Antimicrobial Susceptibility and Distribution of Non-typhoidal *Salmonella* Serovars Isolated in Malaysian Children

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Summary

There is widespread resistance of *Salmonella* species to commonly prescribed antimicrobials the world over. We aimed to determine the antimicrobial susceptibility and serovar distribution of non-typhoidal *Salmonella* (NTS) isolated from blood cultures of Malaysian children. Positive isolates of NTS from blood cultures obtained from children admitted to the pediatric wards of University of Malaya Medical Center (UMMC), a large urban hospital from Kuala Lumpur (1991–2001), and Hospital Kota Bharu (HKB), from the predominantly rural state of Kelantan (1991–1999), Malaysia, were reviewed retrospectively. Serovar distribution and antimicrobial susceptibility were ascertained. A total of 64 and 55 isolates of NTS were obtained from blood cultures of children admitted to UMMC and HKB, respectively. The commonest serovar isolated was *Salmonella enteritidis* in both centers. The NTS isolated were highly sensitive to the antimicrobials tested: ampicillin 98 per cent, chloramphenicol 98 per cent, gentamicin 97 per cent, trimethoprim-sulfamethoxazole (TMP-SMX) 98 per cent, and ceftriaxone 100 per cent in UMMC; ampicillin 100 per cent, chloramphenicol 87 per cent, kanamycin 100 per cent, streptomycin 96 per cent, TMP-SMX 93 per cent, and tetracycline 89 per cent in HKB. There were only one and five multi-resistant isolates in UMMC and HKB, respectively. In conclusion, NTS isolated from blood cultures of Malaysian children from Kuala Lumpur and Kota Bharu were highly sensitive to commonly prescribed antibiotics. We speculate that this is due to the restriction of sales of antimicrobials in Malaysia except by prescription. Continuing vigilance and frequent antimicrobial surveillance is necessary.

Introduction

In infants and young children, gastroenteritis caused by non-typhoidal *Salmonella* (NTS) can result in systemic infections, including septicemia and meningitis.[1–3] NTS is by far the most common cause of bacteremia and septicemia in children in certain parts of the world.[4] Antimicrobials are commonly used for the treatment of patients with moderate-to-severe NTS diarrhea and may be lifesaving for persons with systemic *Salmonella* infections.[1,5] There have been numerous reports on the emergence of resistance of NTS to antibiotics the world over,[6–9] including south-east Asia.[10–12] Resistance of NTS to potent but expensive antimicrobials, such as quinolones,[12–15] and third generation cephalosporins have been noted as well.[5,16]

Malaysia is geographically located in the heart of south-east Asia. In 1989 there was a report of resistance of NTS and other Gram-negative bacteria to commonly prescribed antibiotics in this region.[17] However, there has been no recent documentation of antimicrobial susceptibility of *Salmonella* serovars in children from Malaysia. The aim of the present study was to determine the antimicrobial susceptibility and the type of serovars of NTS isolated from children admitted for NTS septicemia from two large pediatric units in Malaysia.

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Materials and Methods

Location of study

This is a retrospective review involving two large hospitals from Malaysia, i.e. University of Malaya Medical Center (UMMC) from Kuala Lumpur, and Hospital Kota Bharu (HKB) from Kelantan. UMMC is situated in the predominantly cosmopolitan area of Kuala Lumpur, which is the federal capital of Malaysia with a population of more than 1.3 million. It has a large pediatric unit that serves the local population but also serves as a tertiary referral center. The population of Kuala Lumpur comprises all the three major ethnic groups in Malaysia, namely Malays, Chinese, and Indians. HKB is a regional general hospital in the state of Kelantan, which is situated in the north-east of peninsular Malaysia. Kota Bharu is

a medium-size town with a population of approximately 200 000, which is predominantly Malay. Less than 5 per cent of the population is Chinese and other minorities. Paddy plantation, other agricultural cultivation, fishing, and petty trades are the main economic activities.

The present study was a retrospective review of all positive blood cultures of NTS in both units. In UMMC, the review was from 1991 to 2001, while in HKB the review covered the period 1991–1999. All the positive records of NTS from blood were noted and basic demographic data and antimicrobial sensitivity patterns were recorded.

Laboratory methodology

In both centers, positive salmonellae isolates were identified by standard biochemical reactions. NTS was differentiated from *Salmonella enterica* ser. Typhi by its ability to produce gas on sugar fermentation. The identification of *Salmonella* serovars was performed at the Institute of Medical Research, Kuala Lumpur, the national reference laboratory for serotyping *Salmonella* species, using specific antisera.

Antimicrobial sensitivity

In Kota Bharu, prior to 1996, antimicrobial susceptibility testing was carried out using the Stokes method.[18] From 1997 onwards, the NCCLS guidelines were used. In UMMC, susceptibility to antibiotics was tested using the standard disc diffusion method according to NCCLS guidelines throughout the study period. Commercially available discs were used and zone diameters were read as sensitive or resistant according to the manufacturers recommendation. Ampicillin, chloramphenicol, trimethoprim-sulphamethoxazole (TMP-SMX), kanamycin and tetracycline were routinely tested in HKB, while ampicillin, chloramphenicol, TMP-SMX, gentamicin and ceftriaxone were tested in UMMC. Multi-resistance was defined as resistance to more than one antibiotic tested.

Statistics

The chi-squared test was used for statistical analysis. A p-value of < 0.05 was considered to be significant.

Results

Septicemia caused by NTS was not a common occurrence in both UMMC and HKB. During the study period, there were only 64 positive blood cultures of NTS from children aged <16 years in UMMC, while the number of positive isolates in HKB was 55 (Table 1).

Demographics

NTS septicemia is a disease of the young. The median age of children with NTS septicemia in both centers was 2.0 years. The ethnic breakdown of the children with NTS septicemia in each center reflected the ethnic composition of the community served by the respective centers.

Serovars isolated (Table 2)

Salmonella enterica serovar Enteritidis was the commonest serovar isolated in both centers, comprising 42 per cent of the total isolates in UMMC and 44 per cent of total in HKB. *Salmonella serovar* Paratyphi A was the second most common serotype isolated in HKB, but it was not isolated in UMMC at all. *Salmonella serovar* Typhimurium, a common serovar isolated in sub-Saharan Africa, was not common in this study at all.[4,19]

TABLE 1
Demographic data of non-typhoidal Salmonellae *septicemic cases from Kuala Lumpur and Kota Bharu, Malaysia*

	University of Malaya Medical Center, Kuala Lumpur	Hospital Kota Bharu, Kelantan	
Year of study	1991–2001	1991–1999	
No. of positive samples isolated	64	55	
Sex			
Male	32	36	
Female	32	19	
Ethnicity			
Malays	28	53	
Chinese	25	1	
Indians	11	0	
Others	0	1	
Age (years, median)	2.0	2.0	
Salmonella serovar Enteritidis as % of total isolates	42	44	$p = 0.97$

TABLE 2
Serovar distribution of non-typhoidal Salmonella *isolated from children < 16 years old, from Kuala Lumpur and Kota Bharu, Malaysia*

Serovar	UMMC (n = 64) n	HKB (n = 55) n
Enteritidis	27	24
Paratyphi A	0	13
Paratyphi B	4	6
Typhimurium	2	0
Welterreden	0	4
Virchow	0	3
Group B	2	0
Group C	2	0
Group D	10	0
Others[a]	3	5
Non-typable	3	0
Not done	11	0

[a] One each for *Salmonella* serovar Matopeni, Newport, Bovismorbificans

Antimicrobials susceptibility (Tables 3 and 4)
Five antibiotics were routinely tested for sensitivity in UMMC while six antimicrobials were tested in HKB. The vast majority of isolates tested in UMMC were sensitive to the commonly prescribed antimicrobials, ranging from 97 to 100 per cent. There was only one isolate that was resistant to two antibiotics (noted in 1998; resistant to TMP-SMX and chloramphenicol). In HKB, the sensitivity rate was 100 per cent for ampicillin and kanamycin, but lower for TMP-SMX (93 per cent), chloramphenicol (87 per cent) and tetracycline (89 per cent). In the case of chloramphenicol, the resistance rate was significantly higher than that observed in UMMC. There were five multi-resistant strains isolated compared with UMMC ($p < 0.05$). There was no difference in the antimicrobial susceptibility pattern in each center during the first and second halves of the study period (Table 4).

Discussion

There is a disturbing general trend of *Salmonella* species being resistant to commonly prescribed antimicrobials, which has been noted in many parts of the world, including south-east Asia.[9–12] Ling and Wang[12] from Singapore in 2001 reported that 75 per cent of the *Salmonella* serovar Enteritidis isolated were resistant to sulphonamide. The emergence of multi-resistant *Salmonella* is also widespread in Africa.[19] *Salmonella* septicemia in children is often associated with significant morbidity and mortality, especially in those who are immunocompromised, and appropriate antimicrobial treatment is important in its management.[3,20]

Malaysia is in the heart of south-east Asia. The patterns of antibiotic susceptibility are expected to be very similar to those of her neighboring countries. We studied the susceptibility of *Salmonella* serovars isolated from blood cultures taken from children in an urban and a rural setting from Peninsular Malaysia. Surprisingly, in contrast to another study from this region where antimicrobial susceptibility of the causative *Salmonella* isolates varied with their serovars,[3] the present study showed that the vast majority of NTS isolated from children with NTS septicemia from an urban and a rural setting in Malaysia were sensitive to commonly prescribed antimicrobials, such as ampicillin, chloramphenicol, TMP-SMX, and aminoglycoside. In UMMC from Kuala Lumpur, the sensitivity rate ranged from 97 to 100 per cent. In HKB from Kelantan, a northern state that is predominantly rural, the sensitivity rate

TABLE 3
Antimicrobial susceptibility of non-typhoidal Salmonella *in Kuala Lumpur and Kota Bharu, Malaysia*

	University of Malaya Medical Center (n = 64)[a]			Hospital Kota Bhau (n = 55)			
Antibiotics tested	S	R	% sensitive	S	R	% sensitive	p-value
Ampicillin	63	1	98	55	0	100	0.53
Chloramphenicol	63	1	98	48	7	87	0.004
Trimethprim-sulfamethoxazole	61	2	97	51	4	93	0.41
Gentamicin	58	1	98		NT		
Kanamycin		NT		55	0	100	
Streptomycin		NT		53	2	96	
Tetracycline		NT		49	6	89	
Ceftriaxone	60	0	100				

[a] Not every isolate of NTS in UMMC was tested against all the antibiotics listed.
NT: not tested.

TABLE 4
Antimicrobial susceptibility of non-typhoidal Salmonella *in Kuala Lumpur and Kota Bharu, Malaysia*

	Ampicillin		Chloramphenicol		TMP-SMX		Gentamicin		Kanamycin		Tetracycline		Ceftriaxone	
	S	R	S	R	S	R	S	R	S	R	S	R	S	R
Hospital Kota Bharu														
1991–1995	46	0	43	3	43	3			46	0	41	5	ND	
1996–1999	9	0	5	4	8	1			9	0	8	1		
Overall	55	0	48	7	51	4								
University of Malaya Medical Center														
1991–1995	43	1	44	0	43	1	38	1					40	0
1996–2001	20	0	19	1	19	1	20	0					20	0
Overall	63	1	64	0	61	2	58	1					60	

TMP-SMX: trimethoprim-sulfamethoxazole.
S; Sensitive; R; resistant.
ND, not done.

also exceeded 85 per cent in all the antimicrobials tested. The only statistically significant difference between the two centers was the NTS isolated in HKB was significantly less sensitive to chloramphenicol than the NTS isolates from UMMC.

Unlike many countries in this region where over-the-counter sale of antimicrobials is rampant, the sale of antibiotics in Malaysia is restricted to prescription only. This may be the reason for the high susceptibility rate of *Salmonella* serovars to various antimicrobials. Another possible reason for this observation was that this study only included blood cultures obtained from children with NTS septicemia, while most other studies included isolates other than blood cultures.

There was no isolate from UMMC that was resistant to ceftriaxone. However, resistance to ceftriaxone had been reported in many parts of the world.[5,16,21] Ceftriaxone is commonly used empirically to treat children with severe *Salmonella* infection, including septicemia and meningitis. As the present study only covered one pediatric center in Malaysia, the true picture in the whole of Malaysia may be different. Continuing surveillance against ceftriaxone susceptibility in other parts of Malaysia is warranted.

We did not routinely include the quinolone ciprofloxacin in the sensitivity test because it has not been approved for use in children and is not commonly used in pediatric practice. Nevertheless, resistant *Salmonella* species to flouroquinolone has recently been reported by Parry, *et al*.[14] from Vietnam and Chiu, *et al*.[15] from Taiwan.

NTS septicemia is a disease of infants and young children.[3,19,20] It is not surprising that the median age of patients with NTS septicemia in both centers from this study was 2 years. NTS septicemia was not a common occurrence in both UMMC and HKB. This is in contrast with studies from tropical Africa where NTS was the commonest cause of bacteremia and septicemias in children.[4,19] Conditions that predispose to NTS septicemia such as malaria, anemia, and human immunodeficiency virus (HIV) infection, which are all prevalent in sub-Saharan African, are uncommon in Malaysia.[19,20]

Compared with sub-Saharan Africa where *Salmonella* serovar Typhimurium is the commonest serovar-causing septicemia,[4,19] the commonest serovar isolated in both centers from septicemic children in this study was *Salmonella* serovar Enteritidis. This finding was similar to a study in children with *Salmonella* gastroenteritis.[1] There has been a sharp increase in the prevalence of *Salmonella* serovars Enteritidis and Blockley in the 1980s in Malaysia, and a decline of *Salmonella* serovars Typhimurium and Weltevreden, which were the prevalent serovars in the 1970s.[22,23]

This study has shown that unlike many parts of south-east Asia, NTS isolated from children with NTS septicemia in two large pediatric centers from Malaysia were highly sensitive to commonly prescribed antimicrobials. However, continuing surveillance of antimicrobial susceptibility in the NTS isolated from children is necessary.

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