

## THE BACTERIOLOGY OF GASTROENTERITIS IN CHILDREN BELOW THE AGE OF TWO YEARS

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The gastroenteritis syndrome of diarrhoea and vomiting is a major problem in child health. Singapore has an annual infant mortality of 440 to 547 due to this syndrome representing 7.1 to 8.6 deaths per 1,000 live births. Thailand loses more than 3,000 children below the age of one from this cause, and Philippines more than 8,000 (W.H.O. 1957, 1958, 1959). The Federation of Malaya shows a mortality of between 400-600 for the corresponding period. Although these figures are striking in themselves, they reflect only a small proportion of the actual morbidity rate. Our estimates from local experience indicate that out of the average annual admission of 500 gastroenteritis cases into the General Hospital, Penang (1961-1963: admissions 2020) there was a 10% fatality (1960-1963: deaths 192). The magnitude of this problem in terms of morbidity and care needed is self-evident.

The long term study is aimed at elucidating the role of bacteria in the etiology of this syndrome. This is not only of academic interest, because, should there be a definable bacterial pattern, then the choice of an appropriate antimicrobial therapy would also become important in addition to the usual supportive measures. In this respect, the (Great Britain) Medical Research Council (1953) pointed out that Chloramphenicol and Sulphadiazine had significant therapeutic benefit in these cases although *Shigella* or *Salmonella* had not been demonstrated in the stools. Further, it is essential to establish what the local conditions are, as in this field of investigation, the reported findings vary from region to region.

### Material and Methods

All cases of gastroenteritis in the age group studied during the survey period (April 1960 to December 1962) had samples of stools examined on admission. They were either collected without preservative or where some delay was likely, as rectal swabs in glycerol-

saline. The latter were then despatched to the laboratory at the earliest opportunity — not later than 24 hours after collection. On arrival they were plated out on MacConkey No. 3 medium, Salmonella — Shigella agar, Desoxycholate Citrate Agar (all "Oxoid" products) and blood agar plates. In addition, Selenite broth and Robertson's cooked meat medium (with added 10% sodium chloride) were inoculated at the same time and incubated overnight, before a subculture was made on further MacConkey and blood plates respectively. All plates were read after overnight incubation and pathogens identified by standard bacteriological procedures. (Table 1). Where two suspected pathogens were simultaneously isolated, they were reported together.

### Results

The results of this work covering a period of nearly three years and involving some 1,700 specimens are seen in Table 2.

Among the generally recognised pathogens, the largest group is formed by the *Shigella* species — 4.9% of the total number of stools examined. Of these *Sh. flexner* is the most common (Table 3). It is associated with coagulase-positive staphylococci in about a quarter of the cases. A double infection with other pathogens was not found. The pattern of occurrence of these organisms suggests an endemic incidence in the population, with occasional exacerbations as at the end of 1962 (see figure).

*SALMONELLA* as a cause of diarrhoea and vomiting appears to be rare, only three isolations being made during the period under review. *Salmonella typhimurium* was responsible for one case, a Group C1 salmonella in another and a non-typeable (group A to E4) salmonella in the third.

*ESCHERICHIA COLI* of previously reported pathogenic sero-types were found in

TABLE 1

Organisms	Preliminary Isolation	Preliminary Identification	Confirmation
Salmonella	SS agar, DCA or MacConkey No. 3 — direct, and from enrichment media	Kligler Iron agar, Urea agar, Serology	Sugar reactions, Citrate utilisation, Motility Serotyping
Shigella			
Escherichia	Blood agar MacConkey No. 3	Serology	Citrate utilisation, Serotyping Heat test
Pseudomonas	Blood agar	Pigmentation	—
Proteus	Blood agar	Colonial characteristics	Urea agar
Candida	Blood agar	Colonial characteristics	Microscopy
Staphylococci	Blood agar from RCM with 10% NaCl	Colonial characteristics	Coagulase test

Summary of Bacteriological Techniques used in Identification of Organisms in Stools.

TABLE 2

The positive findings in stools of young children with Gastroenteritis.

Year	1960	1961	1962	Total	Per cent
Specimens examined	557	549	597	1703	—
Salmonella	1	2	0	3	0.02
Shigella	16	11	35	62	3.64
+ Staph.	2	8	11	21	1.24
Escherichia	20	12	14	46	2.70
+ Staph.	8	11	11	30	1.76
+ Proteus	2	0	0	2	0.01
Proteus	29	19	17	65	3.82
+ Staph.	14	13	1	28	1.64
Candida	4	1	14	19	1.12
+ Staph.	0	0	1	1	0.01
Pseudomonas	0	1	8	9	0.53
+ Staph.	1	0	5	6	0.35
Staphylococci	306	243	204	653	38.20
Number positive	303	321	321	945	55.04

4.5% of cases. The relative distributions of the serotypes isolated are shown in Table 4. Again, the incidence distribution for the various months of the year (see figure) suggests an endemic occurrence. Whether their presence is of etiologic significance, it is not possible to conclude definitely as the pattern is unlike that in epidemic outbreaks where the circumstantial evidence is strong and suggestive.

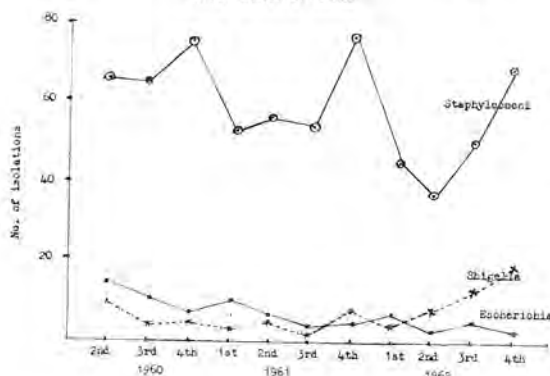
TABLE 4

Serotype	Alone	With Staphylococci	Total	Per cent.
0.119	26	14	40	52.5
0.127	8	6	14	18.4
0.55	7	5	12	15.8
0.26	2	1	3	3.9
0.111	1	1	2	2.6
0.128	1	0	1	1.6
0.126	1	1	2	2.6
0.125	0	2	2	2.6

#### Frequency of Isolation of *Escherichia* Serotypes.

An interesting finding is the comparatively large number of cases — 38.2% — where coagulase-positive staphylococci were present

Seasonal Isolations of Staphylococci, *Shigella* and *Escherichia*



TRIMESTERS OF THE YEAR

Trimesters of the year	1960				1961				1962			
	2nd	3rd	4th	1st	2nd	3rd	4th	1st	2nd	3rd	4th	
Staphylococci	65	65	76	53	57	55	78	46	38	51	70	
Esch. coli	14	11	8	11	7	3	5	7	3	6	3	
Shigella	9	4	5	3	5	2	9	4	9	13	20	

as the only suspected pathogen. In another 5% of cases, it is associated with other suspected enteropathogenic bacteria. This problem will be discussed in detail in a following article (Chan and Lucas, 1964).

The other presumptive pathogens include *Proteus* species — 3.5%; *Candida* — 1.1% and *Pseudomonas Pyocyanea* — 0.9%, in order of their frequency of isolation.

#### Discussion

Different workers in their examination of stools of young children suffering from gastroenteritis have incriminated different bacteria. Although there is strong evidence for certain epidemics due to *Salmonella*, *Shigella* and *Escherichia coli* the findings in different parts of the world in endemic and sporadic cases over an adequately observed period are far from uniform (Taylor, 1960; Ordway, 1960). Part of this discrepancy might be due to choice of investigative techniques or case selection. Moreover, little is known of the pattern in the tropics. Luder (1959) in a report involving two separate series of 100 cases each from Uganda, found an overall incidence of 12.5% *Shigella*, 3.0% *Salmonella* and 1.5% *Escherichia coli*. The majority of his cases had no incriminating pathogen and he suggests parenteral infection and malaria as possible causes. This latter suggestion is difficult to accept on the available data.

*SALMONELLA* AND *SHIGELLA*, the commonly recognised entero-pathogenic bacteria, were found in less than 5% of all our cases. Although asymptomatic infection can occur with these organisms (Floyd et al., 1956) and even more so when the frequency of isolation suggests an endemic occurrence as in this study, it has been pointed out by Watt and Hardy (1945) that in the particular age group studied, the ratio of clinical disease to infection by *Shigella* is high. The causal rela-

tionship therefore, appears to be probable. In about 25% of the cases of *Shigella* infection, coagulase-positive staphylococci were also found. The significance of their relative roles is difficult to assess — it might be that they have a pathogenic synergism or that staphylococcal colonisation in the gut predisposes to clinical *Shigella* infection.

Specific pathogenic serotypes of *Escherichia coli* have been associated with epidemics of infantile diarrhoea (Hodes, 1956) and their role in these outbreaks has been accepted. However, their part in sporadic and endemic diarrhoea and vomiting cannot be said to be as firmly established. The isolations in our series of particular serotypes have been erratic and no specific epidemiological pattern is present. Although we cannot dismiss them as unimportant, to be unduly enthusiastic about their pathogenic relationship is also unwarranted on the available data. The most frequently isolated serotypes are 0119, 0127 and 055. The total incidence of 4.5% in our series compares with that of the United States but is much lower than other countries. For example, England is reported to have an overall incidence of 20.4%, Poland 36% and France 52.6% (Taylor, 1960).

The rest of the organisms we have presented as presumptive pathogens — because they are not generally recognised as causing diarrhoea and vomiting — form an interesting category as the bacterial growth pattern in these cases presents a striking departure from that of the normal intestinal flora. These organisms are present in such overwhelming numbers that they form an almost pure culture. Epidemics of diarrhoea due to *Pseudomonas aeruginosa* (Hunter and Ensign, 1947)

TABLE 3

Species	Alone	With Staphylococci	Total	Per cent.
<i>Sh. dysenteriae</i>	1	0	1	1.2
<i>Sh. boyd</i>	1	0	1	1.2
<i>Sh. flexner</i>	51	19	70	84.3
<i>Sh. sonnei</i>	9	2	11	13.3

Frequency of isolation of various shigella species.

and *Proteus mirabilis* (Pintelon, 1962) have been recorded and thrush enteritis is a feared complication of oral antibiotic therapy. The position, thus, of *proteus*, *candida* and *Pseudomonas* organisms as reported in this study, has to be considered seriously as one of the possible etiological agents.

In reviewing the comprehensive reports of investigations into the etiology of infantile gastro-enteritis, we are impressed by the large proportion of cases, where the etiology of the syndrome escapes the accepted laboratory methods of investigation for enterobacteria commonly incriminated in gastro-intestinal disease. (Walker et al., 1960, Linetskaya-Novorodskaya, 1959; Taylor, 1960) Neither were viral studies particularly enlightening (Light and Hodes, 1949, Walker et al., 1960). Our experiences with standard techniques have also led to the same conclusion. However, with selective methods, the finding of coagulase-positive staphylococci in about 40% of all cases has suggested a possible role of this organism (Chan and Lucas, 1964), either as a primary casual agent or a predisposing factor for hitherto undetected causes. The problem of this finding is dealt with more fully in a following article.

The concept of "Parenteral" diarrhoea as a distinct entity cannot be accepted without more convincing data, although general debility is no doubt an important contributory factor in production of clinical disease in the infected host (Taylor, 1960).

From the therapeutic point of view, unless there are strong valid objections, it would appear that exhibition of an antibiotic sufficient to meet at least all the suspected bacteria is a rational approach to the management of gastroenteritis in young children. This should be given in conjunction with the usual supportive measures to combat dehydration and acid-base imbalance.

### Summary

A bacteriological investigation of the stools of 1703 cases of gastroenteritis revealed the presence of the following: Coagulase-positive staphylococci 38.2%, *Shigella* 4.9%, Pathogenic serotypes *Escherichia coli* 4.5% *Proteus* 5.5%, *candida* 1.1%, *Ps. Pyocyanea* 0.9% and

**Salmonella** 0.02%. The role of these organisms is discussed and specific antimicrobial therapy suggested as part of the management of the cases.

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