

REPORT ON AN OUTBREAK OF BACTERIAL
FOOD-POISONING¹.

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(With 1 Plan.)

OUTBREAKS of bacterial food-poisoning are fortunately of rare occurrence in Ireland. In spite of the attention directed to them in recent years and their apparent increasing frequency in other countries I have been only able to trace one such outbreak—that reported by McWeeney (1909) in 1908. It occurred in an Industrial School in the South of Ireland, 73 persons were affected, and the mortality was unusually high, nine deaths being recorded. The organism responsible for the outbreak was *B. enteritidis* (Gaertner)². Sporadic cases of bacterial food-poisoning appear to be similarly but rarely met with. One such case was recorded, again by McWeeney (1916), in which the causal organism, *B. Aertrycke*, was recovered from the organs after death. It is obvious that isolated cases, owing to the short duration of the illness and the rapid recovery of the patient, are easily overlooked. At any rate, these are the only instances of infection with *B. Aertrycke* or Gaertner which I have been able to trace in this country.

The outbreak which I had an opportunity of studying recently presents some unusual features. It occurred last Autumn in an Institution near Dublin, and the incidents of importance in connection with it, so far as I have been able to piece them together, may be briefly related. On Thursday, October 6th, 1921, it was noticed that one of the drains in the grounds of the institution was stopped. A brush was inserted at the man-hole, *M* (see plan) and the obstruction cleared away. A stoker employed at the engine-house stood at the door a few yards away watching the operations. On the following Saturday, about 10 p.m., this stoker was seized with nausea, vomiting, abdominal pain, and diarrhoea. The diarrhoea was fairly severe, and only ceased on Sunday evening after the administration of a lead and opium pill. On the following Friday night, October 14th, some boys in Block A became severely ill, about eight complaining up to midnight. The numbers rapidly and steadily increased during Saturday, and it was not till midnight that the attack had spent its

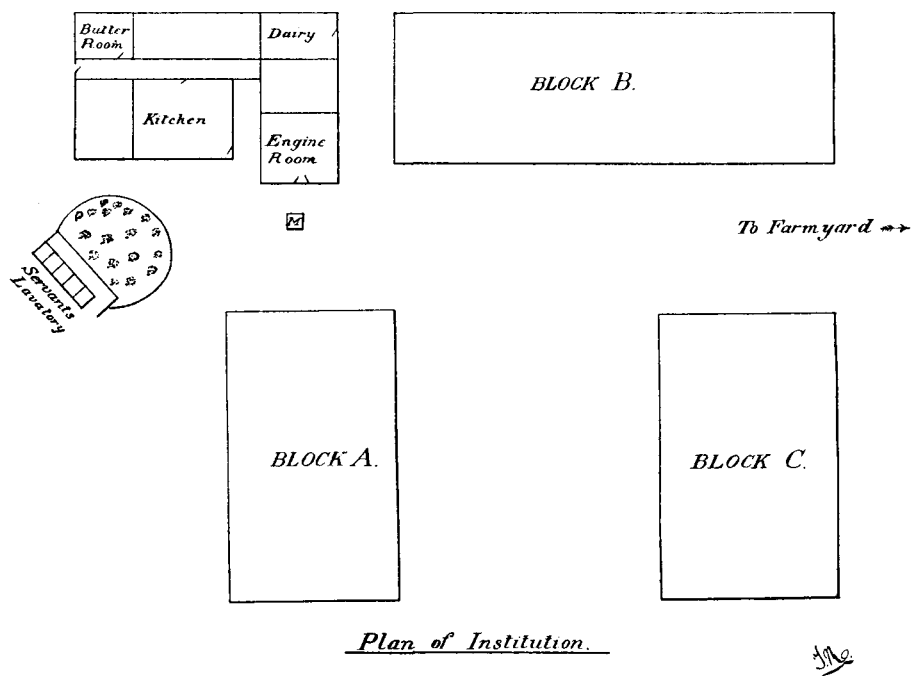
¹ This investigation was carried out under a grant from the Medical Research Council.

² This strain of Gaertner is No. 125 in the Catalogue of the National Collection of Type Cultures. *Med. Res. Council Spec. Rep. Ser.* No. 64.

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force. Four boys were attacked on Sunday. The number of boys stated to have been ill was 148 out of 170, all occupants of Block A, and partaking of the same meals daily. The symptoms were headache, nausea, vomiting, abdominal pain, diarrhoea, and a temperature reaching in some cases 104° F. The motions were liquid, and of a pea-soup appearance. Fortunately there was no fatal case. Most of the invalids were convalescent in four or five days, and in the milder cases in 24 hours. None of the occupants of Blocks B or C were affected, and no member of the staff became ill. Of the servants five were invalidated. Their case will be discussed subsequently.

On my arrival at the Institution on Sunday, October 16th, it was obvious that the outbreak was one of food-poisoning. The patients had all partaken



of the same food, and after an incubation period of 8 to 40 hours¹, had presented the symptoms of gastro-enteritis. The question then resolved itself into the determination of the following points:

1. The peccant food.
2. The causal organism.
3. The source of the infection.
4. The measures to be adopted to prevent the occurrence of similar outbreaks in the future.

¹ In four cases the incubation-period exceeded 40 hours. These may have been secondary infections.

I. THE PECCANT FOOD.

It was clear that the food conveying the infection was partaken of on Friday. On the previous day the occupants of Block A had partaken of exactly the same food as the boys in Blocks B and C. On the day of the outbreak the meals supplied in the affected Block were:

1. Breakfast: porridge and milk, followed by tea and bread and butter.
2. Luncheon: tea and bread and butter.
3. Dinner: lentil soup, fish (cod), vegetables, and stewed fruit.
4. Supper: cocoa and milk with bread and butter.

The only item in these four meals not common to the whole Institution was the porridge and milk which was only served on alternate mornings in Block A. I should at once make it clear that none of the food was available for investigation, any surplus food being sent each day to the farmyard to feed the pigs. The milk served with the porridge on Friday morning had been used on Thursday without ill-effect, and the general impression at the Institution was that in some way the fish served at dinner was responsible for the outbreak. The suspicions with regard to the fish were gone into thoroughly and proved to be groundless. It had arrived the previous day in two boxes which had been placed on the floor in a larder. On examination the next day it had seemed quite fresh. The contents of the two boxes were turned out on a table and those sufficiently large to supply a mess (10 boys) were wrapped separately in muslin and steamed. The steaming was carried out in one large steamer. The remainder of the fish was baked. The steamed fish was served to the boys in Blocks A and B. The sauce served at dinner was common to the whole institution. The possibility of a few of the fish having been infected was negatived by the extent of the outbreak. The boxes in which the fish had arrived were scraped and the scrapings examined bacteriologically, but no recognised food-poisoner was recovered. Again at least 13 boys who did not eat fish were attacked¹. Finally the first person affected complained at 3.45 p.m., $\frac{3}{4}$ of an hour after the commencement of dinner. Very short incubation-periods have been recorded in outbreaks of food-poisoning but in this instance in which the average incubation-period was approximately $25\frac{1}{2}$ hours, such an isolated case would have been very exceptional, the main incidence of the attack occurring 16–40 hours after breakfast (or 9–33 hours after dinner).

The milk-supply to the institution was next examined. The farm which adjoins the buildings supplies all the milk, meat, and vegetables used. The custom was to use the morning-delivery of milk at breakfast and luncheon. The amount required for porridge and for the servant's breakfast on the following morning was then set aside. (The servants took breakfast before the cows were milked.) The evening-supply was used at supper and for puddings on the following day. Any surplus milk was sent to the dairy to be churned. The milk set aside for porridge was stored in two large glazed

¹ *B. enteritidis* (Gaertner) was isolated from the faeces of one of these boys.

earthenware crocks on the floor of the butter-room. On the following morning the amount required for the servants' breakfast was drawn off, and the remainder poured into jugs for each mess. It was not added to the porridge in bulk. It is evident then that if this milk served at breakfast on Friday morning was the peccant food it must have become infected after luncheon on the preceding day as it had been used, as already stated, at breakfast and luncheon on Thursday and had not caused any illness. The available boys belonging to Block A were summoned to their Refectory and asked to sit in their usual places. It was found that in no case had all the occupants of any one mess escaped. Not more than two or three members of any mess were present, and most of the tables were empty. Some of the boys in attendance had suffered from a mild attack and were able to be about again. Others stated that they had suffered from headache but not from diarrhoea. Enquiries were next directed to eliciting complaints about the food. Some stated that the milk served with the porridge on the previous Friday was condensed milk, others noticed nothing peculiar about it. Some complained of the cocoa served at supper on Friday evening, which it will be remembered was common to the whole institution. These latter complaints are readily understood as most of the boys were in the incubation stage of an attack of gastro-enteritis, and a distaste for food would have been expected. Queries of this nature are necessarily "leading questions," and the answers have to be interpreted cautiously, but further questioning elicited the fact that porridge and milk had been left over from breakfast on Friday morning. The suggestion that condensed milk had been used was not verified. Naturally one of the first questions asked was directed to the exclusion of the possibility of tinned foods being at fault. The assurance was immediately forthcoming that tinned foods had never been supplied at any meal. The records of the amount of milk received daily from the farm were examined. They showed an appreciable surplus morning and evening, which did away with the necessity for the use of condensed milk.

Outbreaks of bacterial food-poisoning due to milk are on the whole rare, especially when the rapidity with which micro-organisms would become diffused through it as compared with their rate of spread in meat is borne in mind. Out of 112 British outbreaks tabulated by Savage (1920) milk was only responsible on nine occasions. In a corresponding table dealing with Continental outbreaks compiled by the same author (1913) no case is recorded in which milk was the vehicle. Nevertheless in the outbreak under consideration there can be no doubt that milk was the peccant food. The amount of porridge and milk left by the boys aroused comment. This alone is suggestive. It is also an unusual feature, as it is usually stated in outbreaks of bacterial food-poisoning that the peccant food presented no abnormal appearance and tasted quite good. The case in which a German food-inspector passed sausage-meat as sound and subsequently died from gastro-enteritis resulting from the consumption of the same meat may be recalled in support of this statement.

Also the milk was the only article of food served on Friday which was not common to all the boys in the Institution except the porridge, which may be disregarded as a vehicle for many obvious reasons.

II. THE CAUSAL ORGANISM.

On Sunday, October 16th, 10 c.c. of blood was taken from two boys and inoculated into 100 c.c. of sterile broth. No growth took place. The temperatures recorded before withdrawing the blood were 104° and 102° F. respectively. The blood of six boys, all severely ill, and that of the stoker was collected for the agglutination test. On the following day the seven sera were tested against 24-hour broth cultures of *B. paratyphosus* B. (C.), *B. enteritidis* of Gaertner (Limerick), and *B. Aertrycke* (Healy)—three stock strains of members of the Salmonella group. The microscopic test was applied in dilutions of 1-10 and 1-50. The sera of four of the boys had no apparent effect on any of the three organisms in these dilutions. The sera of two other boys and of the stoker agglutinated *B. enteritidis* Gaertner strongly in the 1-10 dilution in ten minutes, but had no effect in the 1-50 dilution on this organism. These three sera did not agglutinate the remaining organisms at all. The fact that strong agglutination, though only in low dilution, of *B. enteritidis* occurred, suggested that eventually this organism might be found to be the causal organism.

Specimens of faeces from four of the severe cases and a vomit which had been kept were also taken on Sunday. No member of the Salmonella group was isolated from the vomit. The faeces were emulsified in a little broth and rubbed out on the Conradi-Drigalski and Endo media. Lactose fermenters predominated, but there were several non-lactose fermenters present. From three of the samples of faeces (only after considerable "picking" in two cases) an organism was isolated which clumped with an anti-Aertrycke serum in low dilutions but agglutinated with an anti-Gaertner serum to its full titre. This organism subsequently proved to be culturally and serologically a genuine Gaertner bacillus. The sera of the seven persons which had been already tested against the laboratory strains of the Salmonella group were tested a few days later with one of the "raw" strains of Gaertner. In no case did any agglutination occur in a 1-10 dilution.

Efforts were made to obtain a sample of faeces from the stoker, but they were not successful until 44 days after his recovery. From this sample no members of the Salmonella group was recovered either by direct plating out on McConkey, Endo, and Drigalski, or by preliminary cultivation in dulcete peptone water (with and without sodium taurocholate) and brilliant green dulcete broth, and subsequent plating out. The interval which elapsed between the attack and the test is sufficient to account for the negative result, seeing that only 75 p.c. of the samples of faeces collected from boys during the height of the attack yielded positive results.

The isolation of Gaertner's bacillus from three out of four specimens of faeces from victims of the outbreak definitely established this bacillus as the causal organism.

III. THE SOURCE OF THE INFECTION.

Accepting the foregoing conclusions that milk was the peccant food and *B. enteritidis* of Gaertner the causal organism the path of infection had then to be determined. The vehicle was undoubtedly milk. No ill-effects had followed its use in the whole institution at breakfast and luncheon on Thursday. It is therefore very unlikely that it was already infected with Gaertner's bacillus on arrival from the farm. Even if it had then contained a stray organism there would have been time for proliferation to occur by mid-day and some illness would have manifested itself. In all probability some boys in each Block would have been ill. No such observation was recorded. The infection would appear to have occurred between lunch-time (mid-day) and evening. In any attempt to trace the source of the infection the stoker's illness must be taken into account. Five days before the milk became infected the stoker developed gastro-enteritis. This was not a casual attack of diarrhoea. He had severe diarrhoea (10 or 12 motions) and noticed some blood in the stools. The diarrhoea lasted 24 hours. His blood a week later clumped Gaertner's bacillus, and even 54 days later still agglutinated this organism (see Table I, No. 117). It is not unreasonable then to infer that the stoker was a sporadic case of Gaertner infection. In what way is his illness to be linked up with the contamination of the milk? Two hypotheses are put forward. The butter-room (see Plan) in which the milk was stored was not more than 50 yards distant from the servant's lavatory, which was used by the stoker during his illness and on the following days. The hypothesis is that flies may have conveyed the organism from the lavatory to the milk stored in the butter-room. The weather at this time was warm during the day, and flies would still have been about in the daytime. The alternative suggestion is that the stoker was in the habit of surreptitiously taking milk from the butter-room. In the event of his hands being soiled and coming in contact with the milk in the crock infection might easily occur. The importance of these two methods in the spread of typhoid fever is universally recognised. In bacterial food-poisoning it has not been dealt with to any extent in the literature. Two instances may however be mentioned. In the St Johann outbreak studied by Rimpau (1911) a case in the vicinity was definitely traced to the consumption of milk obtained from an infected house. Again in the Clitheroe outbreak in September, 1921, studied by Dr W. E. Barker, to whom I am indebted for the details, the suggestion is put forward that flies may have infected the peccant food while it stood cooling in a room in the proximity of which there was a water-closet used by the workmen. Other possibilities such as contamination of the milk by rodents might be put forward. The butter-room had a tiled floor and no rat-holes were found

in it, and in any event the stoker's illness if unconnected with the outbreak would have been an extraordinary coincidence.

The frequent association of outbreaks of bacterial food-poisoning with the consumption of the meat of "emergency-slaughtered" animals led me to enquire if there was a sick beast on the premises. Strangely enough it transpired that one of the milch-cows had been sick. The details of the animal's illness are as follows:—Some time previously it had developed "blood-murrian" and had been isolated. On its recovery its milk was again brought into use. On the Monday preceding the outbreak it was noticed to be suffering from "dry-murrian" and it was again isolated. During the next few days it became very weak being unable to stand, and death occurred on Saturday morning. The preliminary enquiries about fish and the milk-supply consumed a good deal of time and this information was only elicited after night-fall, when the carcass could not be isolated, and no examination of it would have been possible. I accordingly asked that some of the flesh of this animal should be forwarded to me, and this was done. A careful bacteriological examination of the flesh was made but Gaertner's bacillus was not isolated from it. A mouse was fed with some of the meat on the following day. It was dead next morning. The bedding of the animal was wet. The inguinal and axillary glands were somewhat enlarged. The spleen was not enlarged. The stomach and intestines were distended with gas. There was some meat in the stomach. The contents of the intestine were clear, yellowish, and mucoid with a partially-formed faecal pellet in the caecum and a fully-formed one in the rectum. Cultures were made from the heart's blood, spleen and contents of caecum on agar, malachite green broth, and litmus dultite broth. No member of the Salmonella group was isolated. In addition the intestinal contents were plated out on Endo's medium direct, and after dilution. The direct plate on Endo looked very hopeful after incubation over-night, non-lactose-fermenters being approximately three times as numerous as the lactose-fermenters. Nine of the most likely of the colourless colonies were picked off and studied. One of these, and one only, proved to be a genuine Gaertner bacillus. It behaved in exactly the same manner in agglutination reactions and on nutrient media as did the strains isolated in the outbreak from human sources. A second mouse was fed with the heart and one of the legs of this mouse. It died five days later. Efforts to isolate *B. enteritidis* from the organs and intestinal contents were unsuccessful. The isolation of a member of the Salmonella group of organisms from the intestines of mice, rats, or guinea-pigs is not an uncommon finding. The percentages given by various observers, however, differ very much. Similarly it has been stated by many writers that the feeding of mice with a diet to which they are unaccustomed commonly leads to their death, although the diet is uninfected. If the cow had been suffering from a Gaertner infection on Thursday morning and its milk had been used not merely would the outbreak have been general but the organism would have been isolated with ease from the meat forwarded from the carcass.

I had an opportunity of discussing the post-mortem appearances with the man who disposed of the carcass. He was the butcher attached to the institution, and he stated that he found the food impacted in the "book," the usual condition responsible for "dry-murrain." He noticed no abscesses in the flesh when securing the portion which was forwarded to me. These facts have led me to the conclusion that the sick cow had no epidemiological connection with the outbreak.

IV. THE MEASURES TO BE ADOPTED TO PREVENT THE OCCURRENCE OF SIMILAR OUTBREAKS IN THE FUTURE.

If the foregoing reconstruction of the outbreak is correct the only preventive measure of any value which had not been carried out was the protection of the milk in the butter-room from flies. The butter-room itself appeared to be an ideal room for the storage of milk. It was clean, had a fresh smell, and was provided with a fly-proof gauze ventilator situated in the roof. Still the room was part of the detached building in which the kitchen and pantries were situated, and flies from these portions of the building could gain access to the milk. The simple expedient of covering the milk with butter-muslin was an obvious safeguard against this danger¹. Nevertheless the other arrangements for handling the milk were so satisfactory that one could not attach any blame to the institution for this omission. The exclusion of servants from the butter-room would seem to present greater difficulties, and yet the potential danger of a servant of uncleanly habits is considerable. At any rate short of pasteurising the milk for porridge every morning these two suggestions were the only improvements recommended.

The prevention of sporadic cases of infection with Gaertner's bacillus is naturally impossible. We have very little evidence as to how they originate. We know little or nothing of their frequency. Very probably they occur much more often than a survey of the literature would suggest. In the ordinary course of such a case the danger would be over and the patient on the road to recovery before a bacteriological examination would be thought of. The outbreak, on the other hand, arouses more interest, receives publicity owing to members of different families being involved, and is usually investigated bacteriologically at the instance of the Medical Officer, or of the Coroner, fatal cases being met with commonly. It was suggested that the freeing of the drain might have borne some relation to the stoker's illness, and perhaps even to the outbreak itself, independent entirely of the stoker. Possibly it had. If the rat be accepted as the likely reservoir of Gaertner's bacillus the possibility of one of these organisms being splashed on the ground in front of the engine-house during the operations on the drain must be admitted. Now if the stoker kept a mug of milk in the engine-house the transference of a stray Gaertner organism to the milk if given an opportunity of multiplying

¹ This precaution would apparently have prevented the occurrence of the Clitheroe outbreak also.

under such favourable conditions might be the cause of his developing gastro-enteritis. The stoker, however, was not confined to the institution, and may have become infected elsewhere.

Certain points remain which require further consideration. It will be remembered that the milk which caused the illness of most of the boys in Block A was used by the servants at breakfast a few hours previously. Out of a staff of 40 servants only five were ill. This fact appeared to upset the milk-theory. Investigation showed that the five sick servants were pantry-boys. These boys cleared away the remains of meals in the refectory, and their illness is easily explained on the supposition that they partook of some of the porridge and milk left by the students that morning. How then is the escape of the remaining servants accounted for? The milk set aside on the previous day, Thursday, was placed in two earthenware crocks in the butter-room, where it stood till the following morning. There are three possibilities for consideration:

(1) The servants may have been in the habit of taking their supply of milk from the delivery of the previous evening, in spite of the arrangement supposed to exist. In that case the milk kept over from Thursday morning would have been used for puddings. This may have been so but obviously could not have been verified.

(2) It is unlikely that more than one crock of milk was infected. Now if the servants took their supply from the uninfected crock their escape is at once explained. In that event the milk remaining in that crock must have been emptied into the infected supply because no mess was found in which all the boys had escaped.

(3) The remaining possibility is that again one crock was infected, that the servants drew their supply from it, and then poured the residue into the uninfected portion.

It was felt that the third possibility would have to be investigated if the view that milk was the peccant food was to have a solid foundation. It was ascertained that the servants' tea was served in an urn. As soon as the tea was made the milk was poured into the urn, and it was assumed that it stood about five minutes while the servants were assembling. The viability of Gaertner's bacillus under these conditions was studied as follows:—A culture of *B. enteritidis* (Dublin) was made in broth and allowed to stand at room-temperature for a few days. A loopful was then sown in a large tube of milk at 6 p.m., and growth allowed to take place overnight at room-temperature. On the following morning 200 c.c. of an infusion of tea in an Erlenmeyer flask was heated just short of boiling-point. The flask was then set on the bench. Six c.c.¹ of the inoculated milk was immediately added to the flask and the time noted. The temperature before the admixture was 98° C., after admixture 90° C. Five minutes later 0.1 c.c. of the tea was added to glucose

¹ It was ascertained beforehand that 6 c.c. of whole milk added to 200 c.c. of tea gave a palatable mixture.

agar and a plate culture made. This was repeated at $2\frac{1}{2}$ minute intervals until 15 minutes after the admixture. In none of the plates did any growth occur. A control test showed that the milk contained approximately 4800 Gaertner bacilli per c.c. This experiment was repeated using *B. enteritidis* (Dublin: Strain Mouse 1 Faeces) with the same result, the control tube in this case giving approximately 6500 organisms per c.c. On a third occasion 0.5 c.c. was plated out under similar experimental conditions and again no growth occurred. These experiments satisfactorily explain the escape of the servants. Even if their supply was infected the temperature to which it was subjected was sufficient to kill off the Gaertner bacilli present.

It may be argued that the agglutination of Gaertner's bacillus by the serum of the stoker was unspecific. This may have been so, but the coincidence is hard to explain. Some six weeks later his serum agglutinated the Dublin strain which it had previously failed to do (see Table I). Certain statements in the literature have a bearing on this point. Thus Dawson (1915) reported negative results in agglutination tests with a strain of Gaertner's bacillus (Delépine 7160) against the sera of 50 persons inoculated with typhoid vaccine. Again in the St Johann outbreak Rimpau found that a positive Widal reaction (against *B. enteritidis*) was never present before the second week. On the other hand O'Farrell (1916) has recorded the results of agglutination tests with *B. enteritidis* (Delépine 7160) in which 19.6 p.c. of the cases clumped in a 1 in 25 dilution. Most of these cases were soldiers who had received anti-typhoid inoculation. In the epidemic studied by Perry and Tidy (1919) they considered a positive serum reaction of diagnostic value (they were dealing with an Aerttrycke infection). The general impression gathered from the literature is that agglutinins for the Salmonella group are only present in infected persons, but it must be borne in mind that a small percentage of such people (as verified by examination of the faeces) often develop no symptoms. This whole question is deserving of further study.

As is usual in outbreaks of bacterial food-poisoning there are gaps in the chain of evidence. The outbreak reconstructed in the foregoing pages is no exception in this respect. At the same time the facts presented appear to be conclusive, and no alternative explanation suggests itself. I wish to place on record the fact that the authorities of the institution, at whose request the enquiry was conducted, facilitated me in every way. I have to thank the Medical Officer of the Institution for the trouble taken by him in the collection of material for examination. Lastly I have to acknowledge my indebtedness to Dr T. Dillon, a post-graduate student in the laboratory, for his assistance in the routine work of the investigation.

SUMMARY.

1. The outbreak was one of bacterial food poisoning due to the consumption of infected milk.
2. The causal organism was *B. enteritidis* of Gaertner (Dublin).
3. A case of gastro-enteritis occurred in an employee in the institution some days previously. This man's serum possessed agglutinins for Gaertner's bacillus, but the organism was not isolated from a specimen of his faeces taken 44 days after this attack.
4. This employee is supposed to have been the source of the organism which infected the milk. Two hypotheses are put forward to explain the mechanism of infection.
5. Servants who took the infected milk in their tea escaped infection. The milk was added to the tea in bulk and the experiments described satisfactorily account for their escape.

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APPENDIX.

CULTURAL CHARACTERS OF *B. ENTERITIDIS* OF GAERTNER (DUBLIN).

Cultures of the strains II A_1 , IV B_1 , IV C_1 , IV C_2 , V B_1 , isolated from victims of the outbreak, of Strain Mouse 1 (Faeces), of *B. enteritidis* (Limerick), *B. Aertrycke* (Healy), and of *B. paratyphosus* B. (C_1) were made on agar, broth, peptone water, gelatine, and potato. They presented the usual characteristics of cultures of the Salmonella Group. On neutral-red fluorescence was developed in every case. In milk the rate of change from the initial acid to the strongly alkaline reaction varied. In some cases five days sufficed, in others nine days were required. Indol was not formed in peptone water.

In the fermentation tests no change was observed on inulin, raffinose, lactose, saccharose, and salicin. On arabinose the Dublin strains caused no

change, Gaertner formed acid, Paratyphoid B., and Aertrycke acid and gas. On maltose, glucose, laevulose, galactose, mannite, and dulcite all formed acid and gas except Gaertner, which formed acid, but no gas, on glucose and laevulose, and Aertrycke which formed slight acid, but no gas, on dulcite. The amount of gas produced varied, being frequently very slight with the raw strains, and occasionally so with the laboratory strains. The fermentation tests were repeated using glucose, laevulose, arabinose, dulcite, sorbite, and amygdalin. No change was observed on amygdalin. All strains formed acid and gas on sorbite and dulcite, and all produced acid and gas on glucose, laevulose, and arabinose, except Gaertner's bacillus which again only produced acid on these sugars. After five days the six raw strains had discharged the colour in the arabinose tubes, the three stock strains had not. As before gas production with the raw strains was not vigorous. Andrade's indicator was employed in these tests.

VIRULENCE TESTS.

These tests were conducted on mice with *B. enteritidis*, strains II A_1 , IV B_1 , and Mouse I Faeces, and Limerick being used. One mouse was inoculated subcutaneously, the others intraperitoneally. Both cultures were employed when four hours old, and again after five days incubation, 0.01 c.c. being the amount injected. The findings agreed in every case. The mice became ill, showed some evidence of diarrhoea, and their eyes became glued up. Five of the mice inoculated with the raw strains died in four days, the sixth in three days. The mouse injected with the four-hour culture of the Limerick strain died in seven days. The older culture of this strain caused death in five days. At autopsy the intestines were empty, the spleen slightly or not at all enlarged. Pseudo-tubercles were uniformly found in the liver. They varied in size and number in the individual mice. In each case Gaertner's bacillus was recovered from the organs.

The results of the agglutination tests are set out in Table I. The Medical Officer stated that Nos. 120 and 121 were unaffected. The results of agglutination tests on their sera suggests that they at any rate became infected.

Table II shows the results of the agglutination tests. These tests were conducted in two series separated in the table by a double line. The formalised cultures were diluted, those in each batch being brought to the same opacity. The two series of tests were put up on different days and the standard suspensions selected on these occasions were not necessarily of the same density.

The anti-sera were obtained from the Lister Institute.

Table I. Agglutination results with certain sera against the Salmonella group.

No.	B. enteritidis Gaertner (Dublin)			B. enteritidis Gaertner (Limerick)			B. Aertzycke (Healy)			B. paratyphosus B. (C ₁)					Remarks
	1-10	1-50	1-100	1-250	1-100	1-250	1-10	1-50	1-100	1-250	1-10	1-50	1-100	1-250	
3	++	++	++	++	++	++	+	±	—	—	++	++	++	++	Gaertner in faeces
7	++	++	++	++	++	++	+	±	—	—	++	++	++	++	Severely affected
8	++	++	++	++	++	++	+	±	—	—	++	++	++	++	—
9	++	++	++	++	++	++	+	±	—	—	++	++	++	++	—
95	++	++	++	++	++	++	+	±	—	—	++	++	++	++	Severely affected
97	++	++	++	++	++	++	+	±	—	—	++	++	++	++	" "
109	++	++	++	++	++	++	+	±	—	—	++	++	++	++	" "
111	++	++	++	++	++	++	+	±	—	—	++	++	++	++	—
114	++	++	++	++	++	++	+	±	—	—	++	++	++	++	—
117	++	++	+	—	—	—	±	—	—	—	±	—	—	—	Patient from whose faeces the strain of B. Gaertner (Dublin) used in test was isolated
118	++	±	—	—	—	—	++	+	—	—	±	—	—	—	Stoker, supposed to be cause of outbreak; 54 days after illness serum collected
119	++	++	++	—	—	—	+	—	—	—	++	++	+	—	Pantry-boy was very seriously ill
120	++	++	++	—	—	—	++	±	—	—	±	—	—	—	Lightly affected; suffers from mild diarrhoea still
O'K.	±	—	—	—	—	—	++	±	—	—	++	—	—	—	Unaffected
T. D.	++	—	—	—	—	—	—	—	—	—	++	—	—	—	Worker in laboratory
M.	—	—	—	—	—	—	±	—	—	—	—	—	—	—	" "

++++ = complete agglutination; -- = no agglutination. Results read off after one hour (low power).

Table II. Agglutination tests with formalised cultures.—Macroscopic test—2 hours at 52° C. Final reading overnight.

Organism	Gaertner serum					Suipestifer (Type A) serum					Paratyphosus B. serum				
	1-2000	1-4000	1-6000	1-8000	1-10,000	1-200	1-1000	1-2000	1-4000	1-8000	1-1000	1-2000	1-4000	1-6000	1-8000
Gaertner (Limerick)	++++	++++	++++	++++	++	—	—	—	—	—	—	—	—	—	—
Aertzycke (Healy)	++++	++++	++++	++++	++	++	++	++	++	++	—	—	—	—	—
Gaertner (Dublin); IV B ₁	++++	++++	++++	++++	++	++	++	++	++	++	—	—	—	—	—
" IV C ₁	++++	++++	++++	++++	++	++	++	++	++	++	—	—	—	—	—
" IV C ₂	++++	++++	++++	++++	++	++	++	++	++	++	—	—	—	—	—
V B ₁	++++	++++	++++	++++	++	++	++	++	++	++	—	—	—	—	—
Gaertner, Mouse 1 (Faeces)	++++	++++	++++	++++	++	++	++	++	++	++	—	—	—	—	—
Paratyphosus B. (C ₁)	—	—	—	—	—	++++	++++	++++	++++	++++	++++	++++	++++	++++	++++
Paratyphosus B. (McW.)	—	—	—	—	—	++++	++++	++++	++++	++++	++++	++++	++++	++++	++++
Gaertner (Limerick)	++++	++++	++++	++++	++	++	++	++	++	++	—	—	—	—	—