and Silberstein cases 1928-32	Aberdeen outbreaks: cases 1929-32	
A	Senftenberg-Newcastle	2	—	2	—	—
B	Aertrycke-Breslau	76	89	64	18	20
	Stanley	1	—	7	—	—
	Reading	—	—	3	—	—
C	Derby	2	—	—	—	—
	Suipestifer-American }	7	—	1	1	1
	Suipestifer-European }				1	1
	Thompson-Berlin	7	3	8	5	17
	Oranienburg	—	—	1	—	—
Potsdam	—	2	—	—	—	
D	Newport	5	5	7	—	—
	Morbificans	3	—	1	—	—
E	Enteritidis-Gaertner Jena	14	28	15	—	—
	Dublin-Gaertner Kiel	1		6	4	6
F	<i>Salmonella</i> not fully typed	3	—	—	1	1
		121	143	115	30	46

The biochemical reactions of the various strains isolated.

The results of these biochemical reactions after an incubation period of 7 days are given in Table II and generally conform to previous descriptions. Five *S. aertrycke* strains failed to produce any change in media containing inosite and two in media containing rhamnose, while two only produced acid in this carbohydrate. The Thompson type strains produced acid and gas in all types of carbohydrate media used. Four strains of *S. dublin* produced acid and gas in all sugars with the exception of inosite and arabinose. The *suipestifer* strains failed to ferment dulcitate and inosite and the strain of the American type failed to produce blackening of lead acetate agar.

Clinical and epidemiological findings.

Most of the cases were, apparently, clinically typical cases of enteritis of the type commonly associated with food poisoning, but some were really dysenteric in character. By blood culture in broth or by culture of blood clot in bile it was possible to show that one infection due to *S. aertrycke*, two to

S. dublin, and both the cases infected with the American and European types of *S. suispestifer* were suffering from septicaemia. One case infected with *S. dublin* developed meningitis due to this organism. The single case of septicaemia due to *S. aertrycke* and two cases of septicaemia due to *S. dublin* died, while both cases infected with the *suispestifer* types recovered. Seven of

Table II.

N.B. All strains included in this table failed to ferment lactose, produced acid and gas in glucose, mannite and maltose, and an acid to alkaline reaction in litmus milk.

Strain from case	Type of Salmonella	Dulcitate	Xylose	Inosite	Arabinose	Rhamnose	H ₂ S
5	<i>S. aertrycke</i>	AG	AG	AG	AG	AG	+
6		AG	AG	AG	AG	AG	+
8		AG	AG	AG	AG	AG	+
9		AG	AG	AG	AG	AG	+
10		AG	AG	AG	AG	AG	+
11		AG	AG	AG	AG	AG	+
12		AG	AG	AG	AG	AG	+
13		AG	AG	AG	AG	AG	+
14		AG	AG	AG	AG	AG	+
15		AG	AG	AG	AG	AG	+
16		AG	AG	AG	AG	AG	+
31		AG	AG	AG	AG	AG	+
35		AG	AG	0	AG	0	+
36		AG	AG	AG	AG	AG	+
37		AG	0	AG	AG	AG	+
41		AG	AG	0	AG	AG	+
42	AG	AG	0	AG	AG	+	
44	AG	AG	0	AG	AG	+	
45	AG	AG	0	AG	0	+	
46	AG	AG	0	AG	AG	+	
4	<i>S. thompson</i>	AG	AG	AG	AG	AG	+
17		AG	AG	AG	AG	AG	+
18		AG	AG	AG	AG	AG	+
19		AG	AG	AG	AG	AG	+
20		AG	AG	AG	AG	AG	+
21		AG	AG	AG	AG	AG	+
23		AG	AG	AG	AG	AG	+
26		AG	AG	AG	AG	AG	+
27		AG	AG	AG	AG	AG	+
40		AG	AG	AG	AG	AG	+
1	<i>S. dublin</i>	AG	AG	0	0	AG	+
2		AG	AG	0	0	AG	+
3		AG	AG	0	0	AG	+
32		AG	AG	0	0	AG	+
38	<i>S. suispestifer</i> (American)	0	AG	0	AG	AG	0
39	<i>S. suispestifer</i> (European)	0	AG	0	AG	AG	+
43	Not fully identified	AG	AG	AG	AG	AG	+

the cases occurred in children in the age group 5 years and under, nine of the cases in the age group 6–20 years, 20 of the cases in the age group 21–40 years, and 10 in the age group 41 and over. The distribution as regards sex was 20 female cases and 26 male cases. In certain cases the clinical and epidemiological findings are of considerable interest and an account of these follows.

Case 1. I.L., a boy aged 5 years, had been ill at home with an undiagnosed pyrexial condition for 3 weeks. On February 12th, 1929, he was seen by an aural surgeon on account of symptoms suggesting a mastoiditis. On examination it was found that he had a profuse bilateral otorrhoea and that there was marked oedema over the right mastoid process. There was an indefinite history of ear trouble for years. The patient was removed to a nursing home, and a cortical mastoid operation was immediately performed, pus being found throughout the mastoid cells. There was some improvement in the general condition following the operation; the temperature on the following morning was normal, but in the evening it rose to 99·5° F. and the pulse rate increased proportionately. The patient's temperature continued to show morning remissions. On the 18th the right lateral sinus was explored and found free from clot. A left cortical mastoid operation was performed and a small amount of pus found in the antrum and upper cells. The condition did not respond either to serum treatment or to the second operation. The temperature and pulse rate increased and rigors became frequent, and the general condition of the patient deteriorated. On March 8th slight jaundice was evident, and during the following day the patient became delirious and died on the morning of March 10th. A blood culture made on February 19th showed a Gram-negative, non-lactose fermenting, motile bacillus; and a sample of faeces obtained 2 days later showed a similar organism, both strains being identified later as *Salmonella* type *dublin*. Cultures made from pus obtained from the ear showed *Streptococcus haemolyticus* and staphylococci.

It appears evident, therefore, that this patient had had a septicaemia for some weeks, and as a result of the lowering of the patient's resistance an acute exacerbation of the chronic infection of the middle ears had occurred.

Case 2. N.M., a female infant aged 11 weeks, was admitted to hospital suffering from vague digestive upset. On admission the temperature was normal, but on the following day it rose to 102·2° F., and 3 days later was again normal. During the initial stages of the illness the child vomited frequently, and although there was no actual diarrhoea the stools were green. Clinical examination showed no signs of any definite lesion in chest, abdomen, or nervous system. After the subsidence of the fever the convalescence was uneventful. On May 22nd a small amount of blood was obtained for blood culture, but this specimen remained sterile. On the same day the faeces and urine were examined, and a *Salmonella*-like organism was obtained from the urine only. On May 27th the blood, urine and faeces were examined and the same organism was obtained from the urine. Finally, the faeces and urine were examined for the third time on June 3rd, but no pathogenic types were obtained. The organism obtained from the urine was later identified as a *dublin* type.

Case 3. F.D., a female child, had been born in hospital on February 21st, 1929. The infant progressed favourably until June 6th when there was a slight rise in temperature and it was noticed that she had developed a hard dry

cough, but examination of the chest gave no indication of any intra-pulmonary lesions. On June 8th the temperature again reached 99° and on the following day rose to 102·5° F., and on this day examination of chest showed dullness on percussion over the middle of left lung posteriorly and auscultation showed a raising of pitch of the breath sounds. On the following day the chest signs showed no further change but the child now showed obvious signs of meningitis, there being definite head retraction, bulging of the fontanelle, a positive Kernig, and an internal squint of the left eye. The clinical condition of the infant became rapidly worse and she died on June 11th.

For bacteriological examination the only specimen obtained from this case was the cerebro-spinal fluid taken on June 9th. The specimen was turbid and a qualitative chemical examination showed a marked increase above normal of globulin and a decrease in the sugar content. Microscopic examination showed 540 cells per c.mm., 68 per cent. of the cells being polymorphonuclear leucocytes and 32 per cent. lymphocytes. Microscopic examination also showed numerous Gram-negative bacilli. Cultural examination showed numerous colonies of a Gram-negative bacillus which was found to be a non-lactose motile Salmonella-like organism, later identified as type *dublin*.

Case 5. From F.M., a male patient aged 11 years who had been ill for 5 weeks, a blood specimen was received with a request for a Widal reaction. The patient became ill with diarrhoea and vomiting, and this was followed 2 days later by fever and general malaise. He was admitted to hospital 1 week after the onset, when his temperature was evidently of that type usually associated with a septicaemia, rising every evening to 103° F. or thereabout. The boy complained of pain in the larger joints; the tongue was moist and clean, the abdomen was distended, the spleen was enlarged and he had marked diarrhoea. The fever continued, the patient showed marked wasting, and in the fifth week of the illness developed generalised lymphadenitis and died.

The patient's serum failed to agglutinate *S. typhosus* "H" and "O," *S. paratyphosus* A "H," and *S. paratyphosus* B "O," but agglutinated *S. paratyphosus* B "H" (non-specific) to a dilution of 1 in 6400. In addition the blood clot was cultured in bile and an organism giving typical cultural and biochemical reactions of the paratyphoid group was obtained, and this organism agglutinated to the full titre of a non-specific paratyphoid B serum. Later, absorption tests were carried out, using a specific paratyphoid B serum, with the result it was found that the organism was not a strain of *S. paratyphosus* B and further serological investigation showed it to be a *S. aertrycke* strain.

Cases 8 and 9. Two elderly sisters, age 48 and 50, kept a croft and attended to their cow. The cow took ill with an acute enteritis and was attended by M.M. As the cow appeared to be dying it was slaughtered and the carcass was seized by the Veterinary Inspector. Another cow was bought and arrived at the farm after the first cow had been disposed of and was placed in another

byre. In the meantime, the sister who had been attending the first cow took ill, then the second cow developed an acute enteritis but recovered. The second sister who had been attending the ill cow now developed an acute enteritis. From the faeces of both sisters and from the milk and faeces of the second cow *S. aertrycke* was isolated. Presumably, therefore, the first cow was also the victim of a Salmonella infection.

Cases 18-30. This outbreak is the largest in the present series and involved two households, 13 individuals in all. Mrs D. purchased pressed beef (brisket) from a provision merchant. The meat was placed in the domestic refrigerator until lunch time the following day. Eight out of 11 members of the household partook of the meat, and after an incubation period of 22 hours one male member of the family took ill and other seven became ill within the next 8 hours. The three individuals who did not partake of this particular beef remained well.

In the second household Mrs G. purchased the pressed beef from the same merchant 2 hours later than Mrs D. The beef in this instance was retained for 8 p.m. supper and was partaken of by three members of the family and two friends, while three individuals did not have it. In this instance a boy of 13 years became ill in 12 hours, all the individuals partaking of the beef had become ill within 22 hours.

Four out of five specimens of faeces from the first household and two out of four from the second showed numerous colonies of a non-lactose fermenting organism, later proved to be *S. thompson*. Unfortunately, when inquiry was made at the shop, none of the beef was available. The meat had been cooked on the premises, stored in a refrigerator, and sold.

Cases 32, 33 and 34 are of interest in that all were infected with *S. dublin*. The son, a young man of 22 employed in the post-office, took ill with abdominal pain, vomiting, diarrhoea, and fever. He was attended during his illness by his elderly parents, both of whom developed a mild gastro-enteritis 2 days after the son took ill. Presumably, therefore, they were infected by the son. No possible source of the son's infection could be discovered.

Case 38. Mrs F., aged 19, was confined in a hospital. The labour was entirely normal, but on the day after confinement she had a rigor. The fever continued without sickness, abdominal pain, or diarrhoea, and she was admitted to the City Hospital as a case of puerperal fever. On admission her temperature was 102.5° F., she had no definite symptoms of uterine infection, and nothing definite intra-abdominally. From the blood and from an intra-uterine swab a Salmonella organism was cultured and this was later found to be the American *suipestifer* type. The faeces and urine were negative. Three days after admission the fever subsided, but 4 days later there occurred a slight recurrence lasting 6 days, when recovery took place. Here again no possible source of infection could be discovered as all the patients in the ward from which she was removed remained perfectly well and there was no history of illness amongst the nursing staff.

Case 39. A female, married, aged 28 years, took ill with abdominal pain, vomiting, and slight diarrhoea. The patient was in her fifth month of pregnancy and on examination it was found she was going to abort. After a complete abortion she continued to run a slight fever and the doctor in attendance sent a swab of the uterine discharge for examination. From this material a *Salmonella* organism, later found to be of the European *suipestifer* type, was cultured. The patient presumably had had an intestinal and blood infection, the onset of the illness producing the abortion. Inquiry failed to show that any of the other members of the family had been ill.

As each *Salmonella* infection was recognised by the bacteriological investigation it was personally investigated. Thus, in the series of 46 cases, no less than 26 occurred as single cases unassociated with others. In these instances only a single member of a family or group of individuals was affected, in two instances there were two members of a family infected, and in one instance three members, two being really secondary infections from the primary case. Finally, in one instance, members of two families were infected by a common food substance. Thirty-six of the cases occurred in the months of August, September and October of the respective years.

In previous papers Kinloch, Smith and Taylor (1926) and Bowie, Kinloch and Smith (1926) outbreaks of *Salmonella* food poisoning in Aberdeen have been described. In these large outbreaks little difficulty was found in tracing the source of infection through a common food substance. In this series, however, it has only been possible, despite personal investigation, to fix definitely the source of infection in outbreak No. 8—two cases—due to a cow infected with *S. aertrycke* and in outbreak No. 16—13 cases—resulting from eating pressed beef infected with *S. thompson*. Scott (1930) has definitely shown that duck eggs are also a possible source of infection due to *S. aertrycke*. Inquiry in the present series failed to produce any evidence that this source might account for some of the infections. It can only be suggested that the individual cases of infection, particularly with the *aertrycke* type, must have occurred through the ingestion of material contaminated with the excreta of infected rodents. Again, no evidence has ever been found that the human carrier plays an active part in the dissemination of these infections, since the routine examination of thousands of specimens of faeces has failed to show the presence of *Salmonella* (food poisoning) organisms except when obtained from actual cases.

SUMMARY.

An investigation has been made into sporadic Salmonella infections. The most prevalent infecting organisms have been found to be Salmonella types *aertrycke* and *thompson*. Infections due to *S. dublin*, *S. suispestifer* (both European and American types) have also been encountered. The striking features of the investigation have been the number of individual cases and the lack of definite evidence of the source of infection.

The author is indebted to the Medical Research Council for a personal grant.

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(MS. received for publication 3. II. 1933.—Ed.)