BLOOD CULTURE ISOLATES FROM CHILDREN ADMITTED TO MEDICAL UNIT III, YANGON CHILDREN'S HOSPITAL, 1998

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Abstract. A one year study (August 1998-July 1999) of bacteremia in febrile children was carried out in the Medical Unit (III), Yangon Children's Hospital (YCH), Myanmar, to determine: (1) the bacteria responsible for fever of five days or more in children; (2) the antibiotic sensitivity pattern of these bacteria. Children aged one month to 12 years who had fever for five days or more and who did not receive antibiotics within the first 48 hours irrespective of the diagnosis were included in this study. A total of 120 patients fulfilled the criteria. Bacteria could be isolated from 65 cases (54.2%). The commonest organism isolated was *Salmonella typhi* (43.1%). Others included *Escherichia coli* (12.3%), *Staphylococcus aureus* (7.7%), *Pseudomonas aeruginosa* (7.7%); *Streptococcus, Shigella, Diplococcus, Klebsiella* and *Acinetobacter* were also isolated. The *Salmonella typhi* were resistant to conventional antibiotics (ampicillin, amoxicillin, chloramphenicol, and co-trimoxazole); however, they were sensitive to amikacin, netilmicin, nalidixic acid, and cephalothin. A cluster of enteric fever cases from Mingalartaungnyunt township was noticed and was reported to the Directorate of Health. Changes in the incidence and etiology of bacteremia in hospitals are well documented. Sentinel surveys of bacteremia in major hospitals should be carried out in order to detect the changing patterns of bacteremia and antibiotic sensitivity; such surveys will be of great help in establishing local antibiotic policies.

INTRODUCTION

Bacteremia is often a life-treatening and fatal condition that necessitates immediate treatment. For an accurate diagnosis and an appropriate choice of antibiotics, blood culture, which usually takes a few days, is required. The empirical choice of antibiotics for the treatment of bacteremia is guided by an awareness of previous culture reports. Up-to-date information on the local etiologic patterns and antibiotic sensitivities is also important.

There are several reports on blood culture isolates from various countries (Maiztegui *et al*, 1965; Watt and Okubadejo, 1967; Jansson, 1971; Mycrowitz *et al*, 1971; Cronberg *et al*, 1974). Gedegou *et al* (1984) reported that of 408 bacterial strains *Salmonella* were most often isolated (23%), followed by *Staphylococcus aureus*, *Acinetobacter-Moraxella* (12.0%), *Escherichia coli* (9.0%), and a miscellaneous group of less common organisms. The Gram-negatives constitute 67%, of which most were *Enterobacteriaceae*, suggesting that proportions of both the Gram-negative and -positive isolates represent contaminants in blood. Cheng *et al* (1991) found, in a five year prospective study in Hong Kong, 344 clinically significant episodes of pediatric septicemia: the commonest organisms were *Salmonella* spp (15.0%), followed by methicillin-resistant *Staphylococcus aureus*; they reported that *Haemophilus influenzae* accounted for 2.0% of all episodes; there were no cases of men-

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ingococcal septicemia. *H. influenzae* and *N. meningitidis* usually cause 10-20% and 5-7% respectively of all pediatric septicemias in Japan and the West (McCarthy *et al*, 1976; Winchester *et al*, 1977). These different studies show that the pattern of pediatric septicemia varies regionally. High mortality rates in cases of septicemia due to methicillin-resistant *S. aureus* (MRSA) and treatment with ineffective antibiotics were also recorded (French *et al*, 1988).

Changes in the incidence and etiology of bacteremia in hospitals is well documented (Watt and Okubadejo, 1967; Shah and Watanakunakorn, 1979). The antimicrobial resistance of *Salmonella* (Gedebou and Tassew, 1981), *Klebsiella* (Gedebou, 1982), *Shigella* (Gedebou and Tassew, 1982) is quite alarming. Knowledge of the sensitivity of bacterial strains cannot be the sole factor for the rational prophylactic or therapeutic use of antibiotics, it is certainly essential for the appropriate choice of antibiotics. The aim of this study is the determination of the bacterial agents responsible for fever of five days or more in children admitted to Yangon Children's Hospital (YCH); the antibiotic susceptibilities of the isolated organisms are also examined.

MATERIALS AND METHODS

Case selection

All children admitted to Medical Unit III, YCH, between August 1998 and July 1999 that fulfilled the following criteria were included in the study.

Inclusion criteria: Children aged one month to 12 years with a fever for five days or more (irrespective of the initial diagnosis) and no history of antibiotic use during the 48 hours prior to enrolment.

Exclusion criterion: Children who were on antibiotics (either oral or parenteral) during the 48 hours prior to the time of enrolment.

Blood specimens and primary inoculation

Blood specimens (5 ml) were collected in blood culture bottles that contained 50 ml of tryptose phosphate broth and 0.02% polyanethol sulfonate (liquoid) following standard aseptic procedure. The inoculated culture bottle was immediately placed in an incubator (37ºC) before forwarding to the Bacteriology Research Division, Department of Medical Research (DMR).

Isolation and identification

Based on the Gram-staining characteristics of the bacteria, growth in the blood culture bottles was subcultured onto MacConkey agar, Salmonella-Shigella agar, Nutrient agar and/or blood agar and/or chocolate agar plates. Following incubation, colonies were further characterized by biochemical and serological mehthods (Ewing, 1986).

Widal test and clot culture

Blood specimens (usually a single specimen, but sometimes two were sent to the National Health Laboratory for Widal test and clot culture if indicated. The Widal test was considered positive if either the O or the H agglutinin level was equal to or more than 1:160.

Antibiotic susceptibility testing

All isolates were tested for their susceptibilities by using antibiotic discs form Oxoid (Oxoid Unipath Limited Basingstoke, Hampshire, England) as described by the method of Bauer *et al* (1966).

RESULTS

A total of 120 blood samples were collected from children with different diagnoses. A large majority of cases presented with pyrexia of unknown origin (PUO; 53.3%) and acute viral infections (AVI; 25.8%) (Table 1). The age and sex distributions of the study population are shown in Table 2. A total of 65 isolates (54%) were recovered. The commonest organism isolated was *Salmonella typhi* (43.1%); others included *Escherichai coli* (12.3%), *Staphylococcus aureus* (7.7%) and *Pseudomonas aeruginosa* (9.2%) (Table 3).

Table 1
Admission diagnoses.

Sr No.	Admission diagnosis	Number of cases	Percentage
1	Pyrexia of unknown origin (PUO)	64	53.3
2	Acute viral infection (AVI)	31	25.8
3	Rheumatic fever/congenital heart diseases	9	7.5
4	Acute respiratory infections	6	5.0
5	Malaria	3	2.5
6	Meningitis/encephalitis	2	1.7
7	Cellulitis/multiple pyemic diseases	2	1.7
8	Enteric fever	1	0.8
9	Others	2	1.7
	Total	120	100

Table 2
Age and sex distributions.

Age	Male	Female	Total	Percentage
1 month to 1 year	5	7	12	10.6
>1 year to 5 years	20	26	46	39.4
>5 year to 12 years	29	33	62	50.0
Total cases	54	66	120	100

Table 3
Organisms isolated from blood culture.

Organisms	Number of cases	Percentage
Salmonella typhi	28	43.1
Escherichia coli	8	12.3
Staphylococcus aureus	5	7.7
Pseudomonas aeruginosa	6	9.2
Streptococcus sp	4	6.2
Staphylococcus epidermidis	5	7.7
Gram-negative bacilli	2	3.1
Shigella boydii	2	3.1
Streptococcus faecalis	2	3.1
Acinetobacter sp	1	1.5
Diplococcus sp	1	1.5
Klebsiella aerogenes	1	1.5
Total	65	100

Duration of fever did not influence the recovery of the isolates (Table 4). Isolated bacterial pathogens with respect to admission diagnosis

Table 4
Day of blood culture taken and culture positivity.

Day of fever when blood culture taken	Culture positive (%)	Culture negative (%)	Total
5-10 days	26 (55.3)	21 (44.7)	47
11-15 days	21 (58.3)	15 (41.9)	36
16 or more days	19 (51.3)	18 (48.7)	37
	66	54	120

$\chi^2 = 0.36$; $p = 0.83$

is shown in Table 5. A number of other bacteria were also isolated.

Three isolates, *Staphylococcus, Diplococcus,* and *Pseudomonas aeruginosa* could be obtained from children with heart diseases *P. aeruginosa* was isolated from three cases of AVL and one case of PUO. Bacteremia could be demonstrated in two of the three children

Table 5
Bacterial pathogens isolated from the study population.

Admission diagnosis	*S. typhi*	*E. coli*	Gm(-)	*S. faecalis*	*P. aeru.*	*S. aureus*	*S. epider*	*Sh. boy*	*Strep.* sp	*Kleb.*	*Deplo.*	*Actino.*
PUO (n=64)	17	4	1	1	1	1	4	1	3	1	0	1
Acute viral infection (AVI) (n=31)	10	2	0	1	3	1	1	1	0	0	0	0
Rheumatic fever/con. heart dis. (n=9)	0	0	0	0	1	0	0	0	1	0	1	0
Acute respiratory infection (n=6)	0	0	0	0	1	1	0	0	0	0	0	0
Malaria (n=3)	0	1	1	0	0	0	0	0	0	0	0	0
Meningitis/encephalitis (n=2)	0	1	0	0	0	1	0	0	0	0	0	0
Cellulitis/Multiple pyemic abscesses (n=2)	0	0	0	0	0	1	0	0	0	0	0	0
Others (n=2)	0	0	0	0	0	0	0	0	0	0	0	0
Enteric fever (n=1)	1	0	0	0	0	0	0	0	0	0	0	0
Total = 65/120	28	8	2	2	6	5	5	2	4	1	1	1

clinically diagnosed as having malaria; blood for malaria parasites was negative in all the cases. *Escherichai coli* was isolated from a 5-month-old child with meningitis. Four children died; the causes of death were: malignant lymphoma, multiple lung abscesses, and congenital heart disease with heart failure. In one case, the cause of death could not be determined. Bacteria were not isolated in all these cases.

The salient clinical features of typhoid fever are shown in Table 6. The age distribution of *Salmonella typhi* culture-positive children was mapped. No *Salmonella* organisms nor a positive Widal test could be obtained from children under two years of age. The youngest child in whom *Salmonella typhi* was isolated was two years and two months old. Seventy-three percent of typhoid fever cases were children of more than five years of age. In this study, 45.9% of the children with typhoid fever were both blood culture and Widal positive; 29.7% of the children were Widal negative but culture positive and 24.3% were Widal positive but culture negative (Table 7). The Widal test was found to be positive in four children whose blood culture isolates were organisms other than *Salmonella typhi* (9.7% false positive). Clot cultures were found to be positive in four patients: three Widal-positive and one Widal-negative (Table 8).

The antibiotic sensitivity of *Salmonella typhi* revealed that the organisms was resistant to conventional antibiotics, *eg* ampicillin, amoxacillin, chloramphenicol, and co-trimoxazole. However, it was sensitive to amikacin, netilmicin, nalidixic acid, and cephalothin (Table 9). Table 10 shows the localities from which the typhoid fever culture positives came; more than 39% came from Mingalartaungnyunt.

DISCUSSION

As in other studies, the commonest organism isolated was *Salmonella*. *Salmonella typhi* was isolated from 23.3% of the study population: comparable with the 17% prevalence

Table 6
Salient clinical features of children with typhoid fever.

Clinical features	Number	Percentage
Relative bradycardia	11	29.7
Hepatomegaly	27	72.9
Splenomegaly	8	21.6
Lung signs	5	13.5
Coated tongue	2	5.4
Total	37	100

reported from the same hospital (YCM) by Saw-Aung-Myat-Thein (1979). Both blood culture positive and Widal positivity could be demonstrated in 45.9% of the typhoid cases. However, in 39% (11/28) only the blood culture was positive (false Widal negative) if the cut-off points for typhoid agglutinins was set at 1:160. If the cut-off level was reduced to 1:80, then the proportion of false negatives was reduced to 32%. Different studies have suggested a variety of cut-off levels for O and H agglutinins: 1:40 (Chow *et al*, 1987); 1:50

Table 7
Age distribution of typhoid fever cases.

Age	Culture positive only	Widal positive only	Both culture and Widal positive	Total
1 month to 1 year 11 months	nil	nil	nil	nil
2-4 years	3	1	6	10
5-12 years	8	8	11	27
Total	11 (29.7)	9 (24.3)	17 (45.9)	37

Note: Figures in parentheses are percentages.

Table 8
Antibiotic sensitivity of common organisms.

Antibiotics	*S. typhi*	*S. aureus*	*P. aeruginosa*	*E. coli*
Ampicillin	14.3	50.0	0	25.0
Amoxacillin	21.4	75.0	0	20.0
Chloramphenicol	7.1	50.0	20.0	12.5
Co-trimoxazole	7.1	100	16.7	37.5
Amikacin	96.4	100	100	100
Netilmicin	100	100	100	100
Nalidixic acid	100	NT	75.0	83.3
Cephalothin	89.3	100	0	16.7
Norfloxacin	100	100	100	100
Gentamicin	100	100	100	85.7
Streptomycin	3.6	75.0	16.7	12.5
Ceftriaxone	100	100	33.3	62.5
Carbenicillin	28.6	100	50.0	87.5
Furazolidone	89.3	100	0	25.0
Cefaclor	90.0	100	0	100
Tetracycline	14.3	33.3	50.0	87.5

Note: all figures are percentages.

Table 9
Locality of the children with typhoid.

Sr No	Locality	Number of cases	Percentage
1	Mingalartaungnyunt	11	39.3
2	Thingangyun	3	10.7
3	Tharkayta	2	7.1
4	Dalla	2	7.1
5	Kyauktada	2	7.1
6	Kyaukmyaung	1	3.6
7	Dawbone	1	3.6
8	Tarmwe	1	3.6
9	Hlaing	1	3.6
10	Hlaingtharyar	1	3.6
11	N. Okkalapa	1	3.6
12	Mingaladon	1	3.6
13	Laputta	1	3.6
	Total	28	100

Table 10
Relationship between Widal test (≥1:160) and culture.

Sr No	Types	Number of typhoid fever cases
1	Culture positive (*Salmonella typhi*) and Widal positive	17 (45.95)
2	Culture positive (*Salmonella typhi*) and Widal negative	11 (29.73)
3	Culture negative and Widal positive	9 (24.32)
4	Culture positive (other organisms) and Widal positive	4 (9.76)

Note: Figures in parentheses are percentages.

(Chew *et al*, 1992; Choo *et al*, 1993); 1:80 (Thevanesaan, 1992; Shukla *et al*, 1997); 1:120 (Saha *et al* 1996); 1:160 (Rasaily *et al*, 1993; Kulkarni and Rego, 1994). H antibodies have been said to be as important as O antibodies (Kulkarni and Rego, 1994; Saha *et al*, 1996). A study of adult patients of Mandalay General Hospital (1980) revealed that 46.7% of the patients could be missed if a single titer of TO or TH 1:160 was used as a diagnostic criterion (Paing-Soe, 1980); background information regarding the agglutinin levels of the normal children of that region was needed for accurate interpretation of the test. In this study, the false positive rate was 9.7%; the Widal positive rate in other febrile illnesses has been reported as being 10% (Rasaily *et al*, 1993). The limitations of the Widal test of single specimens is well documented (Khin-Saw-Khine, 1982). Rising agglutination titers was be demonstrated in two out of four cases where second sample of blood was taken for Widal test in our study.

The clustering of enteric fever cases from Mingalartaungnyut township was reported to the Directorate of Health (DOH) for appropriate action. This finding demonstrated that hospital based studies might reflect the disease burden of communities and might, therefore, be very helpful to public health administrators.

The emergence of typhoid strains resistant to antibiotics has been reported in recent years

(Gupta, 1994; Bhutta *et al*, 1997; White *et al*, 1997). In the present study, most of the typoid fever cases were resistant to conventional antibiotics. The knowledge of the antibiotic sensitivity pattern of *Salmonella typhi* in this study might be of help in establishing a current hospital antibiotic policy for typhoid fever cases. In two of the three suspected cases of malaria, bacteremia could be demonstrated; in all the cases, blood for malaria parasites was found to be negative. Bacteria could be isolated from 27.5% of children with presumed acute viral infection. This suggests that children who presented with acute fever of more than five days without any clinical signs need to be properly investigated rather than treated for viral infection.

In conclusion, sentinel surveys of bacteremia in major hospitals should be carried out in order to detect the changing pattern of bacteremia and antibiotic sensitivity. These surveys will be of great help in establishing hospital antibiotic policies.

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