Prevalence and current trends of antimicrobial resistance among Salmonella Typhi and Salmonella Paratyphi A in children

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ABSTRACT

Objective: To determine the frequency of Salmonella typhi and Salmonella Paratyphi A causing enteric fever among children and recent trends of antimicrobial resistance among S.typhi and Paratyphi A to selected antibiotics.
Study Design: Cross sectional observational study
Place and Duration: At Department of Pathology Zubaida Medical Centre, Karachi from January 25th 2017 till 15th June 2018
Methodology: Positive blood samples of Salmonella were inoculated in Brain heart infusion broth and Blood and MacConkey agar for subculture. Age, gender, Salmonella serovar and resistance pattern to seven antibiotics were recorded. Salmonella typhi and Salmonella Paratyphi A were confirmed by using agglutinin serum against O and H antigens. Antimicrobial susceptibility was performed by Kirby- Bauer disk diffusion method
Results: Out of 125 positive blood cultures, 49% Salmonella serovars were isolated. Salmonella typhi (86%) was the most frequent organism isolated and 73.6% resistant Salmonella strains to different antibiotics were found. Overall resistance for both Salmonella typhi and Salmonella Paratyphi A was observed 64% from ampicillin, 63% from chloramphenicol 39% from ciprofloxacin and 28% from third generation ceftriaxone and cefixime.
Conclusion: The resistance to third generation cephalosporins among Salmonella typhi is considerably high. .
Keywords: Typhoid fever, Children, Salmonella typhi, Salmonella paratyphi A, Antibiotic resistance, Third generation cephalosporins, Ceftriaxone, Cefixime

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INTRODUCTION

Typhoid fever or enteric fever is a clinical condition caused by gram negative, facultative anaerobic bacilli, Salmonella, causing approximately 222,000 deaths per year globally[1]. In 2014, World health organization (WHO) documented about 0.2 million deaths due to typhoid fever and most of them occurred in Asia[2]. Pakistan is considered as most ill- famed country along with India, Indonesia and Nepal, in terms of typhoid fever[3] Also high mortality and morbidity have been noticed in children due to typhoid fever especially in Asia and Africa[4,5]. A high incidence rate of enteric fever among children aged between 2-5 years has been reported in India and Pakistan[6]. A current analysis in Asian countries also revealed 12 – 20 million typhoid cases and approximately 30% of mortality and about 90% if untreated[7,8]. Moreover emergence of multi drug resistance (MDR) in Salmonella strains is important phenomenon leading towards the disastrous situation[9].

The human restricted Salmonella pathogen is broadly classified as Salmonella enterica serovar typhi causes typhoid fever and Salmonella enterica serovar Para typhi A, B and C cause paratyphoid fever via oro-fecal route[10]. Traditionally typhoid fever has been treated with ampicillin, chloramphenicol and trimethoprim sulfamethaxaole as first line therapy. But the effectiveness of these drugs became questionable due to emergence of drug resistance back in 1970 and 1980 and resistant strains are known as multi drug resistant (MDR)[11]. In the MDR infection region, Floroquinolones as second line drug was the suitable choice but high resistance to Floroquinolnes have also been documented[12-14]. This trend is more obvious in

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South Asia[15]. Currently, third generation cephalosporin: ceftriaxone and macrolide azithromycin are used where other options are not available. But point of concern is the emerging resistance against third generation cephalosporins[16,17]. As a consequences, less antibiotic treatment is available for typhoid and it became nightmare for the physicians[18,19]. Unfortunately in Pakistan health authorities have reported extensively drug resistance (XDR) salmonella strain in Hyderabad and Karachi in 2016[11,18].

In this scenario of emerging extensively drug resistance, it is required to provide awareness and knowledge regarding the spread, symptoms and treatment strategies of organism. To curtail the situation present study was designed which would be beneficial in generating awareness about the microbe and prevailing issue of drug resistance. We conducted this study with an objective to determine the frequency of Salmonella typhi and Salmonella Paratyphi A causing enteric fever among children and recent trends of antimicrobial resistance among S.typhi and Paratyphi A to selected antibiotics.

METHODOLOGY

This cross sectional observational study was conducted by obtaining samples from Zubaida medical centre and Civil hospital Karachi from 25th January 2017 to 15th June 2018. Blood sample of the patients aged between 0 to 15 years, who attended outpatient department (OPD) or admitted in the ward with, complains of high grade fever lasting for 72 hours or more along with or without abdominal pain were taken and only positive blood cultures of Salmonella typhi and Salmonella Paratyphi A were included in this study. All the patients suffering from fever lasting for less than 72 hours, any congenital abnormality and any tumor or malignancy were exluded from the study.

About 5 ml of blood was drawn and inoculated into brain heart infusion broth and incubated for 24-48 hours. Subculture was performed on Blood agar and MacConkey agar (Oxoid) according to CLSI guidelines[20] and plates were observed after 24 hours. Brain heart infusion broth was observed for turbidity for 7 days. Further biochemical test was performed and Salmonella species were confirmed by using specific 0 and H antisera (Remel Europe). Widal antigen H was added to both positive and negative control placed on agglutination slide and drop of H,O,AH, were added to the reaction circles already containing patient`s serum. Slides were observed for agglutination reaction.

Antimicrobial potential of Salmonella serovars were determined on Mueller –Hinton agar (Oxoid) by Kirby- Bauer disk diffusion method[21] by using bacterial suspension of 0.5 McFarland index and results were interpreted according to CLSI 2011[19]. Antibiotics discs of following concentrations were used; Ampicillin (10µg), Amoxicillin, (25µg), Chloromphenicol, (30µg), Ciprofloxacin, (5µg), Ceftriaxone (30µg), Ceftazidime(30µg) and Cefixime(5µg).

Data Analysis: Results were tabulated and analyzed by using SPSS 20. . Gender, number and type of isolates and number of resistant drugs (categorical variables) were studied and calculated in frequency and percentages.

RESULTS

A total of 125 (17%) blood samples showed positive growth for Salmonella, among 125 positive samples, 46% (n=57) were collected from female and 54% (n=68) were from male patients. About 49% (n=61) organisms were isolated from age group ranged from 0-5 years followed by 40% (n=50) from 5-10 years group and 11% (n=14) from 10-15 years. In 88% (110) of cases, Salmonella typhi was isolated while 12% (15) were positive for S. Paratyphi A. (Fig-1)

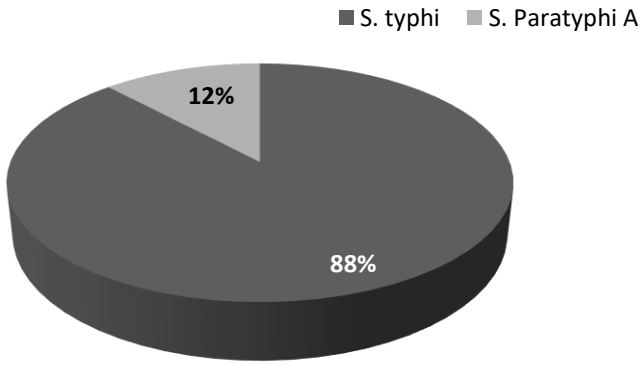

Fig-1: Frequency of isolated Salmonella species (N=125)

S.typhi was observed as the most frequent and resistant isolated organism. S.typhi exhibited high resistance against ampicillin (71% n=78) and chloramphenicol (65% n= 72). About 40% (n=44) of S.typhi was observed resistant to ciprofloxacin whereas 33 % (n=36) showed resistance to ceftriaxone. On the other hand 40% (n=6) of S. Paratyphi A showed resistance to ciprofloxacin and none of the S.Paratyphi A isolate was observed resistant to ceftriaxone. (Fig-2)

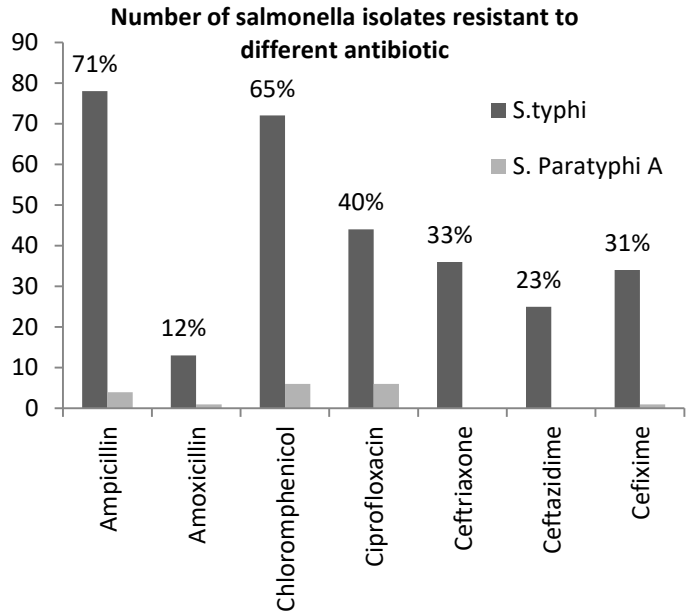

Fig-2: Frequency of Resistance pattern of Salmonella species to different antibiotics (N=125)

Out of 125 positive samples 73.6% (n=92) Salmonella strains showed resistance to one or more antibiotics tested in present study. Among 125, only one strains of Salmonella showed resistance to all tested drugs. Both S.typhi and S.Paratyphi A showed 64%resistance to ampicillin, 63% to chloramphenicol and 39% to Ciprofloxacin. Resistance to third generation cephalosporin also observed as 28% to ceftriaxone and cefixime and 21 % to ceftazidime.

DISCUSSION

In our study, S.typhi was observed as most common isolated organism (88%) followed by S.Paratyphi A (12%). In 2014, Singhal et al[22] observed 83.8% S.typhi. However a study conducted in tertiary hospital of Islamabad showed 78% and 21% results respectively[23]. Many other studies also showed that typhoid fever has become a serious health concern across Asian countries[7,8,24]. The increased frequency of S.typhi in our study may be attributed to improper infrastructure for pure drinking water supply and sewerage, habits of eating unhealthy foodstuff, lack of personal hygiene etc. Furthermore, samples were collected from the tertiary care hospital where large numbers of referral cases from other areas of the city were also received.

During the study prevalence of typhoid fever was observed more in preschool age children (48.8%). This is most probably due to low immunity or frequent exposure to fecal pathogens. Our results are similar to study conducted by Crump and colleagues[25] and Hasan et al[26]. However a research conducted in Quetta Pakistan revealed incidence more in school age going children[6].

Antimicrobial resistance against microbes is now become the utmost problem across the world[27]. This trend of antimicrobial resistance can be seen among Salmonella serovars[17,22,24]. Hasan and his co-mates observed upto 48% resistant S.typhi strains[26]. Our study also revealed 73.6% resistant Salmonella strains and high resistance against ampicillin (64%) and chloramphenicol (63%). Ochiai et al[28] also demonstrated similar findings while Ali et al demonstrated 42% resistance[23]. However, some studies reflect re-emergence of susceptibility to conventional therapy[29,30]. Such variation in susceptibility to conventional drugs is probably due to its limited use in typhoid treatment in some areas[22,31].

Due to documented resistance to conventional drugs, many researchers advocated the use of floroquinolones[32]. However, resistance to florouinolones is increased and well documented now[33]. In this study Ciprofloxacin resistance was observed 39% whereas in other studies resistance was observed 94%[23]and 60%[34]. This resistance is most likely due to frequent use of un-prescribed antibiotics for every fever, flu, diarrhea etc in Pakistan[35]. Moreover self-medication and prescription of antibiotics just to satisfy patient and patient`s parents/guardians and to gain commission or profit from the pharmaceutical agencies are other major contributing factors.

Third generation cephalosporin is the empirical treatment available after the development of resistance against conventional drugs and floroquinoles[19]. Even though resistance to ceftriaxone and cefixime has also been observed[18,34].

Hence emerging resistant strains of Salmonella typhi and Salmonella Paratyphi A to multiple drugs have now become a major challenge[36]. In our study resistance among third generation cephalosporins: ceftriaxone and cefixime was reported considerably high (28%). Moreover in 2016 and 2017 large number of extensively drug resistant (XDR) S.typhi have been reported in Hyderabad and Karachi region[11]. In mega city like Karachi, lack of basic human necessities, unsatisfactory condition of sewerage and drinking water and irrational use of antibiotics are creating such catastrophic nick.

CONCLUSION

The resistance to third generation cephalosporins among Salmonella typhi is considerably high. .

RECOMMENDATIONS

In view of above study we recommend proper induction of antibiotics along with the mandatory susceptibility testing.

CONTRIBUTION OF AUTHORS

Iqbal M: Conceived idea, Designed research methodology, Statistical analysis, Data interpretation, Manuscript writing,
Mirza S: Statistical analysis, Data interpretation, Manuscript writing
Hassan MA: Literature review
Srichand K: Literature review, Statistical analysis
Bugti S: Manuscript final reading and approval

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