ORIGINAL ARTICLE

Drug Sensitivity Pattern in Children with Blood Culture Positive Enteric Fever in a Tertiary Care Hospital in Pakistan

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Abstract

Background
Salmonella infections are endemic in Pakistan and resistance to these infections is increasing which has implications. We aim this study to determine the antimicrobial resistance pattern of *Salmonella infections* among children admitted at tertiary care hospital in Pakistan.

Methods
We have performed an audit in children aged 1 month to15 years with positive blood culture for *Salmonella enterica serovars Typhi and Paratyphi A,* diagnosed at Shifa international hospital's Microbiology Laboratory, from May 2012 to Dec 2013. Blood cultures positive for *S. Typhi* and *S. paratyphi A* isolates were confirmed with specific anti-sera and their antibiogram determined by Modified Kirby-Bauer Disc Diffusion method using 6 relevant antibiotics (Ampicillin, Trimethoprim-sulphamethoxazole, Chloramphenicol, Ciprofloxacin, Cefixime and Ceftriaxone). Interpretation was done according to the guidelines of Clinical and Laboratory Standards Institute.

Results
In our study we had 200 positive cultures, 75% were *Salmonella typhi* and 25% were *Salmonella paratyphi A* isolates. Overall mean age of children was 8.1 +S.D=4.3 years. Majority (61%) were males. Of the total isolates, 88% were resistant to one or more drugs with 75% and 25% being serotype *typhi* and *paratyphi A* respectively. Resistance to 1st line antibiotics was significant in *Salmonella Typhi* compared to *Salmonella paratyphi A* (co-trimoxazole 63% vs. 6%, Chloramphenicol 71% vs 14%, Ampicillin 67% vs 10%). MDR strains were also more common in *Salmonella Typhi* (59%) as compared to *Salmonella Paratyphi (*20%).

Conclusions
The study showed significant resistance to 1st line antibiotics by *Salmonella serotype typhi* compared to *Salmonella serotype Paratyphi A.* Parenterally and orally administered third generation cephalosporins remain effective drugs against infections caused by *Salmonella serotype typhi and Paratyphi A.*

Key Words
Enteric fever, drug resistance, children

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Introduction
Enteric fever caused by *Salmonella typhi (S. Typhi) and Salmonella Paratyphi A (S. Paratyphi A*), affects approximately 27 million people each year throughout the world.[1] The disease is more common in Asian subcontinent.[2] In developed countries, the disease is seen mainly in those returning from endemic areas.[3,4,5] In Indian subcontinent (India, Pakistan, Afghanistan, Nepal and Bangladesh) it is a major public health problem.[6] In Pakistan, the incidence of Enteric fever is comparatively high (451/100000) which supports the data from Kothari *et al*,[7] which claim that Enteric fever is more common in Asia than Africa. The incidence of Enteric fever in India during 2003–2004 was estimated to be 214.2 per 100,000 people/year in a population-based study.[8]

Infection caused by *S. Typhi* is confirmed by isolating the organism from blood, bone morrow, urine or stool. But in developing countries the technical training and laboratory facilities required for this are not available in most primary health care centres.[9,10]

Conventionally Typhoid fever is being treated with first-line antibiotics such as Ampicillin, chloramphenicol and cotrimoxazole. But the excessive use of these antibiotics has led to the emergence of multidrug-resistant (MDR) strains of *S. Typhi*. MDR in *S. Typhi* is almost exclusively conferred by a inchi1 plasmids which carries a suite of resistance genes.[11] Similarly increased use of Ciprofloxacin (one of the Fluoroquinolones) has led to *S. Typhi* with elevated flouroquinolones mics.[12,13]

Empirical treatment of typhoid and paratyphoid fever is usually started before culture and sensitivity test results are available. So it is important to know the treatment options in the beginning. Furthermore antibiotic treatment reduces the mortality from 30% (in untreated patients) to as low as 0.5% (in treated

patients).[14] This study from a tertiary care hospital highlights resistance pattern in *salmonella* infections.

Material and Methods

In this retrospective study children aged 1 month to 15 years with positive blood culture for *Salmonella enterica serovars Typhi* and *Paratyphi A* from May 2012 to Dec 2013 diagnosed at Shifa International Hospital's Microbiology Laboratory were included in the study. They included both admitted and outpatients. Shifa International Hospital is a 550-bed tertiary care hospital at Islamabad, Pakistan. Its clinical laboratory receives samples from both in-patients and out-patients. Blood samples are collected through more than 40 collection points located in various cities of the country.

After approval from IRB (Institutional Review Board Ref. No 2076-125-2013) data was collected. From May 2012 to Dec 2013, 200 blood cultures were positive for *S. Typhi* and *Paratyphi A* isolates. Isolates were confirmed with specific anti-sera and their antibiogram determined by Modified Kirby-Bauer Disc Diffusion method using 6 relevant antibiotics (Ampicillin, Trimethoprim-sulphamethoxazole, Chloramphenicol, Ciprofloxacin, Cefixime and Ceftriaxone). Interpretation was done as per CLSI guidelines.[15]

The data was drawn from the computerized system of the hospital and entered into SPSS version 21 for statistical analysis. In descriptive analysis means and standard deviations of the continuous variables and percentages of the categorical variables were computed. Comparisons between *Salmonella typhi* and *Paratyphi A* by gender, age and antibiotic resistance were done through Chi square test where appropriate. T-test was used to compare the means of age with respect to culture.

Results

A total of 200 blood isolates of *salmonella* species were included in the analysis between May 2012 and Dec 2013. Most of the isolates (N=123, 61%) were from male patients and majority of them were <9 year old. The patients were analysed for demographic and microbiologic characteristics (Table 1). The results show that resistance to 1st line antibiotics was significant in *Salmonella Typhi* compared to *Salmonella Paratyphi A* (co-trimoxazole 63% vs. 6%, Chloramphenicol 71% vs 14%, Ampicillin 67% vs. 10%). MDR strains were also more common in *Salmonella Typhi* (59%) as compared to *Salmonella Paratyphi* A (20%). (Table 2)

Discussion

Typhoid fever is a major public health problem in developing countries including Pakistan.[6] The disease can be prevented by public health education, proper sanitation and vaccination. The development of antibiotic resistant species of *salmonella* is closely related to injudicious use of antibiotics.[16]

In our study the isolation rate of *salmonella Typhi* (75%) was

Table 1: Descriptive characteristics and Pattern of Antibiotic sensitivity in Patients with *S. Enterica serovar Typhi* and *Paratyphi A* infection

	N (%)
Gender	
Male	123 (61)
Age Groups	
0-5	70 (35)
6-15	130 (65)
Organism Isolated	
Salmonella Typhi	150 (75)
Salmonella Paratyphi A	50 (25)
Cotrimoxazole	
Resistant	97 (48)
Chloramphenicol	
Resistant	113 (56)
Ampicillin	
Resistant	105 (52)
Ceftriaxone	
Resistant	0 (0)
Cefixime	
Resistant	0 (0)
Ciprofloxacin	
Resistant	6(3)
Intermediately Resistant	164(82)
Nalidixic acid	
Resistant	114(57)
Intermediately Resistant	85(42)

Table 2: Inferential Analysis in Patients with *S. Enterica serovars Typhi* and *Paratyphi A infection*

	Salmonella Typhi N(%)	*Salmonella Paratyphi A* N(%)	P-value
Age (Mean)	7.87	8.88	0.607
Gender			
Male	92 (61)	31 (62)	0.933
Age Group			
0-5	57 (38)	13 (26)	0.123
6-15	93 (62)	37(74)	
Co-trimoxazole			
Resistant	94 (63)	3 (6)	<0.001
Chloramphenicol			
Resistant	106 (71)	7 (14)	<0.001
Ampicillin			
Resistant	100 (67)	5 (10)	<0.001*
Ciprofloxacin			
Resistant	5 (3)	1(2)	0.099*
Nalidixic acid			
Resistant	86 (57)	28(56)	0.827*
MDR			
(Multidrug Resistant)	88 (59)	10 (20)	<0.001*

*Fischer exact test

significantly greater than *S. Paratyphi A* (25%) which correlates with other data which suggest that typhoid fever is more prevalent than paratyphoid in this part of the world.[17,18] Results from one of the studies showed that out of 62 children 34 were male and 28 were female. The age ranged from 2 to 14 years, the mean age being 7.62 years[19,20] which correlates with our study.

Emergence of drug resistance to 1st line drugs like Ampicillin, chloramphenicol and co-trimoxazole is alarming. In a study from Karachi in 2006 drug sensitivity pattern of *S. Typhi* showed sensitivity of 44.2%, 44.8% and 40.5% for Ampicillin ,co-trimoxazole and chloramphenicol respectively[21] which is greater than the sensitivity noted in our study(Table 2). However sensitivity pattern of *Salmonella Paratyphi A* in the same study from Karachi showed sensitivity of 27.9%, 29.9% and 29.5% for Ampicillin, co-trimoxazole and chloramphenicol respectively which is much lesser than the sensitivity noted in our study (Table 2). This shows that resistance of *salmonella typhi* is increasing against these 1st line antibiotics. However the present study shows decrease in resistance of *salmonella paratyphi A* to these drugs. This fact may be due to geographic variation which needs further evaluation.

MDR strains of *S. Typhi* and *S. Paratyphi A* have been reported from 16.1% to 83.3% in different studies.[22,23] In this study out of 200 isolates 98(49%) were MDR strains with 88 isolates from *S. Typhi* and 10 from *S. Paratyphi A* groups. This shows that MDR strains continue to be alarming sign for the community. One of the studies has shown high incidence of MDR strains of *S. Typhi* from Iraq (83%) and Pakistan (52%) as compared to other countries.[24] Another study indicates that 80% cases of MDR strains of *S. Typhi* arise from the Asian countries.[25] Increase in rate of isolation of MDR strains of *S. Typhi* from 2009 (13.6%) to 2013 (25%) in a study[26] was in agreement with another study from New Delhi which showed isolation of MDR strains of *S. Typhi* was 34% in 1999 which has increased to 66% in 2005.[27]

Resistance to Fluoroquinolone is increasing. Present study reveals out of 150 isolates of *S. Typhi* only 27 (18%) and out of 50 isolates of *S. paratyphi A* only 3 (6%) were susceptible to ciprofloxacin. This reflects the injudicious use of this antibiotic and its availability over the counter in the country. Resistance to Nalidixic acid has been used as a marker of low level resistance to ciprofloxacin in *S. Typhi* infection.[28,29] So it is important to look for nalidixic acid resistance in *S. Typhi* infections along with ciprofloxacin and if the isolates are resistant to nalidixic acid, they should be reported as having intermediate susceptibility to ciprofloxacin.

Both oral and parenteral 3rd generation cephalosporin are the most important drugs used these days for the treatment of enteric fever.[16] In this study both *S. Typhi* and *S. paratyphi A* isolates showed 100% susceptibility to ceftriaxone and cefixime. Similar results were observed by Gautum *et al* from Haryana, India.[16] Both these antibiotics are also effective against MDR strains. But some other studies have also shown increased resistance of *Salmonella Typhi* to both ceftriaxone and cefixime. One of the studies from Bangladesh has shown resistance of *Salmonella Typhi* to ceftriaxone as high as 68% and to cefixime 91% which is in contrast to our study.[30] Another study from Faisalabad, Pakistan out of 19 isolates of *Salmonella typhi* resistance to ceftriaxone was shown in only 1(5%) isolate while to Cefixime it was present in 7(37%) isolates.[31] In another study from Pakistan out of 86 isolates of *Salmonella Typhi* 17.5% were resistant to ceftriaxone.[32] All these 3 studies are in contrast to our study and this needs further studies in our local population.

What this study adds is Resistance pattern from a local tertiary care hospital with comparisons. However there are limitations as this is a Retrospective study from a single centre with 4 years old data.

Conclusion

The study shows significant resistance to 1st line antibiotics by *Salmonella serotype typhi* compared to *Salmonella paratyphi A*. Parenterally and orally administered third generation cephalosporin remain effective drugs against infections caused by *Salmonella serotype typhi* and *paratyphi A*.

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