

BACTERIAL CONTAMINATION OF HOSPITAL FOOD WITH  
SPECIAL REFERENCE TO *CLOSTRIDIUM WELCHII*  
FOOD POISONING

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INTRODUCTION

The role of foodstuffs, contaminated by potentially pathogenic bacteria, has long been established as one of the commonest causes of gastro-enteritis, but the control of this condition remains a major public health problem in all communities particularly among those living in institutions. In hospitals, where a high proportion of the patients might be expected to react more severely to the ingestion of bacterially-contaminated food, it is especially important that the food purchased should be 'bacteriologically clean' and that in the place where it is handled and processed the standards of kitchen hygiene should be high.

In the hospital under review there have been mild outbreaks of gastro-enteritis from time to time, but these have been no more frequent or severe than in other large hospitals where the majority of the patients' meals are prepared and cooked in a central kitchen. It was considered, however, that it would be useful to make a bacteriological survey of the various foodstuffs as purchased by the hospital and of the cooked foods as served to the patients, and also to investigate the various procedures followed in the kitchen, in the hope of providing information which might be of use to hospitals generally. In addition to these special inquiries outbreaks of gastro-enteritis in the hospital continued to be investigated by standard epidemiological and bacteriological methods.

The bacteriological survey of the foods was designed so that any organism which might be responsible for an outbreak of food poisoning would be identified, but as the work proceeded it became clear that the main interest would be centred on the contamination of foodstuffs by *Clostridium welchii*.

MATERIALS AND METHODS

*Source of food specimens*

During a period of 2½ years 300 samples of food were collected from the kitchens of the hospital. The items were selected to include (a) a variety of uncooked foods purchased by the hospital (89 samples), (b) samples of the same foods after they had been cooked and prepared in the hospital kitchens for serving to the patients (173 samples), and (c) foods which were bought already cooked and served to the patients with a minimum of further preparation (38 samples). Specimens were collected in sterile containers, taken to the laboratory and examined the same day. A note was made about the gross condition of the food specimen; if it had a putrid appearance or a bad smell a smear preparation was made and stained by Gram's method for direct microscopical examination.

*Source of faecal specimens*

During the same period all reported outbreaks of diarrhoea within the hospital were investigated epidemiologically and bacteriologically and as far as possible faecal specimens were collected from all the patients affected. A smear of each stool was made and stained by Gram's method to exclude possible cases of staphylococcal enterocolitis.

*Examination of specimens*

The survey of the food was undertaken as a qualitative test rather than a quantitative estimation of bacterial contamination. Nevertheless, in order to assess the potential danger of the food under investigation the degree of contamination was determined roughly by observing the growth obtained after plating an inoculum of standard size on solid media. When the food was of a solid nature a portion of about 15 g. was finely shredded in a sterile Petri dish where it was mixed with 15 ml. of sterile nutrient broth; the mixture was left on the bench for a few minutes to allow the contaminating organisms to disperse evenly before inoculating the solid media. A quantity of the shredded food, to cover two scalpel blades, was added to each of four fluid media. The following media were used for the inoculation of each specimen: two horse-blood-agar plates, one MacConkey-agar plate, 1 × 50 ml. Robertson's cooked-meat-broth, 1 × 50 ml. Selenite F broth, 1 × 10 ml. Robertson's cooked-meat-broth + 10 % NaCl and 1 × 25 ml. nutrient broth.

The nutrient broth was dispensed in screw-capped universal containers (25 ml.) which, after inoculation and before incubation, were immersed in a bath of boiling water for 15 min. to test for the presence of heat-resistant *Cl. welchii*. One horse-blood-agar plate was incubated in a McIntosh and Fildes' anaerobic jar at 37° C.; all other inoculated media were incubated at the same temperature aerobically. After incubation the media were examined by appropriate techniques for the presence of *Cl. welchii*, *Staphylococcus aureus* and also intestinal pathogens such as members of the salmonella and shigella groups.

Faecal specimens from patients were examined by similar methods with the same range of media, but with appropriate adjustments in the size of the sample used for the inocula.

*Identification of Clostridium welchii*

The identity of the strains of *Cl. welchii* isolated was verified by testing for the production of the  $\alpha$ -toxin (lecithinase) which was specifically inhibited by *Cl. welchii*  $\alpha$ -antitoxin. This was a tube-test involving the use of 'lecithovitellin', a filtered preparation of one egg yolk in 250 ml. of saline (Macfarlane, Oakley & Anderson, 1941).

Serological typing of most of the strains of *Cl. welchii* was carried out by Dr Betty Hobbs of the Food Hygiene Laboratory of the Public Health Laboratory Service at Colindale. In addition, I prepared antisera to three strains isolated during outbreaks of diarrhoea. The sera were prepared by intravenous injections of rabbits with successively increasing doses (from 0.1 to 1 ml.) of formalized

bacterial suspension (0.4% formalin) prepared according to the method described by Henderson (1940). The injections were given at intervals of 2 or 3 days over a period of 2 weeks. In practice, because of the death of two rabbits immediately after their final injection due to massive pulmonary emboli containing Gram-positive bacilli, this method of preparing antisera was modified and the last two injections of 1 ml. were administered 0.5 ml. intravenously and 0.5 ml. intramuscularly. Tube-agglutination tests were made with a dense living suspension of the test organism. Readings were made after 4 hr. at 43° C. in a water-bath and again after the tubes had been kept overnight in the refrigerator and 12 hr. at room temperature (Hobbs, Smith, Oakley, Warrack & Cruickshank, 1953).

#### *Identification of Staphylococcus aureus*

The production of coagulase was taken as the criterion of potential pathogenicity and all coagulase-negative strains were discarded. In this paper, therefore, all further references to *Staph. aureus* or staphylococci denote coagulase-positive *Staph. aureus*. The coagulase tests were carried out in tubes with human citrated plasma. The *Staph. aureus* cultures were phage-typed by Dr Morag Timbury of this department.

#### *Identification of specific intestinal pathogens*

The usual routine methods were employed for the identification of salmonella and shigella organisms. The cultures were examined for the production of urease and indole, and the fermentation of glucose, lactose, sucrose, manitol and dulcitol was tested. Final identification was made by specific antisera.

### RESULTS OF THE INVESTIGATIONS

#### *Incidence of potentially pathogenic organisms in the food from the hospital kitchens*

(a) *Uncooked food purchased by the hospital.* Eighty-nine samples of food in this group were examined. The contamination of these items with *Cl. welchii* and *Staph. aureus* is shown in Table 1. Only one specimen, an uncooked sausage, contained a specific intestinal pathogen—namely, *Salmonella enteritidis*.

On direct culture many samples of uncooked food were found to be contaminated with fairly large numbers of saprophytic organisms such as anthracoids, coliforms, and micrococci. However, a potentially pathogenic organism was isolated from only one sample (of chicken) on direct culture; and as this isolate was a single colony of a haemolytic *Cl. welchii* it cannot be regarded as certainly significant.

By enrichment culture *Cl. welchii* was isolated most frequently from sausage and chicken; indeed, it was isolated from every sample of these two foods. Similarly, a heat-resistant strain of *Cl. welchii* was found in one sample of each of these foods, but was not isolated from any other item of diet. However, due to the ubiquitous nature of *Cl. welchii* and *Staph. aureus*, their presence in small numbers on uncooked food cannot be regarded as an indication of uncleanness and, after

arrival at the kitchen, the foods were refrigerated to prevent further multiplication of these organisms before the food was cooked.

(b) *Food cooked and prepared for serving in the hospital kitchens.* In selecting samples of food for this group I attempted to include the same items of diet which were examined in group (a); whenever possible portions of the same batch of food

Table 1. *Contamination of uncooked food with Clostridium welchii and Staphylococcus aureus*

Nature of food	No. of samples examined	No. containing			
		<i>Cl. welchii</i>			<i>Staph. aureus</i>
		Haemolytic	Non-haemolytic	Heat-resistant	
Fish	18	11 E	3 E	0	4 E
Liver	2	0	1 E	0	0
Steak and mince	10	6 E	0	0	3 E
Tripe	6	1 E	1 E	0	1 E
Sausage	38	36 E	9 E	1	5 E
Black pudding	4	2 E	0	0	1 E
Ulster fry	4	2 E	0	0	0
Chicken	7	1 D 6 E	3 E	1	1 E
Total	89	1 D 64 E (1.1%) (71.9%)	16 E (17.9%)	2 (2.2%)	15 E (16.8%)

D = isolated on direct culture. E = isolated after enrichment culture only.

were sampled both before and after cooking. Cooked and uncooked samples of fish cake and savoury croquette were included, because these items were prepared from already cooked materials and then recooked. The total number of samples in this group was 173 and the contamination with *Cl. welchii* and *Staph. aureus* is shown in Table 2.

No salmonella or shigella organisms were isolated; an examination of a cooked sausage from the same batch as that in which an uncooked sausage contained *Salm. enteritidis* proved negative. All bacterial contamination was very much reduced after the foods were cooked and prepared for serving. Contamination with *Cl. welchii* and *Staph. aureus* was commonest in foods which required further handling after cooking. Presumably, therefore, this contamination lay on the surface. The results of the examination of fish cakes and savoury croquettes supported this conclusion.

The incidence of *Cl. welchii* in sausages was very much reduced after cooking, and no sample of sausage, either stewed or fried, contained a heat-resistant strain. One sample of sausages, however, was brought to my attention by members of the kitchen staff on account of its unpleasant smell. After being stewed, this particular batch of sausages was left to cool overnight. A sample of the sausage gravy, collected the day after cooking, gave a heavy growth of haemolytic and non-haemolytic *Cl. welchii* and these organisms were apparent on the direct film.

Haemolytic and non-haemolytic strains were also isolated from a sample of the sausage meat but only after enrichment culture.

The most outstanding finding of this examination was the frequent contamination of cold chicken with *Cl. welchii* (24 of 46 samples). Early in the investigation it was noted that some samples of cold chicken were contaminated with large numbers of *Cl. welchii* and an incident among the laboratory staff, reported below,

Table 2. Contamination of food cooked in the hospital kitchens with *Clostridium welchii* and *Staphylococcus aureus*

Nature of food	No. of samples examined	<i>Cl. welchii</i>			<i>Staph. aureus</i>
		Haemolytic	Non-haemolytic	Heat-resistant	
Fish	6	1E	0	0	0
Liver	1	0	0	0	0
Steak and mince	15	1E	1E	0	0
Tripe	6	1E	0	0	0
Sausage	25	4E	1E	0	0
Sausage gravy	1	1D	1D	0	0
Black pudding	2	2E	2E	0	0
Ulster fry	2	0	0	0	0
Chicken	46	8D 13E	4D 7E	1	9E
Chicken broth	3	0	0	0	0
Cold roast meats	15	4E	2E	0	2E
Other cold meats	37	3E	3E	0	7E
Fish cake (uncooked)	1	1E	0	0	1E
Fish cake (cooked)	1	0	0	0	0
Croquette (uncooked)	8	4E	4E	0	1E
Croquette (cooked)	4	0	0	0	0
<b>Total</b>	<b>173</b>	<b>9D 34E</b> (5.2%) (19.6%)	<b>5D 18E</b> (2.8%) (10.4%)	<b>1</b> (0.6%)	<b>20E</b> (11.5%)

D = isolated on direct culture. E = isolated after enrichment culture only.

indicated that this was a potential source of *Cl. welchii* food poisoning. This incident also indicated that contamination with haemolytic non-heat-resistant strains was not necessarily less dangerous than contamination with non-haemolytic heat-resistant strains. Although three samples of chicken broth prepared from the same birds did not contain *Cl. welchii*, this was presumably because the broth samples were collected by the laboratory staff on the same day as the birds were cooked, whereas the chicken samples were received the next day, after the chickens were carved.

Despite the frequency of contamination with *Cl. welchii*, only two samples of cold chicken appeared bad to the unaided senses. The deterioration of one of these samples did not appear to be due to the growth of *Cl. welchii* but the sample had been reheated before being sent for examination. The second sample of 'bad chicken' examined gave a heavy growth of haemolytic *Cl. welchii* on direct culture and the direct smear showed large numbers of Gram-positive bacilli. Usually, however, the cold chicken, although it sometimes contained large numbers of

*Cl. welchii*, neither appeared putrid nor smelled bad, thus giving no indication from its external appearance of the bacterial contamination.

With the exception of cold chicken the food cooked in the hospital kitchens was relatively free of potentially pathogenic organisms when served to the patients.

(c) *Already cooked foods bought by the hospital.* This group comprised thirty-eight samples and the contamination with *Cl. welchii* and *Staph. aureus* is given in Table 3. The cold meats required slicing after purchase, and the mutton pies and sausage rolls were reheated before serving.

Bacterial contamination of the samples in this group was minimal. Sausage rolls and meat pies are frequently incriminated as the vehicles of food poisoning in the community as a whole and it is of interest to note that these foods were of a very high standard of bacteriological cleanliness. The twelve specimens examined gave no growth on direct culture and enrichment methods revealed only anthracoids and micrococci.

Table 3. *Contamination of cooked food purchased by the hospital with Clostridium welchii and Staphylococcus aureus*

Nature of food	No. of samples examined	No. containing				<i>Staph. aureus</i>
		<i>Cl. welchii</i>				
		Haemolytic	Non-haemolytic	Heat-resistant		
Mutton pie	7	0	0	0	0	
Sausage roll	5	0	0	0	0	
Cold meats	26	1 E	1 E	0	3 E	
Total	38	1 E (2.6 %)	1 E (2.6 %)	0	3 E (7.8 %)	

E = isolated after enrichment culture only.

*Characteristics of the Clostridium welchii and Staphylococcus aureus isolated from food*

*Cl. welchii.* All strains of *Cl. welchii* isolated showed typical morphology. On solid media the colonies of most strains were round with an entire edge, but occasionally rough colonies with an irregular edge and radial striations were observed. The difference in colonial form was unrelated to the production of haemolysis on horse-blood agar. Both haemolytic and non-haemolytic strains were found in foods; only three heat-resistant strains were isolated and all were non-haemolytic. The haemolytic strains showed a well-defined zone of  $\beta$ -haemolysis on horse-blood agar. The non-haemolytic strains usually had no effect on horse blood on primary isolation, but a slight zone of incomplete  $\alpha$ -haemolysis sometimes developed round the colonies after a few days. One of the originally heat-resistant strains retained its heat-resistance in culture, surviving 1 hr. boiling. Cultures of the other two heat-resistant strains were killed by 5 min. boiling. On occasions, serology or growth characters on blood agar showed that more than one strain of *Cl. welchii* was present in an individual sample of food.

*Staph. aureus*. The morphology and cultural characteristics of the *Staph. aureus* isolated were typical, although the individual strains differed among each other in their colony size, haemolysis on horse-blood agar and pigment production. Twenty-five of the thirty-eight strains isolated were phage-typed; 15 (60%) belonged to either group I or II and 10 (40%) were in group III or untypable. No individual sample of food was found to harbour more than one strain of staphylococcus.

*Further observations on the contamination of chicken with Clostridium welchii*

Because of the relatively frequent contamination of cold chicken with *Cl. welchii*, the cooking and storing of the fowls was investigated in detail. It was first thought advisable however, to determine if the high rate of contamination of cold chicken, as compared with other meats, was primarily due to nutrients in chicken which favoured the growth of *Cl. welchii*. To investigate this I studied the growth of two strains of non-haemolytic *Cl. welchii* in chicken-extract and meat-extract broths. Both strains grew better in meat-extract broth.

Table 4. Contamination of cold chicken from the main and special-diet kitchens with *Clostridium welchii*

Kitchen	No. of samples examined	No. containing <i>Cl. welchii</i>		
		Haemolytic	Non-haemolytic	Heat-resistant
Main	31	8D (25.8%)	11E (35.5%)	4D 5E (12.9%) (16.1%)
Special diet	15	2E (13.3%)	2E (13.3%)	1 (6.6%)

D = isolated in direct culture. E = isolated after enrichment culture only.

The chicken samples examined were received from two separate kitchens in the hospital: the main kitchen which served the staff and the majority of the patients, and the special-diet kitchen which supplied meals for the patients on special diet only. Of the forty-six samples examined, thirty-one came from the main kitchen and fifteen came from the special-diet kitchen. The contamination with *Cl. welchii* of the chicken from the respective kitchens is shown in Table 4. There was a striking difference in the number of samples containing *Cl. welchii*; no sample of chicken from the special-diet kitchen yielded *Cl. welchii* on direct culture whereas from the main kitchens eight samples (25.8%) were found to contain haemolytic strains and four samples (12.9%) non-haemolytic strains of *Cl. welchii*. In some instances these organisms were present in large numbers and almost pure culture. The results from the enrichment cultures of samples from the two kitchens showed differences of the same order. It was therefore decided to investigate the methods employed for the cooking and preparation of the fowls for serving to the patients to see whether they revealed any significantly different procedures in the two places.

The actual cooking methods employed in the two kitchens were essentially the same; in the main kitchen batches of about twenty fowls were cooked together in a large steam-jacketed boiler for  $2\frac{1}{2}$ –3 hr., while in the special-diet kitchen two or three birds were cooked in a steam oven for 2–3 hr. The true temperatures attained within the birds during the cooking process were determined by means of a copper-constantan thermocouple. In both types of cooker the temperature within the flesh of the fowls rose to about 97° C. in 55–65 min. from the start of cooking and remained between 97 and 100° C. until the process was completed. In the actual recordings made, temperatures of approximately 100° C. were maintained for 80–115 min.

Zeissler & Rassfeld-Sternberg (1949) and Hobbs *et al.* (1953), reported that heat-resistant spores of *Cl. welchii* could survive temperatures of 100° C. for up to 5 hr. The procedures recorded above for cooking the fowls would therefore be inadequate to kill such heat-resistant spores of *Cl. welchii*, or possibly even some less heat-resistant strains, had they been present in the birds before cooking. Whether they, or other strains that might gain access to the fowls after cooking, could grow and multiply during the period between cooking and serving to the patients would depend upon the conditions of storage. In spite of the similarity of the cooking procedures in the two kitchens there was a very significant difference in the degree of clostridial contamination in the two sets of birds, and it therefore seemed important to make a detailed inquiry in each case as to what happened between cooking and serving.

In the main kitchens the fowls were usually cooked about midday after which the complete batch of birds together with their broth was transferred to a large zinc bath ( $2\frac{1}{2} \times 2 \times 1$  ft.) which was left uncovered outside the kitchen, but not in the cold room. Here the air temperature was lower than in the kitchen itself but not low enough to promote rapid cooling. The next morning the birds were removed from the broth, carved and distributed to the wards to be served for lunch. It will be seen that there was a lapse of nearly 24 hr. between cooking and serving and for the greater part of this time the packed mass of birds was partly immersed in the exposed slowly cooling broth.

In the special-diet kitchen, on the other hand, the fowls were removed from the dishes in which they had been cooked and were placed, well separated from each other, on shallow trays. They did not stand in their broth; this was poured from the cooking dish separately into a jug. The fowls and the broth were left to cool on a side bench in their individual containers for some 3–4 hr. after which they were placed in the refrigerator where they remained until required.

The cooling temperatures were ascertained by the same method as described above, but were determined either on the birds on shallow individual trays or on those at the top of the large bath from the main kitchen. The risk of contamination made it inadvisable to take temperatures of birds deep in the bath. The cooling temperatures fell to about 50° C. in a matter of 1–2 hr. but thereafter the rate of cooling was much reduced. This indicated that the cooked birds, if left unrefrigerated, would remain within the dangerous range of 20–40° C. for long enough to allow considerable multiplication of *Cl. welchii*, and presumably the temperature



of the birds deep in the bath from the main kitchen would remain at this level for considerably longer than those at the top. The high rate of contamination of the chickens from the main kitchen, as compared with those prepared in the special-diet kitchen, would therefore seem to be related to the method of storage between cooking and serving.

Tables 1 and 2 show that out of a total of fifty-three samples of cooked and uncooked chickens examined only two heat-resistant strains of *Cl. welchii* were isolated; the contamination of the cold chicken from the main kitchen could not therefore, be regarded as being mainly due to the presence of heat-resistant strains which had survived the cooking. It was not feasible to judge whether the non-heat-resistant non-haemolytic strains isolated were derived from heat-resistant spores, because large numbers of non-heat-resistant haemolytic *Cl. welchii* were frequently present and these could have gained access to the samples after cooking. The most obvious source (and the most probable) of these organisms seemed to be the dust in the kitchen to which the fowls were exposed during the transfer from the cooking pots to the cooling container and during the cooling period.

Samples of dust were taken from various sites in the kitchen by rotating sterile broth-soaked swabs over the appropriate area. In all, fifty-eight samples were collected from seven sites on the floor and window ledges and both haemolytic and non-haemolytic *Cl. welchii* were isolated from all sites on more than one occasion. Haemolytic strains were found in 89.6% of the samples and non-haemolytic in 81.0% so that most samples contained both types. From the special-diet kitchen eight samples were collected from comparable areas and all eight swabs yielded haemolytic or both haemolytic and non-haemolytic *Cl. welchii*. The organism was therefore widely distributed in the air of the two kitchens and if the same conditions prevailed in the two places there would be ample and equal opportunity for the contamination of the cooked chickens with this infective dust.

#### Laboratory experiments

Various experiments were performed in the attempt to reproduce in the laboratory some part of the kitchens' procedures with the ultimate object of finding methods of reducing the risk of *Cl. welchii* food poisoning among the patients.

Four pieces of fowl, all of which had been found to contain non-heat-resistant *Cl. welchii* before cooking, were cooked in a Koch's steam sterilizer to imitate as far as possible the method followed in the main hospital kitchen. During cooking the thermocouple method of recording the temperatures was employed to ensure that they were comparable to those attained in the hospital kitchens. After removal from the steamer these samples were handled with aseptic precautions and each was placed in a sterile 1 l. beaker and covered with a sterile Petri-dish lid. Each sample was then divided into two, and one half of each in a similar container was placed in the refrigerator and the other half left at room temperature, the accompanying broth being equally divided between the two parts. Samples were removed daily for bacteriological examination.

Neither at room temperature or from the refrigerator were any of the samples found to be contaminated with *Cl. welchii*, although by the sixth day three of the

pieces kept at room temperature showed a moderate contamination with aerobic spore-bearing bacilli and micrococci. The remaining portion kept at room temperature and all the refrigerated pieces showed only a few aerobic organisms by the sixth day and these probably gained access during the manipulation of the specimens.

The experiment clearly indicated that the temperatures attained in the cooking of the fowls were adequate to kill non-heat-resistant *Cl. welchii*. The simple protection offered in the experiment, by covering the beakers with the Petri-dish lids, appeared to be sufficient to prevent subsequent contamination with *Cl. welchii*, even of the unrefrigerated samples, during the period immediately after cooking, when the conditions might be considered suitable for the growth of this organism. The air of the laboratory in which the experiments were conducted was heavily infected with *Cl. welchii* and other organisms, yet the only contaminants found at the end of 6 days storage were moderate numbers of aerobic bacteria which presumably gained entrance during the removal of the specimens for examination from the cold pieces of fowl when they were unprotected from aerial contamination.

Another specimen of fowl, the flesh of which contained haemolytic *Cl. welchii* before being cooked, was divided into two portions. Each portion was inoculated deeply into the flesh with 0.1 ml. of a 6-day culture of a non-haemolytic strain of *Cl. welchii* known to be resistant to boiling for 1 hr. The two portions were cooked in a Koch's steam sterilizer for 2 hr. 40 min. after which each portion was transferred to a lid-covered sterile beaker. The broth was equally divided between the two beakers so that each piece of fowl was about one-quarter immersed in the broth with the upper three-quarters protruding. One beaker was placed in the refrigerator while the other was allowed to remain on the bench at room temperature. Specimens were taken from the flesh and the broth for bacteriological examination after 24 and 48 hr., but all the cultures were negative for *Cl. welchii*. It is evident, therefore, that the cooking procedures were adequate to kill this relatively heat-resistant strain and that only those possessing a high degree of resistance could have survived.

Finally, a piece of fowl was divided into two portions and cooked as above. After cooking and removal from the steamer, the broth of each portion was inoculated with 0.1 ml. of a 6-day culture of a haemolytic *Cl. welchii*; one portion was placed in the refrigerator and the other left at room temperature. A standard loopful of broth from the refrigerated sample yielded ten colonies of the strain of *Cl. welchii* on direct plating after 24 hr. and eight colonies after 48 hr. The organism was identified by slide-agglutination with the homologous antiserum. Cultures from the refrigerated flesh were negative after 24 hr. and only one colony of *Cl. welchii* grew on the direct plate after 48 hr. There was therefore no multiplication of the organism in the refrigerated sample. On the other hand, both the broth and the flesh of the sample left at room temperature yielded heavy confluent growths of *Cl. welchii* after 24 and 48 hr. Here the organism had invaded the flesh from the broth and multiplied extensively overnight; the broth was noticeably cloudy and that part of the flesh which was immersed in the broth was

pink. Even after 5 days there was no putrid smell, indeed, the appearance remained attractive and the smell quite appetizing. The results of this experiment show that in cooked chicken contaminated with relatively large numbers of *Cl. welchii*, multiplication of the organism could be prevented by adequate refrigeration.

#### *Outbreaks of food poisoning investigated*

During the period of this inquiry there were four outbreaks of diarrhoea reported from the wards of the hospital and in all four cold chicken was the suspected food. Two other incidents, involving cold chicken, are also described.

##### *Outbreak 1*

This incident was the first to confirm the suspicion held by many of the hospital staff that cold chicken might be the source of food poisoning in the hospital—a suspicion which was strengthened when the results of my investigations of the hospital food were known.

On the afternoon of 24 November 1955 a sample of cold chicken was collected from the hospital main kitchen and after removing a sample for laboratory examination, the remainder was divided up and eaten by four members of the laboratory staff. Two of these had gastro-intestinal disturbances within 12 hr. of eating the chicken; the other two had no ill effects. The signs and symptoms of those affected were in accordance with those of *Cl. welchii* food poisoning. The only food eaten in common was the chicken from the hospital kitchen. Faecal specimens were not made available.

Bacteriological examination of the sample taken yielded almost pure cultures of a haemolytic *Cl. welchii*.

##### *Outbreak 2*

On 19 January 1956 an incident, involving three patients and a ward orderly, was reported in one of the wards in the hospital. The previous day the three patients had eaten a lunchtime meal consisting of cold chicken, the remains of which were consumed by the ward orderly. All four people had abdominal pain and diarrhoea but had recovered within 24 hr. No specimens of stool or food was available for examination.

Due to insufficient details, no definite conclusions can be drawn from this incident, but the clinical features were such that it could have been a small outbreak of *Cl. welchii* food poisoning caused by the ingestion of cold chicken.

##### *Outbreak 3*

Two of six patients in one ward who ate chicken for lunch on 2 February 1956 suffered symptoms typical of *Cl. welchii* food poisoning 10 hr later. The faeces of both the affected patients yielded non-haemolytic heat-resistant *Cl. welchii* of serotype 3. No sample of chicken was available for examination.

##### *Outbreak 4*

This outbreak was reported on 31 August 1956 and involved three patients in one ward. These patients had eaten chicken on the previous day and suffered

abdominal discomfort and diarrhoea some 8–12 hr. later. The diarrhoea continued throughout the first day but all the patients were well by the next day.

Specimens of faeces from the three patients were received on 31 August. A non-haemolytic heat-resistant *Cl. welchii* of serotype 4 was isolated from all three patients and this was presumably the cause of the outbreak.

#### *Outbreak 5*

This outbreak was well documented, thanks to the clinical co-operation of Dr M. McNicol, formerly a member of this department and, at the relevant time, a senior house officer in the hospital concerned. A report of this outbreak has been published (McNicol & McKillop, 1958).

Nine patients in one ward, together with a maid attached to the ward, developed gastro-enteritis, the signs and symptoms of which were strongly suggestive of *Cl. welchii* food-poisoning. The only article of diet common to the patients and the ward maid was cold chicken; the maid had eaten the remains of the chicken left over from the patients' lunch. Rectal swabs were taken from all ten persons and from eight of the specimens heat-resistant non-haemolytic *Cl. welchii* were isolated.

An outbreak of diarrhoea was reported from another ward on the same day and specimens of stool were received from six of the patients involved, all of whom had eaten chicken for lunch on the same day. The chicken came from the same cooking-batch as that consumed by the ten persons referred to above. Heat-resistant non-haemolytic *Cl. welchii* were isolated from all six specimens so that out of a total of sixteen persons suffering from diarrhoea following a meal prepared from the same batch of chicken fourteen were shown to have heat-resistant non-haemolytic *Cl. welchii* in their stools.

Subcultures of the organisms isolated were examined by Dr Betty Hobbs but they proved to be untypable by her range of typing-sera. I therefore immunized a rabbit with a killed suspension of one of the strains isolated and found that the serum from this animal not only agglutinated its homologous suspension but also suspensions of 11/13 of the other strains of *Cl. welchii*. The two that failed to agglutinate appeared to belong to a different serotype. The fact that out of sixteen individuals with diarrhoea, apparently attributable to a common article of diet, twelve were excreting precisely the same strain of organism is highly significant; it is unfortunate that specimens of the incriminated chicken were not available for examination.

#### *Outbreak 6*

On the evening of 22 September 1957, while visiting an elderly lady, the son of a member of the staff ate a meal of cold chicken. Some 8 hr. later he had colic with diarrhoea which persisted with lessened intensity throughout 23 September, but by the next day he had completely recovered. Two specimens of faeces were received on 23 September and also samples of the chicken and the broth from the cooking; the broth when it arrived in the laboratory showed marked gas production.

A direct smear of the broth and of the chicken showed numerous Gram-positive

bacilli with the typical morphology of *Cl. welchii*. Cultural examination of both specimens resulted in heavy and almost pure growths of haemolytic *Cl. welchii*. The two faecal specimens from the patient also gave good growths of haemolytic *Cl. welchii*; the coliform flora appeared to be normal and other pathogenic organisms were not found in any of the cultures.

Antisera were prepared against one of the chicken and one of the faecal strains, respectively, and cross agglutination tests confirmed the single identity of all the strains. There would seem, therefore, clear evidence in this case that the *Cl. welchii* contaminating the cold chicken was the cause of the colic and diarrhoea.

#### DISCUSSION AND CONCLUSIONS

The food, as purchased by a particular hospital in Glasgow, was found to be relatively free from potentially pathogenic organisms. It is true that some of the samples contained small numbers of *Staph. aureus* and *Cl. welchii*, but there was no evidence of gross contamination and such food could not be regarded as an obvious cause of food poisoning or other food-borne infections.

After the food had been cooked in the hospital kitchens and prepared for serving to the patients the number of contaminating organisms was generally much reduced. The presence of small numbers of *Staph. aureus* and *Cl. welchii* was more common in the foods which received considerable handling after they had been cooked than those which had not been so handled. These observations suggest that in most cases the post-cooking contamination was light and superficial, but in one particular article of diet—as served to the patients—heavy contamination was frequently found. In 10/46 samples of cold chicken examined large numbers of *Cl. welchii* were present. Whether or not the chickens had been contaminated before or after cooking, it seemed clear that there had been considerable multiplication of this organism during the period between cooking and serving the chicken from the main hospital kitchens.

Although all thirteen samples of uncooked chickens which I examined contained *Cl. welchii* in only one of these samples was a heat-resistant strain identified. The period of cooking, at the temperatures recorded in the experiments with the thermocouple, would have been insufficient to ensure the death of highly resistant strains but would be adequate to deal with strains of lower heat-resistance. Indeed, subsequent experiments showed that both the non-heat-resistant and the mildly heat-resistant strains were killed by the cooking. Some of the non-haemolytic strains found on the cooked chicken may have been vegetative forms derived from heat-resistant spores, but the high rate of contamination seems unlikely to have been due to the survival of heat-resistant strains. Only one of the uncooked birds contained non-haemolytic heat-resistant organisms and the majority of all the strains isolated were haemolytic and not resistant to heat; the evidence indicates that nearly all the important contamination occurred after the cooking procedures were completed. The dust in the kitchens and annexes, and presumably the unprotected zinc bath used for cooling the cooked birds, was heavily contaminated with *Cl. welchii* and this seemed to be an obvious source of the infection.

There was no evidence from experiments that chicken broths were more favourable to the growth of *Cl. welchii* than other meat broths, but chicken was the only item in the hospital diet which was, as a general practice, left overnight in bulk to cool after cooking. It was high incidence and heavy contamination with *Cl. welchii* of the chickens from the main kitchens, as compared with the relatively low incidence in those from the special-diet kitchen, that led to an investigation of the full procedures followed in the two places. Although the methods employed in the two kitchens for the actual cooking were satisfactory and fundamentally similar the after treatment was very different. In the special-diet kitchen the removal of the birds from their broth immediately after cooking, their separation and storage on shallow trays during preliminary cooling and finally early refrigeration, until required for use, prevented any significant multiplication of contaminating organisms. In the main kitchen, however, the chickens were not separated after removal from the cooker; they were simply transferred *en masse* with their broth to the large open receptacle where they were allowed to remain for nearly 24 hr. without refrigeration or any form of protection in an environment heavily infected with *Cl. welchii*. While the post-cooking conditions in the main kitchen were favourable to both bacterial contamination and growth of the infecting organisms the methods employed in the special-diet kitchen reduced the chances of contamination and prevented the multiplication of any organisms which might have gained access to the chickens after cooking. In experiments where large numbers of *Cl. welchii* were deliberately introduced after cooking and the chicken then refrigerated there was no evidence of any multiplication.

It is one of the basic principles of modern kitchen hygiene that all foods not intended for consumption immediately after cooking should be cooled as rapidly as possible and then refrigerated until required for use. These principles are not always fully appreciated or, at any rate, still not widely practised. As pointed out by McClung (1945), even where there are full facilities of refrigeration proper arrangements for pre-refrigeration cooling are often lacking. The system adopted in the main kitchens of the hospital under consideration gave no opportunity for rapid cooling; it resulted in the maintenance of a dangerous temperature over a prolonged period. The draining of the broth from the birds and their wide separation on shallow trays would have effected more rapid cooling and done much to reduce the growth of contaminating organisms. Although perhaps not applicable to large-scale cooking, the use of a pressure-cooker would enable temperatures to be attained which would deal with heat-resistant strains of *Cl. welchii* and by keeping the lid closed the birds would remain sterile until required for serving.

During the period of the present investigation no outbreaks of food poisoning due to salmonella organisms or *Staph. aureus* were disclosed; all the outbreaks indicated an association with the consumption of cold chicken and the clinical picture suggested a *Cl. welchii*-type of infection. Inquiries showed that in addition to the four hospital outbreaks reported, cases of diarrhoea frequently occurred which the nursing and medical staff considered were related to the days when cold chicken was served.

The recognition of *Cl. welchii* as a cause of food poisoning in man is of compara-

tively recent origin. Knox & Macdonald (1943) and McClung (1945) reported outbreaks of enteritis in which food contaminated with *Cl. welchii* was suspected as the cause, but whether or not the organisms produced typical  $\beta$ -haemolysis was not indicated. Zeissler & Rassfeld-Sternberg (1949), described the isolation of a heat-resistant form of *Cl. welchii* from cases of *Enteritis necroticans* and Hobbs *et al.* (1953) published reports on outbreaks of food poisoning which were associated with the presence of non-haemolytic heat-resistant strains of *Cl. welchii* in the incriminated food and also in the gut of those affected. The latter authors also isolated haemolytic strains from food and affected patients in similar incidents, but in subsequent investigations attention has mostly been focused on the non-haemolytic heat-resistant types of *Cl. welchii*.

In the work reported here two of the outbreaks were considered to have been caused by haemolytic non-heat-resistant strains and there is good evidence for the acceptance of non-haemolytic heat-resistant strains as the cause of two of the others; the evidence in the remaining two cases is not conclusive. Although the investigations disclosed serious, but easily remediable faults in kitchen hygiene it would appear that the very wide and significant pollution of the air with both haemolytic and non-haemolytic strains of *Cl. welchii* suggest hazards that are beyond the scope of this paper (see Lowbury & Lilly, 1958); only the observance of the highest standards of kitchen hygiene can eliminate the possibility of food poisoning caused by contamination of the food with *Cl. welchii*.

#### SUMMARY

1. An examination of eighty-nine samples of uncooked and thirty-eight samples of cooked food purchased by one particular hospital showed that the purchased food was bacteriologically clean.
2. A similar examination of 173 samples of food after it was cooked and prepared for serving in the hospital kitchens showed that, with the exception of cold chicken, the bacterial flora was greatly reduced by cooking. Ten of forty-six samples of cold chicken, however, were contaminated with fairly large numbers of *Cl. welchii*.
3. An investigation into the cooking and handling of the fowls indicated that contamination of the cooked fowls with kitchen dust was a probable explanation for the presence of *Cl. welchii*.
4. Immediate refrigeration of the fowls, well separated on shallow trays, was shown to be a satisfactory method of preventing the growth of contaminating *Cl. welchii* to any dangerous extent.
5. Six outbreaks of food poisoning, in which there was an association between cold chicken and the clinical symptoms of *Cl. welchii* food poisoning, are reported and discussed.

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