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INCREASING AZITHROMYCIN RESISTANCE IN SALMONELLA TYPHI: ARE WE HEADING TOWARDS A CATASTROPHE

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Abstract

Background: Typhoid fever continues to remain a major public health problem in low to middle income countries (LMIC) where increasing resistance to antibiotics, our primary defense against typhoid fever, is posing a serious challenge in its management. We aimed this study to assess the latest antimicrobial resistance (AMR) pattern of salmonella typhi and paratyphi isolated from pediatric typhoid fever cases between 2019-2023.

Methods: This retrospective cross sectional observational study included assessing antibiotic sensitivity and resistance pattern of salmonella isolates from pediatric enteric fever cases against 10 antibiotics, tested by quality control disc diffusion method.

Results: Salmonella typhi was the predominant isolate 98.9% (91/92). These isolates showed persistently high resistance to conventional first line antibiotics (ampicillin 53.8% , cotrimoxazole 53.8% and chloramphenicol 50.5%) and ciprofloxacin (65.9%). Azithromycin resistance (AziR) was observed in 48.3%. The isolates showed 100% sensitivity to ceftriaxone, cefotaxime and meropenem.

Conclusion: Salmonella typhi continues to show high rates of resistance to conventional first line antibiotics and ciprofloxacin. A high percentage of isolates were azithromycin resistance in our study and this may pose a serious challenge in the out-patient treatment of uncomplicated enteric fever cases. There is strong need for continuous surveillance of AMR of salmonella species.

Keywords: Salmonella typhi, antimicrobial resistance pattern, Azithromycin resistance, North India.

Introduction

Global burden of Enteric fever was estimated at approximately 14.3 million cases in 2017, a decline from 25.9 million cases in 1990 [1]. In developed countries, improvement primarily in water, sanitation and hygiene (WASH) alongwith the use of antibiotics lead to a decrease in typhoid related mortality from approximately 30% (in early 1900s) to <1% [2]. Currently majority of the disease burden is concentrated in South-east Asia and sub-Saharan Africa where there has not been a significant improvement in implementation of WASH and antibiotics remain the only weapon to fight this disease [2]. Salmonella

Spp. are notable for the ongoing emergence of heterogeneous new patterns of drug resistance [1]. The emergence of extremely drug resistance (XDR) salmonella typhi (S.Typhi) has severely limited therapeutic options and a potential threat of global spread [3]. Both Centers for Disease Control and Prevention (CDC) and World Health Organization (WHO) recommend Antimicrobial resistance (AMR) surveillance for Salmonella typhi and paratyphi [4,5]. Moreover, knowledge of the local antibiogram pattern is crucial for the management of enteric fever. Therefore, we planned a retrospective study to analyze the pattern of antibiotic resistance in salmonella spp over the last 5 years.

Materials and methods

This retrospective cross-sectional observational study was conducted in the department of pediatrics at Sher-e-Kashmir Institute of Medical Sciences (SKIMS) medical college and hospital, Jammu and Kashmir, India. The hospital is located in the capital city, Srinagar (Bemina) and caters both urban and rural population. Case records of children between 6 months to 18 years of age, admitted between January 2019 till December 2023, were analyzed. Only Culture proven enteric fever cases were included in the study.

Five to ten mililiters of venous blood was drawn aseptically from children above 3 years of age while in children below 3 years 3-5 ml of blood was withdrawn and inoculated into 50 ml of brain heart infusion maintaining a temperature of 37°C. Blood culture broths were incubated for 7 days and subcultures were performed at 24 and 48 hours and then after 7 days. All bottles were also examined daily, and if a bottle showed visible signs of growth, subcultures onto sheep blood agar, MacConkey's agar and Xylose-Lysine Deoxycholate (XLD Hi Media, India) agar were performed. The transparent pale colonies on MacConkey agar and the red colonies with a black center on the XLD agar were selected for the pure culture of the bacteria. These colonies were subcultured to the XLD agar plate through the streak plate method. These colonies were further subjected to species level identification by standard biochemical tests (glucose. Lactose fermentation and production of gas, indole and hydrogen sulphide production, citrate utilization, motility and ability to split urea). The Triple Sugar Iron (TSI) agar was checked for the production of H2S gas and alkalinity. The TSI agar showed alkaline/ acidic reaction in case of salmonella typhi with a spec of H2S gas on the slant and butt of the media while salmonella paratyphi showed only alkaline/acidic reaction without H2S production (Table I). Finally, the colonies were confirmed as salmonella typhi with the help of slide agglutination with salmonella 09 and Vi- specific antisera while salmonella paratyphi A was confirmed with agglutination with 02 antisera.

Table I: Biochemical identification of Salmonella Spp.

TEST	REACTION
Indole test	-
Catalase test	+
MR test	+
Oxidase test	-
Citrate test	+
Urease test	-
Voges Proskauer (VP) test	-
H_2S production (TSI)	+
Motility	+

Antimicrobial susceptibility testing

The isolates were tested for susceptibility to ampicillin [10µg], chloramphenicol [30µg], ciprofloxacin[5µg],meropenem[10µg],piperacillin/tazobactam[100/10µg],cotrimoxazole(trimethoprim/sulfamethoxazole[1.25/23.75µg]),ceftriaxone[30µg], cefotaxime [30µg], cefixime [5 µg] and

Azithromycin [15 µg] on Mueller-Hinton agar plates by Kirby Bauer disc diffusion method with standard antimicrobial discs. Overnight cultures grown on nutrient broth (cultures were adjusted to 0.5 McFarland units) were spread evenly on Mueller-Hinton agar (Hi media, India). The respective antibiotic discs were placed on the culture plates. The plates were incubated at 35 °C for 24 h and inhibition zones were measured. The sensitivity and resistance of the isolates towards the antibiotics were determined as per the criteria of the Clinical Laboratory Standards Institute (CLSI) and European Committee on Antimicrobial susceptibility testing [6,7]. Isolates resistant to ampicillin, chloramphenicol and co-trimoxazole were defined as multidrug- resistant (MDR) salmonella with additional resistance to ciprofloxacin and ceftrixone were defined as extremely drug-resistant (XDR) cases.

Results

Total number of culture proven enteric fever cases admitted from January 2019 to December 2023 were 92 with Salmonella typhi (S.typhi) and Paratyphi isolated from 91(98.9%) and 1(1.1%) cases respectively. Table II shows the number of culture proven Salmonella typhi cases in each year from 2019 to 2023 and the antimicrobial resistance (AMR) pattern in each corresponding year. Table III shows the clinico-demographic profile of these cases.

Salmonella typhi isolates showed high resistance rate to first line anti-typhoid antibiotics ampicillin (53.8%), chloramphenicol (50.5%) and cotrimoxazole (53.8%). Fifty percent of the isolates were MDR. Approximately 66% of the isolates showed resistance to ciprofloxacin. Azithromycin resistance (AziR) was also high 44(48.3%) cases. Resistance to cefixime and piperacillin-Tazobactum was observed in 22 (24.2%) cases each. Salmonella typhi showed 100% sensitivity to ceftriaxone, cefotaxime and meropenem throughout study period. Single isolate of Salmonella Paratyphi A showed sensitivity to ciprofloxacin, piperacillin-tazobactum and meropenem only.

Table II: Culture proven S.Typhi cases per year and their antimicrobial resistance pattern

Antibiotics/year	**2019 (n=23)**	**2020 (n=11)**	**2021 (n=9)**	**2022 (n=30)**	**2023 (n=18)**	**Total (n=91)**
Ampicillin	12 (52.2%)	7 (63.6%)	6 (66.7%)	13 (43.3%)	11 (61.3%)	49 (53.8%)
Chlorempheni col	13 (56.5%)	5 (45.5%)	5 (55.5%)	14 (46.7)	9 (50%)	46 (50.5%)
Cotrimoxazole	12 (52.2%)	5 (45.4%)	5 (55.6%)	15 (50%)	12 (66.7%)	49 (53.8%)
Ciprofloxacin	14 (60.8%)	8 (72.7%)	6 (66.7%)	19 (63.3%)	13 (72.3%)	60 (65.9%)
Azithromycin	9 (39.1%)	4 (36.4%)	6 (66.7%)	13 (43.3%)	12 (66.7%)	44 (48.3%)
Ceftrioxone	0	0	0	0	0	0
Cefotaxime	0	0	0	0	0	0
Cefixime	3 (13%)	0	2 (22.2%)	15 (50%)	2 (11.1%)	22 (24.2%)
Piperacillin-Tazobactum	4 (17.4%)	2 (18.2%)	1 (11.1%)	11 (36.7%)	4 (22.2%)	22 (24.2%)
Meropenam	0	0	0	0	0	0

Table III: Clinico-demographic profile of patients

Characteristics	Number (%)
Age (years) Upto 5 6-12 13-18	 23 (25.0%) 41(44.5%) 28 (30.4%)
Male Female	48 (52.2%) 44 (47.8%)
Urban: Rural	38:54 (1.4:1)
Fever History	92 (100%)
Hypothermia at admission	03 (3.3%)
Anorexia	78 (84.7%)
Coated tongue	43 (46.7%)
Diarrhea	39 (42.4%)
Vomiting	27 (29.3%)
Constipation	11 (11.9%)
Abdominal pain	32 (34.7%)
Splenomegaly	25 (27.2%)
Hepatomegaly	41 (44.6%)
Hepatitis	04 (4.3%)
Iliopsoas abscess	01 (1.1%)
Septic Shock	01 (1.1%)

Discussion

This study provides crucial information about recently evolving antimicrobial resistance (AMR) pattern of salmonella Spp. from 2019 to 2023 from north India. In this study, salmonella typhi was the predominant causative agent of enteric fever accounting for 91 (98.9%) cases. Similar results were reported by other Indian studies [8,9]. However, some studies from India and Nepal have reported Salmonella Paratyphi as the predominant isolate [10,12]. Antimicrobial resistance (AMR) pattern in our study depicted 46 (50.5%) and 60 (65.9%) isolates of S. typhi as MDR and ciprofloxacin resistant respectively. These isolates showed persistently high resistance pattern to conventional first line antibiotics (ampicillin, chloramphenicol and cotrimoxazole) and ciprofloxacin throughout the study period. Globally, widespread resistance to first line antibiotics started to appear in 1990s followed by resistance to ciprofloxacin in early 2000s [13]. However, a recent multicentric surveillance study on enteric fever in India reported a decreasing trend of resistance to first line anti-typhoid antibiotics [14]. In this study, out of 2373 Salmonella isolates, 2032 (85.6%) were identified as S. Typhi and 341 (14.4%) were S. Paratyphi A [14]. Approximately 2% of S. Typhi isolates were multidrug-resistant (MDR), whereas all 341 (100%) of S. Paratyphi A isolates were sensitive to the first-line antimicrobials [14]. Low levels of resistance to ampicillin, chloramphenicol and co-trimoxazole were also reported by other Indian studies [15, 16] and also from neighboring country, Nepal [17]. While resistance to first line antibiotics may be declining but resistance to ciprofloxacin continues to persist at a high rate as reported in recent Indian studies [14-17]. Similar high ciprofloxacin resistance was observed in our study also. Ceftriaxone resistance in S.Typhi was first reported from a patient in Bangladesh in 1999 [18]. A study from Pakistan has reported largest outbreak of ceftriaxone-resistant S. Typhi in the world [3]. Kokare et al in a study from Mumbai, India, reported 12.5% resistance to ceftriaxone in S.typhi isolates [19].However, Veeraraghavan et al in their 2 year prospective multicentric systematic surveillance study in India (November 2017 – January 2020) reported 100% sensitivity to ceftriaxone [14]. Manoharan A et al in their retrospective (2011-2020) study on 1010 Salmonellae isolates (849 S. Typhi and 161 S. Paratyphi

A) reported 100% sensitivity to cephalosporins[20].Our study also reported 100% sensitivity to ceftriaxone, cefotaxime and meropenem. A recent study from Pakistan conducted in 2020 reported 48% resistance to meropenem in Salmonella isolates [21].

All enterobacteriaceae members are resistant to macrolide antibiotics due to their hydrophobic nature and low permeability through the outer membrane except azithromycin which is active against enterobacteriaceae because of its ability to achieve very high intracellular concentration, 50-100 times more than the plasma levels, due to its basic nature and hence enhanced uptake by bacterial cells [22]. Due to this unique property of azithromycin, it was expected to act on intracellular bacteria like S. typhi [22]. Therefore, azithromycin has been in use for the out-patient treatment of uncomplicated enteric fever after the emergence of ciprofloxacin resistance [22]. Unfortunately, azithromycin resistant (AziR) strains of S.typhi, defined as MIC breakpoint of >16 µg/ml [7], have been reported from Bangladesh, Pakistan and Nepal [23-25]. Azithromycin resistance is gradually increasing in Bangladesh [2]. Sharma et al reported 34.7% azithromycin resistance in their study from 2005- 2008 [22]. A Kolkata based, Indian, study analyzing the AMR trend of 45 salmonella isolates (35 S.typhi and 10 S. paratyphi A) from 2019-2022 reported only 2.2% (1 out of 10 Paratyphi A isolates) azithromycin resistance[26]. Veeraraghaven et al and Manoharan et al reported >95% and 100% azithromycin sensitivity in their studies respectively [14,20]. On contrary, our study demonstrates a high, 48%, resistance to azithromycin. Moreover, the AMR pattern showed progressively increasing trend of AziR throughout the study period and this increase in resistance was more pronounced during the last 3 years of the study. There can be three main reasons for high azithromycin resistance in our study. First, the study was conducted in Kashmir valley where viral upper respiratory tract infections (VURTIs) are frequent almost throughout the year due to its temperate climate and azithromycin is frequently (irrationally) prescribed for this self-limiting illness in children and adults both by physicians and pharmacists, thus putting selective antibiotic pressure on azithromycin. Second, Enteric fever is one of the most common causes of undifferentiated acute febrile illness in India and patients frequently seek medical attention at peripheral health care facilities where azithromycin is often prescribed as first line treatment for uncomplicated enteric fever, considering widespread community resistance to ciprofloxacin. Moreover, its widespread availability, low cost and less frequent dosing (once a day) for short duration make it a potential candidate drug for irrational over the counter use and hence increased resistance. Third, marked increase in azithromycin resistance during the last 3 years of the study coincides with the COVID-19 pandemic, the period during which azithromycin was widely empirically used for treatment of COVID-19.

Currently the dominant clone of S. typhi in India is H58 haplotype which has two main lineages namely I and II [27]. Lineage I is MDR and was dominant in 1990s[27]. Lineage II is now more prevalent and is associated with FQ resistance [27]. These MDR isolates often carry resistance genes (*catA, sul1, sul2, dfrA, bla*$_{TEM-1}$*, strA, strB, tetA, tetB, tetC* and *tetD*) on mobile genetic elements such as IncHI1 plasmids which also have been found integrated on the chromosome of H58 S.typhi isolates from countries including India [28,29]. Mutations in *gyrA* and *parC* and to a lesser extent in *parE* and *gyrB* were responsible for FQ resistance in S.typhi isolates in India[30]. Indian isolates of cephalosporin resistant S.typhi carried IncX3, IncA and IncN plasmids which encoded bla $_{SHV-12}$, bla $_{CMY-2}$, bla $_{TEM-1B}$ and bla $_{DHA-1}$ [31,32]. Isolates from the world's largest outbreak of ceftriaxone-resistant S.typhi reported from Hydrabad city of Sindh, Pakistan in 2016 confirmed a novel strain of S.typhi belonging to H58 lineage carrying plasmid encoded additional resistance elements including bla$_{CTX-M-15}$ alongwith *qnrS* FQ resistance gene [3]. Eighty-five percent of the circulating isolates in Pakistan were XDR [2]. Studies on mechanism(s) of azithromycin resistance in S.typhi are limited. A north-Indian study described mutation in the *acrB* gene as the mechanism of azithromycin resistance in S.typhi [13]. This gene encodes a component of the AcrAB efflux pump[13]. Similar observations were reported in studies from Bangladesh and Pakistan[23,33]. However, azithromycin-resistant isolates from India were phylogenetically distinct (arisen independently) from those reported from Pakistan and Bangladesh [13].

To our knowledge this is the first study from India to report such high rate of AziR in S.typhi isolates. There are two important limitations of our study. First, being a single center study, data mentioned in the study represents regional AMR pattern of S.typhi and may not be the representative data from a larger geographical area. Second, drug history was incomplete in most case files.

Conclusion

This study provides crucial information about recently evolving antimicrobial resistance (AMR) pattern of S. Typhi in India. This is probably the first study from India to report such a high rate of Azithromycin resistance. Spread of azithromycin resistance would require treatment of uncomplicated enteric fever cases with parenteral antibiotics and hence, inpatient department treatment, putting an extra burden on already struggling health care system in LMICs. Acquisition of cephalosporin-resistance by AziR strain or azithromycin-resistance by XDR strain can pose serious threats to the health care system globally and LMICs in particular. To tackle this potentially imminent threat there is urgent need for a multipronged approach which includes (i) Improvement in water, sanitation and hygiene (WASH) interventions (ii) introduction of Typhoid conjugate vaccine (TCV) in endemic regions (iii) typhoid surveillance to monitor the emergence and spread of other AMR phenotypes (iv) local antibiotic stewardship, that is, formulating guidelines for empirical treatment based on local data (v) policy on prescription and sale of antibiotics by local health authority. Based on the findings of our study we recommend the use of 3rd-generation cephalosporins (cefixime and ceftriaxone) for the empirical treatment of enteric fever. Ciprofloxacin in adults and Azithromycin in both children and adults should not be empirically used for enteric fever treatment.

What is already known:

Salmonella typhi continues to have high rate of resistance against Ampicillin, Cotrimoxazole, Chloremphenicol and Ciprofloxacin.

What this study adds:

High rate of resistance against azithromycin is evolving in north India.

References

1. GRAM Typhoid Collaborators. Estimating the subnational prevalence of antimicrobial resistant Salmonella enterica serovars Typhi and Paratyphi A infections in 75 endemic countries, 1990-2019: a modelling study. Lancet Glob Health. 2024 Mar;12(3):e406-e418. doi: 10.1016/S2214-109X(23)00585-5. PMID: 38365414; PMCID: PMC10882211.
2. Senjuti Saha, Mohammad Saiful Islam Sajib, Denise Garrett, Farah N Qamar. Antimicrobial Resistance in Typhoidal *Salmonella*: Around the World in 3 Days. *Clinical Infectious Diseases*.2020 Aug 15; Vol 71(Issue Supplement 2): 91–95.
3. Yousafzai MT, Qamar FN, Shakoor S, Saleem K, Lohana H, Karim S, et al. Ceftriaxone-resistant Salmonella Typhi outbreak in Hyderabad City of Sindh, Pakistan: High time for the introduction of typhoid conjugate vaccine. Clin Infect Dis 2019; 68:16-21.
4. CDC. Antibiotic Resistance Threats in the United States, 2013 | Antibiotic/ Antimicrobial Resistance | CDC. 2014 [cited 2017 Feb 3]. Available from: https://www.cdc.gov/drugresistance/threat-report-2013/
5. World Health Organisation. Vaccines and Biologicals. Background Document: The Diagnosis, Treatment and Prevention of Typhoid Fever. Geneva. 2003. Available online: https://www.glowm.com/pdf/WHO-diagnosis%20treatment%20prevention%20of%20typhoid%20fever-2003-CustomLicense.pdf (accessed on 18 April 2019).

6. Clinical and Laboratory standards Institute. Performance Standards for Antimicrobial Susceptiblity Testing: 29th Information Supplement. CLSI document M100-S29. Wayne, PA; Clinical and Laboratory Standards Institute; 2019.
7. Leclercq R, Cantón R, Brown DF, Giske CG, Heisig P, MacGowan AP, et al. EUCAST expert rules in antimicrobial susceptibility testing. Clin Microbiol Infect. 2013 Feb;19(2):141-60. doi: 10.1111/j.1469-0691.2011.03703.x. Epub 2011 Nov 25. PMID: 22117544.
8. Manchanda, V.; Bhalla, P.; Sethi, M.; Sharma, P.K. Treatment of enteric fever in children on the basis of current trends of antimicrobial susc
9. Kumar, S.; Rizvi, M.; Berry, N. Rising prevalence of enteric fever due to multidrug-resistant Salmonella: An epidemiological study. J. Med. Microbiol. 2008, 57, 1247–1250.
10. Kapil A, Sood S, Reddaiah VP, Das B, Seth P. Paratyphoid fever due to Salmonella enterica serotype Paratyphi A. Emerg Infect Dis 1997; 3: 407
11. Woods CW, Murdoch DR, Zimmerman MD, Glover WA, Basnyat B, Belbase RH, et al. Emergence of Salmonella enterica serotype Paratyphi A as a major cause of enteric fever in Kathmandu, Nepal. Trans R Soc Trop Med Hyg 2006; 100: 1063-7.
12. Shlim DR, Schwartz E, Eaton M. Clinical importance of Salmonella paratyphi A infection to enteric fever in Nepal. J Travel Med 1995; 2: 165-8.
13. Carey ME, Jain R, Yousuf M, Maes M, Dyson ZA, Thu TNH, et al. Spontaneous Emergence of Azithromycin Resistance in Independent Lineages of Salmonella Typhi in Northern India. Clin Infect Dis. 2021 Mar 1;72(5):120-127.
14. Veeraraghavan B, Pragasam AK, Ray P, Kapil A, Nagaraj S, Perumal SPB, et al. Evaluation of Antimicrobial Susceptibility Profile in Salmonella Typhi and Salmonella Paratyphi A: Presenting the Current Scenario in India and Strategy for Future Management. J Infect Dis. 2021 Nov 23;224(Supple 5):502-516.
15. Wattal C, Goel N. Pediatric Blood Cultures and Antibiotic Resistance: An Overview. Indian J Pediatr. 2020 Feb;87(2):125-131. doi: 10.1007/s12098-019-03123-y. Epub 2019 Dec 21. Erratum in: Indian J Pediatr. 2020 Jun;87(6):486.
16. Iyer RN, Jangam RR, Jacinth A, Venkatalakshmi A, Nahdi FB. Prevalence and trends in the antimicrobial susceptibility pattern of Salmonella enterica serovars Typhi and Paratyphi A among children in a pediatric tertiary care hospital in South India over a period of ten years:a retrospective study. Eur J Clin Microbiol Infect Dis. 2017;36(12):2399–04.
17. Shrestha KL, Pant ND, Bhandari R, Khatri S, Shrestha B, Lekhak B: Re-emergence of the susceptibility of the Salmonella spp. isolated from blood samples to conventional first line antibiotics. Antimicrob Resist Infect Control. 2016, 5:22.
18. Saha SK, Talukder SY, Islam M, Saha S. A highly ceftriaxone-resistant Salmonella Typhi in Bangladesh. Pediatr Infect Dis J 1999; 18:387.
19. Kokare RS, Bari AK, Pereira JV, Patel K, Poojary A. Minimum inhibitory concentration (MIC) of Ceftriaxone and Azithromycin for blood culture isolates of Salmonella enterica spp. J Infect Dev Ctries. 2021 Apr 30;15(4):538-543.
20. Manoharan A, Dey D, Putlibai S, Ramaiah S, Anbarasu A, Balasubramanian S. Epidemiology of Multidrug Resistance among Salmonella enterica serovars typhi and paratyphi A at a Tertiary Pediatric Hospital in India Over a Decade; In-silico Approach to Elucidate the Molecular Mechanism of Quinolone Resistance. Int J Infect Dis. 2022 Jun;119:146-149.
21. Ali Shah SA, Nadeem M, Syed SA, Fatima Abidi ST, Khan N, Bano N. Antimicrobial Sensitivity Pattern of Salmonella Typhi: Emergence of Resistant Strains. Cureus. 2020 Nov 29;12(11): e11778.
22. Sharma P, Kumari B, Dahiya S, Kulsum U, Kumar S, Manral N, Pandey S, Kaur P, Sood S, Das BK, Kapil A. Azithromycin resistance mechanisms in typhoidal salmonellae in India: A 25 years analysis. Indian J Med Res. 2019 Mar;149(3):404-411.

23. Hooda Y, Sajib MSI, Rahman H, Luby SP, Bondy-Denomy J, Santosham M, Andrews JR, Saha SK, Saha S. Molecular mechanism of azithromycin resistance among typhoidal Salmonella strains in Bangladesh identified through passive pediatric surveillance. PLoS Negl Trop Dis. 2019 Nov 15;13(11):e0007868. doi: 10.1371/journal.pntd.0007868. PMID: 31730615; PMCID: PMC6881056.
24. Iqbal J, Dehraj IF, Carey ME, Dyson ZA, Garrett D, Seidman JC, Kabir F, Saha S, Baker S, Qamar FN. A Race against Time: Reduced Azithromycin Susceptibility in *Salmonella enterica* Serovar Typhi in Pakistan. mSphere. 2020 Jul 22;5(4):e00215-20. doi: 10.1128/mSphere.00215-20. PMID: 32699118; PMCID: PMC7376502.
25. Duy PT, Dongol S, Giri A, Nguyen To NT, Dan Thanh HN, Nhu Quynh NP, Duc Trung P, Thwaites GE, Basnyat B, Baker S, Rabaa MA, Karkey A. The emergence of azithromycin-resistant *Salmonella* Typhi in Nepal. JAC Antimicrob Resist. 2020 Dec 21;2(4):dlaa109. doi: 10.1093/jacamr/dlaa109. PMID: 34223059; PMCID: PMC8210228
26. Biswas M, Biswas S, Gupta B, Mascellino MT, Rakshit A, Chakraborty B. Changing Paradigms in Antibiotic Resistance in *Salmonella* Species with Focus on Fluoroquinolone Resistance: A 5-Year Retrospective Study of Enteric Fever in a Tertiary Care Hospital in Kolkata, India. Antibiotics (Basel). 2022 Sep 26;11(10):1308.
27. Britto CD, Dyson ZA, Duchene S, Carter MJ, Gurung M, Kelly DF, Murdoch DR, Ansari I, Thorson S, Shrestha S, Adhikari N, Dougan G, Holt KE, Pollard AJ. Laboratory and molecular surveillance of paediatric typhoidal Salmonella in Nepal: Antimicrobial resistance and implications for vaccine policy. PLoS Negl Trop Dis. 2018 Apr 23;12(4):e0006408
28. Wong VK, Baker S, Pickard DJ, Parkhill J, Page AJ, Feasey NA, et al. Phylogeographical analysis of the dominant multidrug-resistant H58 clade of Salmonella Typhi identifies inter- and intracontinental transmission events. Nat Genet 2015; 47 : 632-9
29. International Typhoid Consortium, Wong VK, Holt KE, Okoro C, Baker S, Pickard DJ, Marks F,et al. Molecular surveillance identifies multiple transmissions of typhoid in west Africa. PLoS Negl Trop Dis 2016; 10 : e0004781.
30. Britto CD, John J, Verghese VP, Pollard AJ. A systematic review of antimicrobial resistance of typhoidal *Salmonella* in India. Indian J Med Res. 2019 Feb;149(2):151-163.
31. Devanga Ragupathi NK, Muthuirulandi Sethuvel DP, Shankar BA, Munusamy E, Anandan S, Veeraraghavan B. Draft genome sequence of blaTEM-1-mediated cephalosporin-resistant Salmonella enterica serovar Typhi from bloodstream infection. J Glob Antimicrob Resist 2016; 7 : 11-2.
32. Rodrigues C, Kapil A, Sharma A, Devanga Ragupathi NK, Inbanathan FY, Veeraraghavan B, et al. Whole-Genome Shotgun Sequencing of cephalosporin-resistant Salmonella enterica serovar Typhi. Genome Announc 2017; 5: e01639-16.
33. Klemm EJ, Shakoor S, Page AJ, Qamar FN, Judge K, saeed DK, et al. Emergence of an extensively drug-resistant Salmonella enterica serovar typhi clone harboring a promiscuous plasmid encoding resistance to fluoroquinolones and third generation cephalosporins.