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Changing Pattern of Resistance in Typhoid Fever in An Era of Antimicrobial Resistance: Is It Time to Revisit Treatment Strategies?

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ABSTRACT

Objectives: Multi-drug-resistant *Salmonella Typhi* is a major concern in the current era of antimicrobial resistance. The emergence of ceftriaxone-resistant *Salmonella Typhi* strains (CRST) has left very few therapeutic options for the treatment of these infections. The objective of this study was to evaluate the typhoid cases diagnosed with CRST and retrospectively analyze the antimicrobial sensitivity pattern of *Salmonella Typhi* over the past three years.

Methods: Laboratory data from our hospital were reviewed for the past three years to examine the trends related to the isolation of *Salmonella Typhi* from blood and their antimicrobial susceptibility. The case records from the patients whose blood culture detected Ceftriaxone resistant *Salmonella Typhi* (CRST) were reviewed to assess the clinical condition and outcome.

Results: Analysis of the antimicrobial susceptibility of *Salmonella Typhi* showed a decline in multidrug-resistant strains. Nalidixic acid resistance has remained high during this period. The resistance to ceftriaxone has shown an increase from 0% in 2016 to 11(12.6%) isolates in 2018. Also, azithromycin resistance has shown a steady increase from 0% in 2016, 3 (3.2%) in 2017 to 7 (8.5%) in 2018. There was no clustering of cases by time or space indicating a non-outbreak spread of ceftriaxone resistant strains.

Conclusion: Development of resistance to ceftriaxone and azithromycin necessitates revision of therapeutic choices. All this necessitates an approach to do further extensive surveillance at the community level and improvement in hygiene, sanitation, and drinking water supply. Maybe it is time to revisit the older antibiotics such as ampicillin, cotrimoxazole, and chloramphenicol as therapeutic options for uncomplicated typhoid fever. *J Microbiol Infect Dis 2021; 11(1):1-7.*

Keywords: *Ceftriaxone-resistance S. Typhi, MDR S. Typhi, Typhoid fever, antimicrobial susceptibility*

INTRODUCTION

Enteric fever continues to be a major health problem especially in developing countries such as India. It is estimated that 12 million cases occur annually worldwide [1]. A meta-analysis from India has reported a pooled estimate of incidence at 377 per 1,00,000-person-years with the highest incidence between 2-4 years of age [2].

Enteric fever is further complicated by the development of multidrug resistance strains (strains resistant to ampicillin, cotrimoxazole, and chloramphenicol). The emergence of multidrug resistance strains (MDR) strains saw the treatment options to change to fluoroquinolones. The last few years have seen over-usage of fluoroquinolones for multiple indications. The resultant increase in antimicrobial pressure caused *S. Typhi* strains to become resistant to fluoroquinolones leaving ceftriaxone and macrolides as the last options. Over time, ceftriaxone-resistant Salmonella strains (CRST) have started emerging from various countries including the Indian subcontinent [3]. This is alarming as it leaves very few therapeutic options for the treatment of these infections. Our center caters to a patient population from Delhi and neighboring areas. In 2018 we noticed a number of *S. Typhi* strains resistant to ceftriaxone. This prompted to study of these cases and also retrospectively analyze the antimicrobial sensitivity data of *Salmonella Typhi*.

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METHODS

Microbiological methods

The study was conducted at a 1200 bedded Tertiary care center after due ethical approvals. The microbiology laboratory receives samples from both adults as well as children's hospitals. The blood cultures are performed using automated BacT Alert™. The isolates were identified and serotyped as per laboratory protocol. The antibiotic sensitivity is performed as per CLSI guidelines using the Kirby Bauer method [4]. All isolates exhibiting multi-drug resistance patterns were subjected to identification and antimicrobial sensitivity analysis by Vitek™. The laboratory is part of the National AMR surveillance network. As part of the network mandate, any isolate exhibiting an unusual resistance pattern is sent for confirmation to National Referral Center at National Center for Disease Control, Delhi. Quality assurance of laboratory with positive and negative isolates is also carried out at NCDC and at EQAS center of Indian Association of Medical Microbiologists at Sir Gangaram Hospital, New Delhi. The antimicrobial susceptibility data is collated using WHONET™. The antimicrobial susceptibility data were analyzed using the data analysis tool of WHONET™ for 2016, 2017, and 2018. Blood culture records were reviewed from 2016 to 2018. Laboratory data from our hospital was also reviewed for the past three years to examine the trends related to the isolation of *Salmonella Typhi* and their antimicrobial susceptibility.

Clinical Data Review

The case records from the patients whose blood culture detected Ceftriaxone resistant *Salmonella Typhi* (CRST) were reviewed to assess the clinical condition and outcome. The clinical data were reviewed to assess the outbreak or non-outbreak situation of CRST and to examine and correlate the conditions attributing to such a high level of resistance.

RESULTS

Microbiological data

In the study period (between 2016 and 2018) a total of 5,807 bacterial pathogens were isolated from 50,479 blood samples received in the laboratory for culture. An overall culture positivity rate of 11.5% was observed. Of these 248 isolates of *Salmonella Typhi* were isolated over a three-year period accounting for 4.3% of total bacterial isolates (Table1).

Analysis of the antimicrobial susceptibility of *Salmonella Typhi* (Table 2) isolates showed a decline in multi-drug resistant strains (Strains of *S. Typhi* resistant to ampicillin, chloramphenicol and trimethoprim sulphamethoxazole) from 9.7% (7 isolates) in 2016 to 5.8% (5 isolates) in 2018. Resistance rates to ampicillin and trimethoprim sulphamethoxazole have remained fairly constant over the past three years but the resistance rate to chloramphenicol have decreased from 13.8% (10 isolates) in 2016 to 9.2% (8 isolates) in 2018. Amongst fluoroquinolones, nalidixic acid resistance has remained very high in our hospital (91.8% in 2016 to 96.5% in 2018). This makes the treatment options with fluoroquinolones impossible. Overall the past three years have seen a fall in MDR strains along with rising resistance to azithromycin and ceftriaxone in *Salmonella Typhi* (Figure 1).

A total of 11 isolates showed resistance to ceftriaxone by disc diffusion. Minimum inhibitory concentration of all these isolates was found to be more than 4 by Vitek™. All these isolates were sensitive to chloramphenicol and meropenem, while all of them were resistant to nalidixic acid. Table 2 shows the antimicrobial susceptibility pattern of ceftriaxone resistant *Salmonella Typhi*. Comparative analysis of CRST with *Salmonella Typhi* isolates showed higher resistance to other antibiotics (Figure 2).

Clinical Review of Ceftriaxone Resistant Salmonella Typhi cases

Analysis of all 11 patient records retrospectively revealed that all cases were unimmunized and had a severe clinical presentation with some complications. They were all admitted for a mean hospitalization period of 28.7 (range 17-45) days. All presented with prolonged fever (>10 days) and abdominal pain. Nine patients reported changes in bowel habits, with diarrhea being the most common symptom. Three patients had vomiting and rashes while one patient presented with signs and symptoms of acute abdomen. Seven cases had massive hepatosplenomegaly while all cases had

pancytopenia (Table 3). All cases had elevated CRP while 8 (72%) had deranged liver enzymes. All these patients lived in different areas of the city. Only two cases were residents of rural areas residing with relatives in Delhi. Also, all cases were reported at different times of a year indicating non-outbreak potential. None of these patients had received typhoid vaccination. One patient died while three cases developed complications. One patient developed septic arthritis. Fluid aspiration from the joint did not reveal any cultivable bacteria. Another patient developed intestinal perforation and was transferred to surgery for management. The third case was complicated by sepsis caused by methicillin-resistant Staphylococcus aureus. The septicemia along with typhoid resulted in the death of this patient.

All cases had prolonged fever which did not respond to multiple antibiotics. Nine out of eleven patients were started on ceftriaxone initially, while two patients were given cefixime. The blood culture report was made available within 72-96 hours still ceftriaxone was continued for a mean of 11.1 days. Six patients with favorable outcomes responded to azithromycin while one case was given meropenem. All patients were prescribed oral cefixime for 10-14 days, while 4 patients were discharged on the same for 10 days.

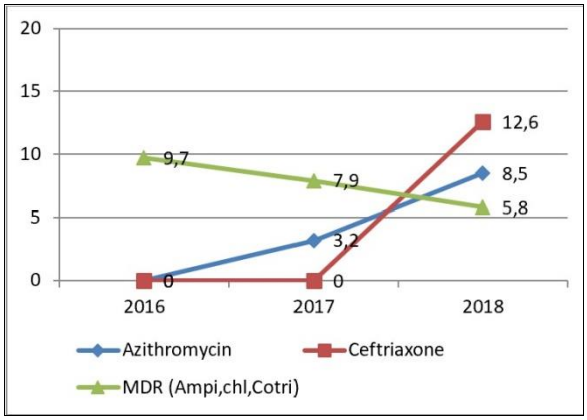

Figure 1. Changing trend of antibiotic sensitivity patterns of Salmonella Typhi (% resistance).

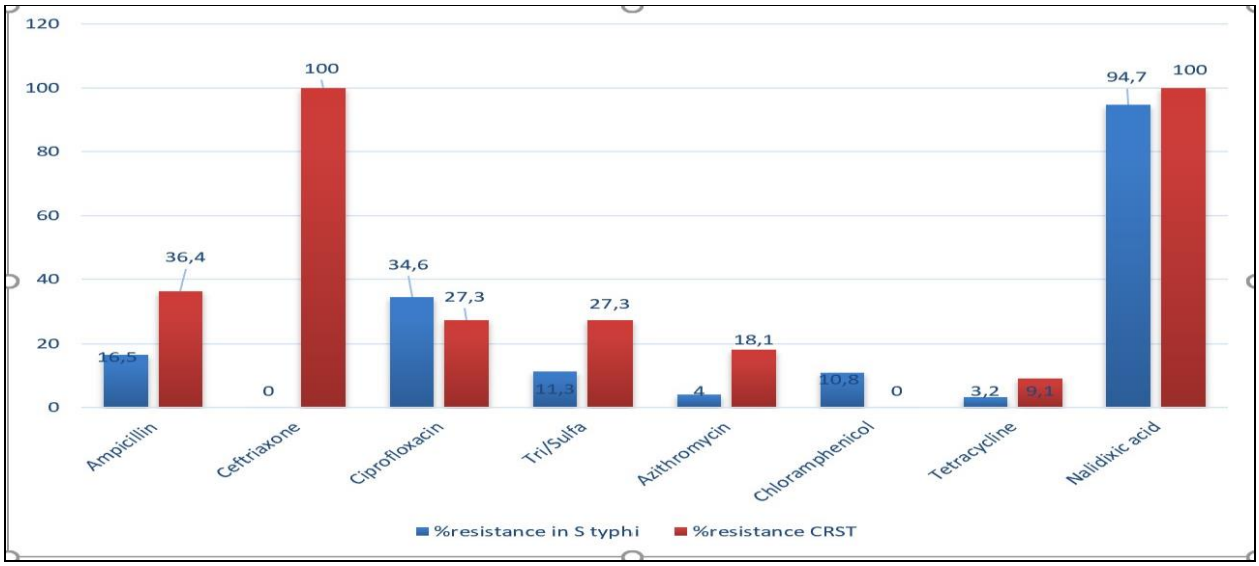

Figure 2. Resistance profile of CRST and Salmonella Typhi.

Table 1. Isolation rates of Salmonella.

Year	Total number of blood cultures	Total number of isolates (%)	S. Typhi (%)	S. Paratyphi A (%)
2016	15,111	1805 (12)	72 (3.9)	12 (0.7)
2017	16,979	2178 (13)	89(4)	5 (0.3)
2018	18,389	1824 (10)	87(4.7)	18(1)
Total	50,479	5807(11.5)	248 (4.3)	36 (0.6)

Table 2: Resistance pattern of *Salmonella Typhi* 2016-2108

Antibiotic	2016, n=72(%)	2017, n= 89 (%)	2018, n=87 (%)	Cumulative resistance, n=248 (%)	Resistant pattern of CRST
Ampicillin	10 (13.8)	15 (16.9)	16 (18.4)	41 (16.5)	4 (36.4)
Ceftriaxone	0 (0)	0 (0)	11 (12.6)	11 (4.4)	11 (100)
Ciprofloxacin	22 (30.5)	33 (37)	31 (35.6)	86 (34.6)	3 (27.3)
Trimetoprim/Sulphametaxozol	7 (9.7)	13 (14.6)	8 (9.1)	28 (11.3)	3 (27.3)
Azithromycin	0 (0)	3 (3.3)	7 (8)	10 (4)	2 (18.1)
Chloramphenicol	10 (13.8)	9 (10.1)	8 (9.2)	27 (10.8)	0 (0)
Tetracycline	0	0	8 (9.2)	8 (3.2)	1 (9.1
Nalidixic acid	66 (91.7)	85 (95.5)	84 (96.5)	235 (94.7)	11 (100)
MDR*	7 (9.7)	7 (7.9)	5 (5.8)	19 (7.7)	0 (0)
Cefixime	0 (0)	0 (0)	0 (0)	3 (27.3)	3 (27.3)

*Strains of *S. Typhi* resistant to ampicillin, chloramphenicol and trimethoprim-sulphamethoxazol.

Table 3. Clinical characteristics of Ceftriaxone resistant *Salmonella* Typhi

Variables	Number (%)
Age mean (Pediatrics; n=8)	7.18 (0.25-14 yrs)
Age mean (Adult; n=3)	38.66 (28-45 yrs)
Duration of fever, mean (range)	11.3 (7-20) days
Setting urban	9 (81)
Complications	
Intestinal perforation	1 (9)
Pancytopenia	11(100)
Idiopathic cytopenic purpura	1(9)
Septicemia with different bacteria	3(27)
Pneumonia	1(9)
Massive hepatosplenomegaly	7 (63)
Septic arthritis	1(9)
Antibiotics prescribed (Mean days of therapy & range)	
Ceftriaxone	9 (11.1 {10-17 days})
Meropenem	4 (8.5 {5-14 days})
Azithromycin	6 (11.6{8-14 days})
Cefixime	11 (12.4 {10-14 days})
Doxycycline	1 (7 days)

DISCUSSION

Over the past few years, there has been a decline in the isolation rates of MDR Salmonella Typhi [3,5-8]. At our center, the isolation rates of multidrug-resistant Salmonella Typhi have shown a decline from 9.7% in 2016 to 5.8 % in 2018. Trend analysis from Vellore has shown a decline from 50% to 5% over a period of 15 years from 2000 to 2015 [3]. MDR strains

accounted for 5% of all S. Typhi analyzed in 2017 by the ICMR surveillance network (6). Earlier Chande et al. from Central India in 2002 have also reported a low isolation rate in MDR isolates (22%) [7]. A study from Delhi also found a reduction in MDR rates from 36% in 1999 to 8% in 2008 [8].

Another systemic review from Asia has reported a decline in MDR strains in Asia to <20% in 2011-2015 while resistance to nalidixic acid and fluoroquinolone continued to increase from 20% in 2001-2005 to 65% in 2011-2015 [5]. A study from Indonesia has also reported MDR rates below 5% [9]. A retrospective study from Nepal in 2017 has reported a decline in MDR strains over a period of 23 years [10). Studies from Indonesia, Bangladesh, and Vietnam in 2017 have reported low MDR rates of 1.8% to 4.3 % over a span of five years [9,11-13].

Ampicillin resistance rates have remained below 20% at our center. Studies from India have reported highly variable rates of ampicillin resistance ranging from 3.7% to 34.1% [3,14-18]. A study from Delhi in 2006 has reported a 98.2% sensitivity rate to ampicillin [19]. Another study from Delhi has reported a decline in ampicillin resistance rates from 36% in 1999 to 8% in 2008 [8].

Resistance rates against trimethoprim-sulphamethoxazole and chloramphenicol have remained around 10% in the past three years. Most studies from India have reported similar rates [15,21]. Resistance rates of 3.6 % against chloramphenicol and 30.4% against trimethoprim-sulphamethoxazole have been reported from Delhi in 2006 [19]. A reduction in chloramphenicol resistance rates from 10% in 2008 to 36% in 1999 has been reported by Raveendran et al. from Delhi. They also reported a decrease in resistance to trimethoprim-sulphamethoxazole from 36% to 24%]8]. Patel et al. from Varanasi have reported resistance of 19.1% to trimethoprim-sulphamethoxazole and 2.1 % to chloramphenicol in 2011-13 [21]. ICMR surveillance network data for 2017 reported resistance rates of 4%, 7.6%, and 22.5 % to chloramphenicol, ampicillin, and trimethoprim-sulphamethoxazole respectively [6].

In our study, we found >90% of isolates were resistant to nalidixic acid-resistant Salmonella Typhi (NARST). Resistance to ciprofloxacin has also remained above 30%. Similar trends have been reported by other authors as well. Variable but high resistance rates to nalidixic acid have been reported from India ranging from 51% to 100% [8,15-18, 22]. ICMR surveillance network reported ciprofloxacin and pefloxacin resistance at 37% and 73.3% respectively in 2017 [6].

In the past few years, resistance to 3rd generation cephalosporin has increased in Asia from 1.5% in 2006-2010 to 4% in 2011-2015 [5]. Sporadic cases of azithromycin and ceftriaxone-resistant S. Typhi have been reported from Pakistan, Bangladesh, and India [3,21,25,26]. Ceftriaxone-resistant Salmonella Typhi was initially detected in Hyderabad, Pakistan [27]. ICMR surveillance network reported 0.4% isolates resistant to ceftriaxone [6). Raza et al. from Nepal reported 4.3% in 2012 while another study from Vellore in 2015 reported 2.4 % isolates resistant to ceftriaxone [3,20]. Chandane et al. have reported the CRST rate of 9.3% in Mumbai while another study from Gangetic plains reported 23.4% resistance to ceftriaxone [21].

S. Typhi isolates resistant to azithromycin have been reported from South Africa and Bangladesh in the past two years [26,29]. Our study has also found a rising trend in azithromycin resistance (Figure 2). Patel et al. in 2017 have reported 21.3 % isolates resistant to azithromycin from Varanasi [21].

Cases of typhoid fever caused by CRST have been associated with more complications, prolonged antibiotic treatments, and more duration of hospitalizations thereby increasing the costs [25]. Similar trends were observed in or study also.

Patient demographics like age, sex, and clinical presentation of our cases were found to be similar to Qamar et al. and Yousafzai et al. [25,27]. The mean duration of fever was found to be for 14 days by Yousafzai et al. in contrast to 11.3 days in our study [25]. Lack of any relevant travel history, absence of any clustering over time and space indicated that these cases were not part of an outbreak but seem to be sporadically present in the community. This is in sharp contrast to earlier studies that have reported CRST as an outbreak [25,27].

Conclusion

The emergence of strains resistant to ceftriaxone and azithromycin clearly indicates the time to revisit our treatment strategies. Infection with such strains predisposes patients to develop complications and will need treatment with newer antibiotics.

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