

Salmonella isolation from hospital areas

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SUMMARY

Evidence of the presence of salmonellas in a paediatric ward, a special care baby unit, a maternity unit and a hospital kitchen was obtained by culture of sewer swabs, faeces and food samples. The survey was designed to cause as little administrative interference as possible. The technical aspects of the survey did not strain laboratory facilities. Minimal secondary spread of salmonella infection was experienced.

INTRODUCTION

In a previous communication, we noted areas in a bacteriological laboratory from which salmonellas could be isolated. We discussed these as potential sources of infection for laboratory personnel and noted dangers of cross-contamination of samples leading to false positive reports (Harvey, Price & Joynson, 1976). The current study is an extension of that investigation, but the emphasis here is on recording potential sources of salmonellosis for man. Samples investigated were foodstuffs from hospital kitchens and the canteen, sewer swabs from a ward in the paediatric unit, and faecal specimens from patients in the hospital occupational health centre, the special care baby unit and a maternity unit.

MATERIALS AND METHODS

The materials examined were faeces samples of patients and staff, sewer swabs (Moore 1948), chicken giblets and cooked ham slices prepared for buffet meals in the hospital canteen. Sewer swabs were first used in Cardiff, in 1955, to associate a butcher's shop with an outbreak of salmonellosis (Harvey & Price 1970). Since then, they have often been employed in the investigation of salmonella epidemiology. We wished to discover if this technique was of value in demonstrating salmonella excretion in a small population in the University Hospital of Wales. The area selected was a ward in the paediatric unit. The chicken giblets, obtained weekly by one of the authors from the hospital kitchens, were seldom used in meals except as a basis for gravy and stock. Liver, crop, heart and gizzard were represented in each pack of giblets. Ham slices, as eaten at lunch in the staff

canteen were examined for total colony counts for twelve months. These latter samples were not examined for salmonellas, but provided background information on the relation between bacterial content and season. Faeces samples from staff reporting with gastro-enteritis to the Occupational Health Centre of the Cardiff hospital over a period of a year were examined for intestinal pathogens. Faecal specimens were also cultured from pregnant women at the 36th week, or at term, in the Maternity Unit in Neath General Hospital and from infants in the Special Care Baby Unit in the University Hospital of Wales.

The methods of salmonella isolation were fairly conventional, but differed in detail as two hospital laboratories were involved in the study. Faeces samples and rectal swabs in Cardiff were plated on MacConkey agar, deoxycholate citrate agar and brilliant green MacConkey agar (Harvey, 1956; Harvey & Price, 1974). These specimens were also enriched for salmonellas in selenite F broth. Enrichment cultures were incubated at 37 °C for 24 h and plated on deoxycholate citrate agar and brilliant green MacConkey agar. In plating media, the concentration of agar is kept low enough to perform slide agglutination on colonies picked from the selective agar. This means that a salmonella can be presumptively diagnosed and reported 24–48 h after receipt of specimen. In Cardiff, we seldom check biochemistry on salmonella-like colonies unless we are working with material containing aberrant serotypes such as reptile faeces or imported crushed bone. Rectal swabs in Neath General Hospital were enriched in selenite F broth, incubated 18–24 h at 37 °C, plated to XLD agar (Oxoid CM 469). Selective agars were incubated at 37 °C for 18–24 h and examined for pathogens. Salmonellas isolated were confirmed by the Division of Enteric Pathogens of the Public Health Laboratory Service in London from both Laboratories.

Sewer swabs were covered with single-strength selenite F broth in their original containers (220 ml screw-capped jars) and incubated at 43 °C for 24 h (Harvey & Thomson, 1953). These enrichment cultures were plated on brilliant green MacConkey agar and deoxycholate citrate agar. The brilliant green plates, after 24 h incubation at 37 °C, were examined for salmonella colonies which were picked and identified. The growth on the deoxycholate citrate agar plates was rubbed off with a short swab and inoculated into a modified Craigie tube (Craigie, 1931; Tulloch, 1939; Harvey & Price, 1967). After incubation of the Craigie tubes at 37 °C for 18–24 h, the surface agar outside the inner tube was subcultured to brilliant green MacConkey agar. Selective agars, after incubation at 37 °C for 18–24 h, were examined for salmonella colonies.

Poultry samples were enriched in a modified Muller-Kauffmann tetrathionate broth and in selenite F broth. The fluid media were prepared by methods already described (Harvey & Price, 1974). Both enrichment cultures were incubated at 43 °C (Harvey & Thompson, 1953; Edel & Kampelmacher, 1969) and were plated after 24 h incubation on deoxycholate citrate agar and brilliant green MacConkey agar. The brilliant green agars, after 24 h incubation at 37 °C, were examined for salmonella colonies which were further identified. The deoxycholate citrate agar plates were examined by the selective motility technique used in the sewer swab examination (Harvey & Price, 1974).

Table 1. *Salmonella* isolation from hospital faecal samples 1974–1978

| Sample origin | Study period | Total samples | Positive samples | Serotypes |
|--|------------------------|---------------|------------------|---|
| Staff suffering from gastro-enteritis reporting to Occupational Health Centre, UHW | Jan–Dec. 1974 | 21 | 0 | — |
| Pregnant women maternity unit, Neath G. H. | Sept. 1976 – Aug. 1978 | 3538 | 9(0.25) | <i>S. agona</i> 2 <i>S. cubana</i> 1 <i>S. eimsbuettel</i> 1 <i>S. hadar</i> 1 <i>S. indiana</i> 1 <i>S. indiana</i> 1 <i>S. mbandaka</i> 1 <i>S. saintpaul</i> 1 <i>S. virchow</i> 1 |
| Special Care Baby Unit, UHW | Jan. 1974 – Sept. 1977 | 5393 | 4(0.07) | <i>S. infantis</i> 3 <i>S. typhimurium</i> |

Figures in parentheses are percentages of total samples. Figures following serotypes record number of times a serotype was isolated.

The ham slices were examined for total bacterial counts at 37 °C by the Miles and Misra technique (Miles & Misra, 1938).

For a period of twelve months, information on cases of gastro-enteritis in hospital staff was recorded and faecal samples examined for salmonellas.

RESULTS

The results are presented in tables 1–5. Table 1 records salmonellas cultured from faeces samples. Isolations were made from specimens taken in the maternity unit and the Special Care Baby unit but not from staff reporting to the Occupational Health Centre U.H.W. with gastro-enteritis. Table 2 records serotypes recovered from swabs placed in a shallow sewer serving a paediatric ward in U.H.W. The survey continued from 1972–1975 when complaints of drain blockage forced termination of the project. Serotypes characterized by asterisks were known to be present in patients in the ward during the investigation. Serotypes without asterisks are more interesting as their isolation suggests the presence of unknown excretors in the paediatric area. Note that all isolations in 1974 were serotypes not corresponding to known clinical salmonella infections. Table 3 gives information on salmonellas cultured from chicken giblets. Two hundred and twenty eight positive samples were found out of a total of 408. The use of two enrichment media increased the number of isolations but there was no significant advantage for either enrichment medium. Table 4 presents the serotypes isolated. Table 5 records the total counts on ham slices as served in the canteen. High counts are more frequently seen in the summer months.

Table 2. *Salmonella* isolations from gauze swabs placed in sewer in paediatric ward 1972-1975

| Period | Total Positive | | Serotypes isolated | | | |
|------------|----------------|--------|--------------------|--|--|--|
| | swabs | swabs | 1972 | 1973 | 1974 | 1975 |
| Jan.-Mar. | 58 | 8(14) | <i>S. agona</i> | <i>S. agona</i> * <i>S. infantitis</i> | <i>S. infantis</i> <i>S. derby</i> | <i>S. typhimurium</i> * <i>S. panama</i> * |
| April-June | 58 | 3(5) | | <i>S. paratyphi B</i> <i>S. java</i> | <i>S. virchow</i> <i>S. newington</i> | <i>S. heidelberg</i> * <i>S. friedenaui</i> * |
| July-Sept. | 51 | 13(26) | | <i>S. stanley</i> * <i>S. livingstone</i> | <i>S. agona</i> <i>S. typhimurium</i> | <i>S. agona</i> * <i>S. derby</i> |
| Oct.-Dec. | 62 | 19(31) | | <i>S. indiana</i> | | |

Figures in parentheses are percentages to the nearest integer of total swabs in period studied.

* Serotypes known to be infecting a clinical case in the area sampled. Other serotypes were not known to be present in the ward and would have remained undiscovered without sewer swab surveillance.

Table 3. *Salmonella* isolations from chicken giblets collected from hospital kitchen. Direct enrichment and selective motility enrichment results combined

| | |
|--|----------|
| Selenite F positive, Muller-Kauffmann tetrathionate positive | 106 |
| Selenite F positive, Muller-Kauffmann tetrathionate negative | 57 |
| Selenite F negative, Muller-Kauffmann tetrathionate positive | 65 |
| Selenite F negative, Muller-Kauffmann tetrathionate negative | 180 |
| Total samples | 408 |
| Positive samples | 228 (56) |

Figure in parentheses expresses positive results as integral percentage of total samples.

Table 4. *Salmonella* serotypes isolated from chicken giblets: crop and gizzard cultured separately from liver and heart

| Serotype | No. of isolations |
|--------------------------------|-------------------|
| <i>S. agona</i> | 30 |
| <i>S. bredeney</i> | 50 |
| <i>S. enteritidis</i> type 13 | 36 |
| <i>S. fischerkietz</i> | 12 |
| <i>S. heidelberg</i> | 23 |
| <i>S. infantis</i> | 9 |
| <i>S. kentucky</i> | 15 |
| <i>S. livingstone</i> | 11 |
| <i>S. montevideo</i> | 20 |
| <i>S. muenster</i> | 51 |
| <i>S. new-haw</i> | 11 |
| <i>S. senftenberg</i> | 2 |
| <i>S. sladun</i> | 5 |
| <i>S. typhimurium</i> RDNC (1) | 2 |
| <i>S. typhimurium</i> U247 | 2 |

Table 5. Counts on sliced ham in hospital canteen

| Month | Year | No. of samples with counts in each category | | | | | | | | | Total tests per Month | |
|-----------|------|---|------------------|------------------|------------------|------------------|------------------|------------------|------------------|------------------|-----------------------|------------------|
| | | < 10 ¹ | 10 ⁻¹ | 10 ⁻² | 10 ⁻³ | 10 ⁻⁴ | 10 ⁻⁵ | 10 ⁻⁶ | 10 ⁻⁷ | 10 ⁻⁸ | | 10 ⁻⁹ |
| April | 1974 | 2 | 4 | 1 | 5 | 2 | — | — | — | — | — | 14 |
| May | | — | — | 2 | 8 | — | 3 | 3 | — | — | — | 16 |
| June | | — | — | — | 1 | 7 | 4 | 3 | 5 | — | — | 20 |
| July | | — | — | — | 1 | — | — | 5 | 12 | 2 | — | 20 |
| August | | — | — | — | — | — | — | — | — | — | — | 0 |
| September | | — | — | — | — | 5 | — | 2 | 5 | 4 | — | 16 |
| October | | 1 | — | 2 | 8 | — | — | — | — | 2 | 4 | 17 |
| November | | — | — | 4 | 6 | — | — | — | 2 | 3 | — | 15 |
| December | | — | — | 2 | 9 | — | — | — | — | — | — | 11 |
| Total | | 3 | 4 | 11 | 38 | 14 | 7 | 13 | 24 | 11 | 4 | 129 |

Medium; Blood agar; incubation temperature; 37 °C; time of incubation, 24 h. Counts are expressed in organisms per g.

DISCUSSION

Some authorities consider routine screening of pregnant mothers for salmonella excretion to be impracticable and suggest that most laboratories would be unable to offer such a service and that the examination would be of poor quality and without significance (Lowbury *et al.* 1975). We certainly admit to administrative difficulties, but in our study neither hospital found it hard to conduct the technical aspects of the survey. We realize that the use of rectal swabs can be criticized (Thomas, 1954) but this is the most suitable sample for nursing staff and if the swabs are soiled with faeces they are culturally adequate. If an investigation is to continue over a prolonged period, which must be the case as the salmonella incidence is low, nursing convenience is of primary concern. The incidence found in the maternity series (0.25 %) is what would be expected in a study of symptomless normal persons (Taylor, 1960). It is possible, if a pre-enrichment technique was employed for examining rectal swabs similar to that used in food and water bacteriology (Edel & Kampelmacher, 1973; Harvey & Price, 1977) that the sensitivity of selective culture would be increased. Speed of reporting results would be diminished but improved collection of data might compensate for this.

The entry of a symptomless excreter of salmonellas into the maternity ward and special care baby unit was a relatively rare event (table 1). All 13 excreters would have remained undiscovered without routine laboratory surveillance. Little spread to other patients occurred, but this is not invariable. Salmonella outbreaks in babies in hospital wards were recorded in 1975 and in 1976 (Anon, 1975; 1977). In both incidents, wards had to be closed to further admissions. The 1975 outbreak was discovered by routine faecal screening but early diagnosis did not prevent ultimate ward closure. Infection of babies can have other consequences. Neonatal salmonella infection often involves prolonged excretion of the organism with secondary infection in the household after discharge of mother and baby from hospital (Rubinstein, Femmster & Smith, 1944; Rubinstein & Fowler, 1955; Szanton, 1957).

Salmonella surveillance in a hospital is possible using indirect methods (table 2). Discovery of an excreter is not obtained by sewer swab examination but the method

has been of value in the past in relating staff excretion and contamination to infected food ingredients (Harvey, 1957). The current study in the paediatric ward demonstrated probably entry of unknown salmonella excreters into a sensitive area. Twelve serotypes were isolated from paediatric sewer swabs which could not be accounted for by serotypes known to be infecting patients in the ward investigated. Multiple salmonella serotypes are infrequently excreted by man in this country and it is therefore likely that these 12 serotypes represent undiscovered salmonella excreters in patients or staff. Only one ward in the department was investigated. The percentage of sewer swabs containing salmonellas showed a seasonal incidence, the greatest number of positive isolations occurring between July and December. Indirect surveillance does not disturb patients or staff but problems of blocked drains were sometimes experienced, and the survey had to be abandoned in 1975. The 'wipe' swab technique described by Harvey & Phillips (1955) could be used. This was devised for drains where rats caused removal of swabs before collection by the Environmental Health Officer. The 'Wipe' swab method is more akin to a grab sample than the swab which is left for seven days in a sewer and might be less efficient in demonstrating the presence of salmonellas.

Chicken giblet contamination is only a guide to the presence of salmonellas in the hospital kitchen. Examination of rinse and drip fluid from frozen poultry might be a more accurate means of estimating carcass contamination (van Schothorst *et al.* 1976). The giblet sample is, however, easy for laboratory staff to collect and avoids interfering with kitchen routine. Salmonella contamination of giblets is necessary knowledge for kitchen staff, as 56% were found to contain these organisms. Fortunately, danger of contamination of cooked foods was slight. Two different enrichment media were used for isolation but there was no significant difference, in this series, between selenite and Muller-Kauffmann tetrathionate broths. Pre-enrichment was not used in this study but is now incorporated in our routine examination of food and water for salmonellas (van Schothorst *et al.* 1978; Harvey & Price, 1977).

Buffet meals are often served in the canteen at the Cardiff hospital. We have no figures for bacterial counts on cooked cold chicken but, in 1974, total colony counts were made on cold ham slices to obtain background information on contamination (table 5). The association between season of the year and magnitude of total count is evident and suggests bacterial multiplication in the summer months. The circumstances under which cold ham and chicken salads are presented in the canteen are similar and ham counts may be taken as representative of the bacterial content of cold meats in general. A recent study on cooked cold chicken kept in a domestic refrigerator whose temperature varied between 2 and 13 °C shows the extent of bacterial multiplication that can take place at relatively low temperatures (Toule & Murphy, 1978).

Data from the occupational health centre in the University Hospital of Wales did not reveal any salmonella infections (table 1). It was hoped to correlate infection in staff with infection in food from the hospital kitchen. In the years 1974-1978, no incidents of salmonellosis in staff could be confidently attributed to hospital catering.

We have attempted in this study to examine some areas in hospitals where salmonellas were likely to be found or where spread of salmonella infection could have serious consequences. In the Neath Hospital series isolation of infected mothers with their babies was practised and only one neonate developed salmonellosis. We feel that monitoring of maternity units for salmonellas is of particular value in prophylaxis, does not strain laboratory facilities and can be integrated into the routine of certain hospitals. If the epidemiology of hospital salmonella infection is worth studying, we think that routine surveillance of selected areas can be rewarding as an alternative to retrospective investigation of outbreaks.

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