ORIGINAL ARTICLE

Clinical Spectrum of XDR and MDR Enteric Fever in Children Aged Between 6 Months to 15 Years at The Children's Hospital, Lahore

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Abstract

Background
To determine the clinical spectrum of XDR and MDR enteric fever in children.

Methods
A cross sectional study conducted at pediatric medical units of the Children's Hospital and the Institute of Child Health, Lahore from May 2019 to Feb 2020. 135 children of both genders between 6 months to 15 years of age fulfilling the operational definition of enteric fever were included in the study. All study cases were evaluated for clinical features, possible complications, antimicrobial sensitivity pattern and duration of stay in the hospital. Data was analyzed by using SPSS version 23.

Results
Mean age of our study cases was 6 years with male preponderance (69.6%). Majority (44.4%, n=60) were between 1 to 5 years of age out of which significant number of children (20%) were between 6 months to 2 years. Among patients between 6 months to 2 years, 67.8% (n=19) were XDR. Fever was present in 100% followed by GI symptoms (40%) including nausea, vomiting, diarrhea and abdominal pain. Among all bacterial isolates of enteric fever, 54.1% were XDR, 20% were extended spectrum beta lactamase positive (ESBL), 18.5% were MDR and 7.4% were non-resistant. Levofloxacin was found to be sensitive in 41% of the total cases as compared to ceftriaxone (23%) and ciprofloxacin (22%). 8.8% of cases developed complications majority of them were XDR Enteric. Hepatobiliary complications (splenic infarcts, icteric hepatitis, pancreatitis) were found in 5.1 %, followed by CNS complications (cerebellar ataxia, meningitis (2.2%). XDR enteric was significantly associated with male gender (p-value 0.025), prolonged durations of fever (p-value 0.047) and hospital stay (p-value 0.010).

Conclusion
XDR enteric fever constitutes more than 50% of total enteric fever cases with prolonged hospital stay and chance of complications as compared to MDR enteric fever. ESBL is the second most common type of drug resistant enteric fever.

Key words
Enteric fever, XDR, MDR, ESBL, levofloxacin, children

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Introduction

The commonly known Enteric fever includes both typhoid and paratyphoid fevers. *Salmonella enterica* subspecies *enterica* serovar Typhi (*Salmonella typhi*) causes typhoid fever, while paratyphoid fever results from infection with any of three serovars of *Salmonella enterica* subspecies *enterica*, namely *S. paratyphi A*, *S. paratyphi B* and *S. paratyphi C*.[1] Typhoid fever is still at large a major contributor to global morbidity.[2] Millions of new typhoid cases are being reported globally each year despite of treatment and preventive effors.[3] Typhoid fever causes an estimated 20 million infections and 2,00,000 deaths annually in resource limiting countries of the world.[4] Significant percentage of enteric fever cases in endemic areas comprise of children.[5] According to a local study conducted in Karachi, incidence of typhoid fever in children in Karachi was 170-450 per 1,00,000 children annually.[6] The clinical presentations of enteric fever are non-specific, vary in different communities, and many a times it's difficult to differentiate those clinically from other febrile illnesses and it results in delay in diagnosis and treatment.[7] Typhoid fever is a systemic illness. The common symptoms of typhoid fever are fever, malaise, anorexia, myalgia, headache, diarrhea and abdominal pain. Complications can involve any system and can cause intestinal hemorrhages or perforation, sepsis, overt hepatitis, cholecystitis, pneumonia, myocarditis, neurological complications, bone marrow necrosis, pyelonephritis, nephrotic syndrome, parotitis, orchitis, osteomyelitis, suppurative arthritis etc. Untreated typhoid fever can lead to mortality at 10% or more.[8] Gold standard for the diagnosis of typhoid fever is microbial culture of blood or bone marrow.[9] Antimicrobial resistances to typhoid fever is emerging as a big problem worldwide.[10] The common factors contributing to drug resistance in typhoid fever are over-prescription, over the counter use of antibiotics and under dosing of drugs.[11] Typhoid fever strains were sensitive to all first line antibiotics before 1970's.[12] Since then, resistance started appearing worldwide. One study showed that by 1990, MDR *S. typhi* strains were being documented in India and Pakistan.[13] Following emergence of MDR strains, fluoroquinolones were introduced for the treatment of typhoid fever. By 2000, resistance to

quinolones also appeared so third generation Cephalosporins were added to the treatment of typhoid fever.[14] In November 2016, first case of XDR typhoid fever was reported in Sindh province in Pakistan. Since then more than 300 cases have been reported in Pakistan.[15] New drugs have been recommended including azithromycin and carbapenems for the treatment of XDR typhoid fever in children.[15] Different factors had been identified in one study which were associated with emerging ceftriaxone resistance and those were male gender, eating food from outside of the house, contamination with *S. typhi* infected patient and history of antibiotic use.[16] It is the need of the hour to conduct studies to assess the evolving trend of antimicrobial resistance in typhoid fever in children in Pakistan and even CDC has also issued health warning against XDR typhoid fever in Pakistan.[17]

The rationale of the study was to conduct a study on typhoid fever in children keeping in view the alarming situation of MDR and XDR typhoid fever in Pakistan so that the different clinical presentations of typhoid fever were assessed and evolving antimicrobial susceptibility was also assessed.

Patients and Methods

This cross-sectional study was conducted at the Department of Pediatric Medicine, The Children's Hospital & Institute of Child Heath, Lahore, Pakistan. Children admitted in medical wards or presenting in OPD were assessed according to the clinical case definition of suspected typhoid fever (Table 1). Sample size was calculated by using prevalence base formula that is:

$$n=(z^2 P(1-P)) / d^2$$

Z= level of confidence= 95% = (1.96)
P= expected prevalence= 10% = 0.1
D= precision = 0.05

Total 135 patients were included in the study fulfilling the inclusion criteria over the period of 1 year, from May 2019 to April 2020. Patients were selected by consecutive sampling. Inclusion criteria was defined as children between the age of 6 months and 15 years fulfilling the operational definition of confirmed typhoid fever admitting in hospital or presenting in outdoor. All those Children with clinical picture explicit for diagnosis of measles, chickenpox, otitis, infected wounds, malignancy, connective tissue disease were excluded.

Sample Collection, Processing and data analysis

Informed consent was taken before enrolling every patient. All 135 patients fulfilling the case definition of either suspected typhoid fever or confirmed typhoid fever initially were enrolled

Table 1. Classification of Typhoid Fever Cases by Drug Resistance Status, Pakistan [19]

Typhoid fever type	Operational Definitions
Confirmed typhoid fever	A patient with persistent fever (38 °C or above) lasting 3 or more days and S. Typhi isolated on blood or bone marrow culture.
Probable / Suspected case of typhoid fever	A patient with documented fever (38°C and above) for at least 5 days prior to presentation, with rising trend in fever and having no other focus to explain the cause of the fever (e.g. UTI, pneumonia, abscess etc.) OR A clinically compatible case that is epidemiologically linked to a confirmed case of typhoid fever
Antibiotic sensitive typhoid fever	Typhoid fever caused by *S. typhi* and *S. paratyphi* A,B,C strains which are sensitive to 1st line drugs[1] and 3rd generation cephalosporins, with or without resistant to second line drugs[2].
MDR (multi drug resistant) typhoid fever	Typhoid fever resistant to 3 first line drugs including Chloramphenicol, Trimethoprim -Sulphamethoxazole and Ampicillin.[15]
XDR (extensively drug resistant) typhoid fever	Typhoid fever resistant to first line antibiotics and second line antibiotics including fluoroquiolones and 3rd generation cephalosporins. [15]
Extended spectrum Beta lactamase positive Enteric fever	Typhoid fever caused by *Salmonella typhi* which is resistant to 3rd generation cephalosporin but may be sensitive to chloramphenicol, co-trimoxazole or flouroquinilones.[19]

1. Ampicillin, chloramphenicol, trimethoprim- sulfamethoxazole
2. Flouroquinolones

and assessed. Complete blood count (CBC) and blood culture was sent in each case by collecting 1 to 3 ml of blood in pediatric blood cultures (Becton Dickinson) and incubated at 37^0C for 7 days in Bactec 9240 automated blood culture analyser. Upon detection of positivity by the instrument, samples were sub-cultured on blood agar (Oxoid, UK) and MacConkey agar (Oxoid, UK) plates at 37^0C for another 18-24 hours. Organism identification and classification was performed by using API 20 E kits (bioMerieux, France). Antimicrobial Susceptibility testing was performed by Kirby Bauer disc diffusion method and reported according to Clinical Laboratory Standard institute (CLSI) guidelines 2019. Double Disc Synergy Test (DDST) was used for phenotypic detection of Extended Spectrum B-Lactamase (ESBL) producing isolates (CLSI, 2019). In this method, test organism was inoculated on Mueller Hinton Agar (Oxoid, UK) and cephalosporin disc (Oxoid, UK) was placed 30mm centre to centre apart from co-amoxiclave disc (Oxoid, UK). After overnight incubation at 37^0C, increase in zone size toward co-amoxiclave disc indicated ESBL production. Liver function tests (LFTs), Urine complete examination, chest radiograph, ultrasound abdomen and other investigations were done in relevant cases.

Only confirmed cases of typhoid fever on culture were included in the study and divided into two groups. One group comprised of patients with MDR typhoid fever and second group comprised of patients with XDR typhoid fever. A detailed history regarding clinical presentation was recorded and thorough examination was performed on each patient. All information was put on a questionnaire sheet. Data was entered and analyzed by Statistical Package of Social Sciences (SPSS) version 23.

Results

Initially 457 cases of typhoid fever were suspected and only 135/457 (29.5%) were culture proven cases which were included in the study. Among a total of 135 confirmed cases, forty-one (30.4%) were females and ninety-four (69.6%) were males. Enteric fever was found more common 42(42.9%) between 1 to 5 years of age. Common presenting complains were fever (100%) followed by rigors (60.7%), GI symptoms (40%) including nausea, vomiting, abdominal pain and diarrhea, arthralgia (30.4%) and headache (9.6 %). Patients with XDR enteric fever had longer duration of fever as compared to the MDR and ESBL positive cases. Common clinical findings were coated tongue (80.7%), tender abdomen (25.9%) hepatosplenomegaly (14.8%). Out of all 135 blood cultures isolates, one hundred and twenty (88.9%) were *Salmonella typhi*, five (3.7%) were *Salmonella paratyphi*, ten (7.4%) were undifferentiated *Salmonella* Species. Among all *Salmonella* species, seventy-three (54.1%) were XDR typhoid, twenty-seven (20%) were extended spectrum beta lactamase positive (ESBL) typhoid, twenty-five (18.5%) were MDR and 10 (7.4%) were non- resistant strains. Clinical spectrum is summarized in Table 2.

Salmonella typhi were found in 100 % of cases of MDR and

Table 2. Chi Square test, *p-value significant at 0.050. Typhoid type was found associated with gender (p-value 0.025), duration of fever (p-value 0.047) and duration of stay at hospital (p-value 0.010).

		Typhoid Type			
		MDR (n= 25)	XDR (n=73)	Total	P-value
Age in years	6 months to < 1 year	2(8.0%)	1(1.4%)	3(3.1%)	0.254
	1 to 5 years	9(36.0%)	33(45.2%)	42(42.9%)	
	>5 to 10 years	9(36.0%)	32(43.8%)	41(41.8%)	
	>10 to 15	5(20.0%)	7(9.6%)	12(12.2%)	
Gender	Male	12(48.0%)	54(74.0%)	66(67.3%)	0.025*
	Female	13(52.0%)	19(26.0%)	32(32.7%)	
Duration of fever in days	3 to 7 days	10(40.0%)	13(17.8%)	23(23.5%)	0.047*
	8 to 14 days	8(32.0%)	28(38.4%)	36(36.7%)	
	15 to 21 days	7(28.0%)	22(30.1%)	29(29.6%)	
	22 to 30 days	0(0.0%)	7(9.6%)	7(7.1%)	
	31 to 60 days	0(0.0%)	3(4.1%)	3(3.1%)	
Hospital stay	≤7 days	17(68.0%)	25(34.2%)	42(42.9%)	0.010*
	8-14 days	6(24.0%)	37(50.7%)	43(43.9%)	
	15-21 days	1(4.0%)	10(13.7%)	1(11.2%)	
	> 21 days	1(4.0%)	1(1.4%)	2(2.0%)	

89% cases of XDR typhoid fever, while 4.1% (n= 3) of XDR cases were having *Salmonella paratyphi* and 6.8% (n= 5) were having undifferentiated *Salmonella* species in blood cultures. Among ESBL positive cases, 93.3% were *Salmonella typhi* isolates and 6.7 % were undifferentiated *Salmonella species*. There was no statistically significant association between Culture finding and typhoid type (p-value 0.334). (Figure 1).

Complication were found in twelve (8.8%) patients. CNS complications were found in 3 (2.2%) patients with XDR enteric; 1 patient with acute cerebral ataxia and 2 patients were having Enteric meningitis. GIT complications were also found in 7 (5.1%) patient with XDR Enteric including pancreatitis (n=2), enteric hepatitis (n=2), GI hemorrhage (n=2) and splenic infarct (n=1). There was no death documented in children with enteric fever. All patients (100%) got discharged with no mortality.

Discussion

Drug resistant enteric Fever is posing a major health burden in developing World including Pakistan. In our study male patients were affected more (67.3%) than female (32.7%), that is consistent with some other studies.[20] The highest incidence of the disease occurred during the monsoon months (July – September). Common Age group was 1 to 5 years that is consistent with other studies in the past.[21,22,23] We observed significantly higher incidence of culture proven enteric fever (20%) between 6 months to 24 months of age. This could be explained by the fact that children require low dose of bacterial count to develop infection. Majority of the patients had fever of average 1 to 2 weeks before they presented to hospital. Only 10 patients (7.4%) were Non-resistant enteric which is in contrast with the study conducted in 2010 in southern coastal Pakistan, which concluded 80% susceptibility to 1st line antibiotics.[24] This Alarming situation is the outcome of injudicious and early change of antibiotics in our community during febrile illnesses. In our study we have found an alarming developing resistance against Carbapenum and Azithromycin 6.8% and 2.7% respectively. Our study proved that we are heading towards the scenario in which we will have to face

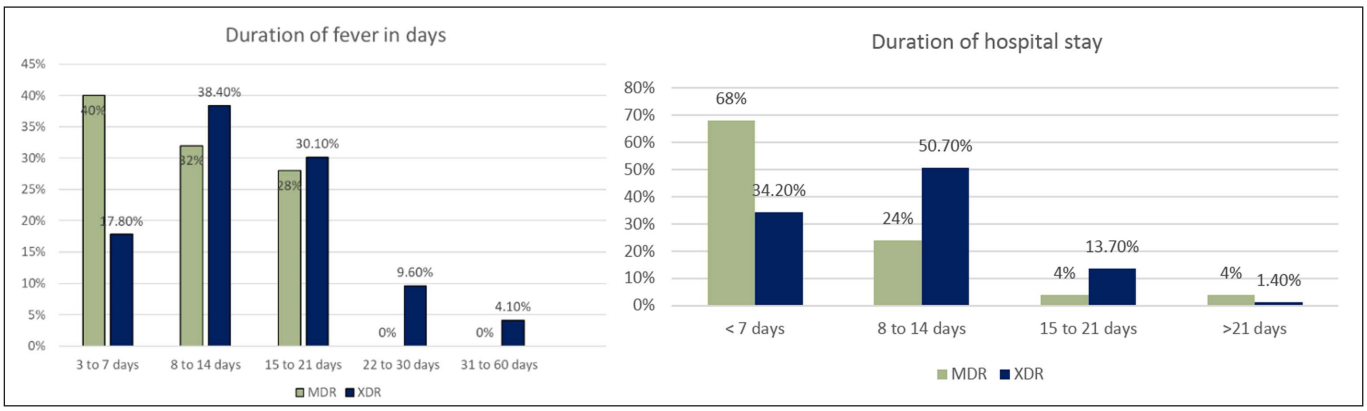

Fig1. Comparison of duration of fever and hospital stay in MDR and XDR enteric fever

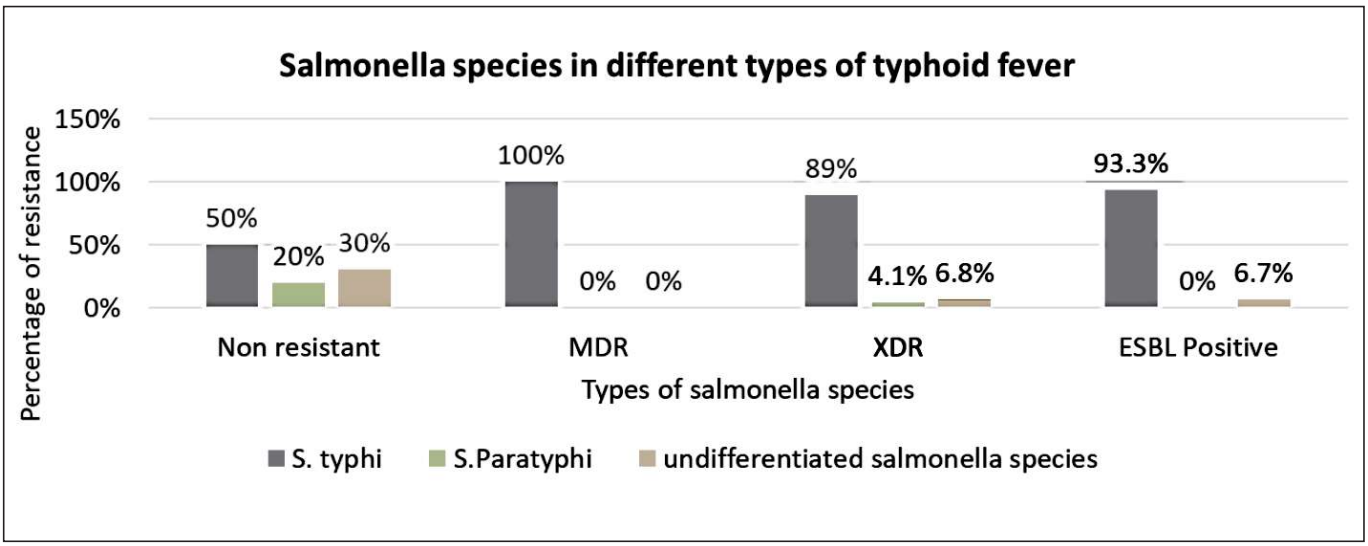

Figure 2. Type of typhoid fever and salmonella species, isolated in patients.

Azithromycin resistant strains of salmonella species. This would have left with treatment option of carbapenum (Meropenum, Imipenum, ertapenum) as a first line antibiotics for treating Enteric fever. These medications have disadvantage of being expensive, administered parentally and needs hospital admission that would add more hurdles in treating drug resistant enteric fever in resource limited countries. Levofloxacin (41%) was found more susceptible as compare to ciprofloxacin (22%) amongst fluoroquinolones. There are limited studies regarding clinical trial of levofloxacin in the treatment of enteric fever. There was one study which was conducted on adult patients with uncomplicated enteric fever.[25] It stated 100% clinical efficacy with 7-days treatment of levofloxacin in uncomplicated enteric fever with average defervescence within 3 days. There is a need to conduct more studies in the clinical trial of levofloxacin in pediatric population suffering from uncomplicated enteric fever especially in resource limited countries.

Conclusion

XDR enteric fever constitutes more than half of the total enteric fever cases leading to prolonged hospital stay and complications as compared to MDR enteric fever. ESBL is the second most common type of drug resistant enteric fever in our study. There is enough evidence regarding enteric fever burden in pediatric patients of 2 years, as evident by other studies as well,[21] warrant introduction of new safe typhoid vaccine in in Expanded Program of Immunization (EPI) schedule for children less than two years.

Limitations

1. Due to budget constraints, MIC (Minimum Inhibitory Concentration) could not be performed. Antimicrobial sensitivity testing was performed by Kirby Baur disc diffusion method according to CLSI, 2020 guide lines.[26]
2. Genetic testing could not be done due to non-availability of logistics.

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