FATAL EPIDEMIC ENTERITIS DUE TO *B. DYSENTERIAE* SONNE.

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DURING an enquiry into the bacteriology of summer diarrhoea in infants there was encountered in a certain hospital in Glasgow a small outbreak of acute enteritis which on investigation was found to be due to infection by *B. dysenteriae* Sonne. Although isolated cases of Sonne dysentery have been discovered in Glasgow during the past few years this is the first recorded outbreak in the city of epidemic enteritis due to this organism. A remarkable feature of the outbreak was the high fatality rate; in one ward there were seven deaths, all of which were probably due to infection with the Sonne bacillus. The outbreak appeared to originate in the nursery of the institution, and after the first case of this severe type of diarrhoea had been removed to ward X the infection spread in that ward and involved infants who were recovering from minor medical ailments with which they had been admitted. Two additional cases occurred subsequently in the nursery and were removed to ward X.

It is impossible to dogmatise as to the source of the infection as the bacteriological examination did not extend to the nursery attendants. The first case occurred in the nursery and may have been due to infection from a carrier member of the nursery staff. In Table I the cases have been enumerated in the order in which they developed diarrhoea: it will be noted that Cases I, III and VII had diarrhoea when admitted from the nursery. There was no fatal case of acute enteritis in ward X until after the admission of Case I which was proved subsequently to be suffering from Sonne dysentery.

Table I. *Fatal cases of diarrhoea in ward X during the Autumn, 1928.*

Case	Age in months	Removed from	Date of admission to ward X	Diarrhoea developed	<i>B. dysenteriae</i> Sonne	Date of death	Day of disease when examined
I	10	Nursery	Aug. 26	Aug. 26	+	Sept. 19	10
II	3	District	" 25	Sept. 1	-	" 14	6
III	12	Nursery	Sept. 3	" 3	.	" 12	—
IV	3	District	Aug. 22	" 5	-	" 29	9 & 21
V	3	"	" 28	" 5	-	" 14	7
VI	22	"	Sept. 18	" 30	+	Oct. 5	3
VII	—	Nursery	" 30	" 30	.	" 5	—

The milk supply of the nursery, ward X and ward Y—another infant ward—was the same, viz. Grade A T.T. milk, and no case arose *de novo* in ward Y. Six milk samples from the nursery were bacteriologically examined and no non-lactose fermenting organisms were isolated. During the epidemic four batches of flies were caught in ward X and examined for *B. dysenteriae* Sonne with negative result.

Although *B. dysenteriae* Sonne was isolated in only two instances, it is probable that all the cases recorded in Table I died from Sonne dysentery; they each presented the same clinical picture (see clinical notes), and it is possible that the bacteriological examination was carried out too late to recover *B. dysenteriae* Sonne from the stools in the three negative instances. Wiseman (1927), however, has isolated the bacillus from the faeces as late as the thirteenth day of disease.

CLINICAL NOTES OF CASES I AND VI.

Case I. Male child, aged 10 months, was admitted from the nursery to ward X on 26. viii. 1928 with the diagnosis of broncho-pneumonia and enteritis. The child was very cross: temperature 101.4° F., pulse-rate 107 and respiration rate 52. The temperature fell, but rose on the fifth day to 101° and continued intermittent for the next five days; it then became subnormal and remained so until death occurred on 19. ix. 1928. During the first week the chest condition attracted more attention than the diarrhoea but cleared up on 5. ix. 1928. The motions were very frequent, foul-smelling, grass-green and mixed with mucus: blood was never detected by the naked eye. There is no record of vomiting. Prostration and pronounced wasting were later features of the disease.

Case VI. Male child, aged 1 year and 10 months, the salient features of whose case are given chronologically in order to demonstrate that Sonne dysentery was contracted in ward X.

1928. Sept. 18. Admitted from district to ward X, with the complaint of vomiting after feeds: temperature normal: child was well nourished. (Vomiting ceased on admission; child noted to be constipated.)
- Sept. 23. Very listless, has an irritating cough; motions normal, temperature 97° F.
- Sept. 24. Vomited: appears in pain.
- Sept. 26. Child appears ill, motions dark and slimy.
- Oct. 1. Diarrhoea continues; motions green, foul-smelling and slimy. Intractable vomiting commenced.
- Oct. 5. Death occurred.

Note. The temperature was never recorded above 97° F., but as it was taken in the morning only there may have been an evening rise.

Remarks on Cases I and VI.

The type of diarrhoea presented by these two cases was that of the choleraic form of enteritis. In the animal inoculations also (*vide infra*) the lesions produced were invariably in the small intestine. This corresponds with the experimental findings of Mackie (1919) with atypical dysentery bacilli, of which one of the strains gave the biochemical reactions of *B. dysenteriae* Sonne. The bronchitis noted in the earlier stages of these two cases, transient in Case VI, but lasting eleven days in Case I, has not been found uniformly by other observers. Nevertheless, Fraser, Kinloch and Smith (1926) state that "involvement of the respiratory system in a catarrhal process is a notable feature of Sonne dysentery." The most noteworthy feature in these cases is that they each terminated fatally. Mita (1921) discussing his fatal cases which were all of a fulminant type refers the high mortality rate to a con-

stitutional disposition of the patient rather than to the toxic action of bacteria: yet there were no deaths among 24 cases of Sonne dysentery which occurred in the marasmus ward of the City Hospital, Aberdeen. There appears, therefore, to have been considerable exaltation in the virulence of the organism in this small epidemic which may have been favoured by the season and by host-susceptibility.

BACTERIOLOGICAL EXAMINATION.

A rectal swab taken from Case I was stroked on MacConkey's bile-salt-lactose-agar and gave an almost pure culture of *B. dysenteriae* Sonne accompanied by scanty colonies of *B. coli*, *B. mucosus capsulatus* and enterococci. A sample of faeces obtained from Case VI was emulsified in saline and two loopfuls spread on MacConkey's medium; a few colonies of *B. dysenteriae* Sonne were found and many colonies of *B. mucosus capsulatus*. When first isolated the colonies of *B. dysenteriae* Sonne had the appearance originally described by Thjøtta (1919) and Mita; but this appearance was found to vary as the strain grew older and as lactose fermentation occurred correspondingly earlier (although, as Thjøtta pointed out, lactose was fermented earlier in fluid than in solid media). After two months' sub-culture the colonies on MacConkey's medium were large, irregular and smooth, with a red inner portion heaped up towards a pale central papilla and greyish more transparent margins whose edges were deeply crenated. The appearance of the colony varied also with different supplies of media, being on some not definitely pale and on others small, rugose and thickened. This variation may have been associated with surface dryness of the medium, as in one series of animal experiments, in which inoculations were made on plates of lactose-bile-salt-agar using Andrade's indicator instead of neutral red, very beautiful Sonne colonies appeared which after several days developed a central pink papilla.

BIOCHEMICAL REACTIONS. (See Table II.)

Acid was produced within 24 hours in glucose, mannite and maltose by both strains; lactose was not fermented by either strain when newly isolated, but after several months' sub-culture, acid production was noted in this sugar on the third day with one strain, and on the fifth day with the other strain, which also produced acid in saccharose on the sixth day (Table II). Dulcitate, xylose and sorbitol were not fermented. Neither of the strains produced indol nor liquefied gelatin. A slow alkaline reaction was observed in litmus milk with a preliminary acidity in old strains. The fact that these strains were xylose-negative and indol-negative is important in bearing out the findings of Kerrin (1928) who asserted that *B. dysenteriae* Sonne might in this way be differentiated from *B. coli anaerogenes*, an organism which it otherwise closely resembles in its biochemical reactions¹.

¹ Two strains of *B. coli anaerogenes* have been encountered which are also xylose-negative and sorbitol-negative, but they produced indol within 24 hours.

Epidemic Enteritis

Acid production in lactose varied during the animal experiments and fermentation of saccharose appeared in both strains. The most interesting results in this connection were given in one of the animal experiments in which a strain was used which produced acid in lactose on the third day. After intracardiac injection into a guinea-pig the animal died and the strains recovered from the heart, lung and the small intestine had lost the capacity to

Table II. *Biochemical reactions of two strains of B. dysenteriae Sonne.*

Date of testing	Glucose	Lactose	Saccharose	Dulcite	Mannite	Maltose	Xylose	Sorbite	Litmus milk. Days			Gelatin	Indol	Motility
									1	3	10			
Case I:														
10. ix. 28.	A.	-	-	-	A.	A.	-	-	.	.	Alk.	-	-	-
21. xii. 28.	A.	A.*	-	-	A.	A.	-	-	Acid	.	Alk.	-	-	-
Case VI:														
29. ix. 28.	A.	-	-	-	A.	A.	-	-	.	Alk.	Alk.	-	.	-
19. i. 29.	A.	A.†	A.‡	-	A.	A.	-	-	Acid	Alk.	Alk.	-	.	-
	* 3rd day.				† 5th day.				‡ 6th day.					

- in Indol column = non-production after 14 days. Sugars read after 10 days' incubation at 37° C.

ferment lactose, whereas strains from the large intestine, liver, spleen and peritoneal exudate produced acid in lactose on the fourth day. A comparable result was got from another guinea-pig in this experiment. This phenomenon was not observed in the first series of animal experiments where guinea-pigs were given living culture intra-peritoneally and where the organism probably reached the blood-stream via the portal circulation.

SEROLOGICAL REACTIONS. (See Table III.)

A supply of agglutinating serum for *B. dysenteriae* Sonne was obtained from the Standards Laboratory, University of Oxford. Two months after isolation each strain agglutinated up to a dilution of 1/120, the controls in saline being negative. Two additional strains from Case VI which otherwise

Table III.

Organism	Dilutions of serum					
	1/100	1/200	1/600	1/800	1/1600	1/3200
Case I	-	+	++	++	++	+
Case VI (a)	-	- +	++	++	++	+
„ (b)	-	-	-	-	-	-
„ (c)	-	-	-	-	-	-

resembled *B. dysenteriae* Sonne were not agglutinated with this serum. At the same time Dr J. C. Kerrin (Aberdeen) supplied an antiserum which he had prepared against his Matheson strain (titre 1/3000). Table III shows the results of agglutination experiments with this antiserum tested against the strains isolated from Cases I and VI. Controls were negative. Both strains showed zonal phenomenon.

One of the inagglutinable strains, Case VI (*b*), was subjected to an agglutinin absorption test with the Oxford serum and was found to absorb the specific agglutinins for *B. dysenteriae* Sonne. This inagglutinable strain differed from Case VI (*a*) strain in that when recently isolated it produced acid in lactose and saccharose within 48 hours. All strains were tested against a polyvalent Flexner serum obtained from the Standards Laboratory, Oxford, but were not agglutinated.

Nine months after isolation the original strain from Case VI was found to have developed "roughness" on agar slopes and on MacConkey's bile-salt-lactose-agar medium. This phenomenon was not observed in the inagglutinable strains from Case VI nor in the original strains from Case I. It developed, however, in a Case I strain derived from one of the animal inoculations. These rough strains agglutinated spontaneously in normal saline.

Sonne's researches proved the high specificity of serum agglutination with *B. dysenteriae* Type III, and it may be accepted as the result of the above serological reactions that the organism which caused this outbreak of fatal enteritis was *B. dysenteriae* Sonne (Type III).

EXPERIMENTS ON ANIMALS.

A series of animal experiments was undertaken to study the virulence of *B. dysenteriae* Sonne, and the nature of the lesions which it produced. In addition some feeding experiments were carried out. All the animal inoculations were made by Dr R. Cruickshank. The protocols of individual experiments are not given; only the deductions drawn from these experiments are stated.

In testing the virulence of *B. dysenteriae* Sonne guinea-pigs and rabbits were used. Intravenous and intraperitoneal injections of unkilld and killed cultures were given, substituting the intracardiac route for the intravenous in the case of guinea-pigs. Three strains were used, namely, those from Cases I and VI and Dr Kerrin's Matheson strain added for the sake of comparison.

Emulsions of these strains were standardised as in vaccine work in order that the dosage might be fairly accurate, thus eliminating the vagueness of the terms "agar slope" or "half agar slope" used by many observers. Owing to the fact that cultures tended to lose virulence after three to four months' growth *in vitro*, there was some difficulty in assessing the results. Virulence was only partially restored by passage. In general, however, guinea-pigs were more susceptible than rabbits; a dose of 1000 million organisms (unkilled) of the recently isolated strains was lethal to guinea-pigs, whereas double that dose produced only sickness in rabbits. Killed cultures were less toxic than unkilld, similar doses (1000 million) producing merely temporary sickness in guinea-pigs and mild enteritis in rabbits.

In one experiment, *killed* cultures (2000 million) injected intravenously into rabbits resulted in death and the organism was recovered from the small and large intestine, spleen, liver and heart's blood. The emulsion had been

heated for one hour at 60° C. prior to injection and cultures made from this heated emulsion remained sterile. As the rabbits receiving the killed cultures were kept in the same "run" as those receiving unkilld organisms, they presumably became infected from the latter and being rendered less resistant by the injection of killed culture, they succumbed to the infection.

Similar doses of living organisms were slightly less toxic by intraperitoneal injection than when injected intravenously. With regard to differences in the virulence of the strains, Case I strain was more toxic than Case VI strain; the Matheson strain, owing to its age, was found to have practically no effect. The fact that live cultures were more virulent than killed cultures suggested the presence of an exotoxin as well as an endotoxin. Accordingly Case I strain was grown for ten days in 100 c.c. bouillon which was then filtered through a Berkefeld filter; 5 c.c. of this filtrate injected intravenously into rabbits and guinea-pigs was without effect.

Autopsies were done on nineteen animals and bacteriological and histological examinations were made of the tissues and organs of these animals. The most noteworthy fact which emerged from the macroscopic examination was the uniformity of the enterotropic effect of the organism, the lesions being situated in the proximal half of the ileum in each case; the large intestine was not affected. The degree of inflammation varied with the toxicity of the strain used, and histological examination revealed in one animal microscopic ulceration of the mucosa of the small intestine. A generalised toxæmia, accompanied by areas of focal necrosis in the liver and kidneys, was found in the case of any animal which succumbed rapidly. When freshly isolated Case I strain had a particularly toxic effect on the suprarenals of guinea-pigs, the lesions being identical with that produced by *B. diphtheriae*. With the less toxic Case VI strain the animals survived two to three days after inoculation and lobular pneumonia was found in addition to the intestinal lesion. The organism could usually be recovered *post-mortem* from the small and large intestine, liver, spleen, lungs, heart's blood and peritoneal exudate.

For the feeding experiments rabbits, guinea-pigs and white mice were used. Previous observers (Bamforth, 1924, Kerrin 1928) have had entirely negative results under ordinary conditions of atmospheric temperature and humidity, but Arnold (1928) in his recent experiments on gastro-intestinal auto-disinfection found that increased susceptibility to enteritis was produced under such conditions as exposure to high temperatures and increased humidity. Various methods were accordingly tried to render the animals more susceptible: (a) Four guinea-pigs were kept in a room at 95° F. and emulsions of the live organisms were given in the drinking water. It was found that some sickness was produced and that the organism could be recovered from the faeces, while a control lot kept at the same temperature were unaffected. (b) An attempt was made to reduce the resistance of rabbits and guinea-pigs by injecting killed cultures of *B. dysenteriae* Sonne before feeding, but this experiment was negative owing to the fact that the strains had lost virulence.

Mice did not prove to be susceptible to feeding with *B. dysenteriae* Sonne under normal atmospheric conditions.

CONCLUSIONS FROM ANIMAL EXPERIMENTS.

There is a close resemblance between Sonne dysentery as it affected infants during the epidemic recorded and as it affected rabbits and guinea-pigs which had received inoculations of the organism. The assumption made clinically that *B. dysenteriae* Sonne caused lesions in the small rather than in the large intestine was corroborated by the foregoing experiments on animals. The resemblance was most striking in those animals which survived inoculation long enough to develop lobular pneumonia. From these experiments it is also inferred that the two strains isolated during the epidemic were highly virulent as relatively small doses were lethal to the animals used. The negative results of the feeding experiments are probably partly due to the fact that the strains had lost virulence and partly that the animals used were not susceptible to the action of *B. dysenteriae* Sonne when administered orally.

SUMMARY.

1. An outbreak is described of fatal epidemic enteritis due to *B. dysenteriae* Sonne which occurred in the autumn, 1928, in the infant wards of a hospital in Glasgow.
2. The clinical histories of two fatal cases are given.
3. The bacteriological and immunological examination of two strains of *B. dysenteriae* Sonne is described.
4. The results are given of a series of animal inoculations undertaken to study the virulence of *B. dysenteriae* Sonne and the nature of the lesions which it produces. To these are added the results of certain feeding experiments.

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