

## **Detection of Multidrug Resistant *Salmonella* Typhi and *Salmonella* Paratyphi A isolated from enteric fever patients, in a tertiary care hospital of Dhaka city.**

Farzana Hoque Promi<sup>1</sup>, Dr. Saika Farook<sup>2</sup>, Dr. Aunta Melan<sup>2</sup>, Dr. Shariful Alam Jilani<sup>2</sup>,  
Dr. Fahim Kabir Monjurul Haque<sup>1\*</sup>

<sup>1</sup>Microbiology Program, Department of Mathematics and Natural Science, BRAC University, 66 Mohakhali, Dhaka, Bangladesh

<sup>2</sup>KA. Monsur Laboratory, Department of Microbiology, BIRDEM General Hospital and Ibrahim Medical College, Dhaka

**Abstract:** Multidrug resistant *Salmonella* has become prevalent in most of South Asia, with a frequency ranging from 50% to 80% of all *Salmonella* Typhi and *Salmonella* Paratyphi A isolated from enteric fever patients. The positivity rate of enteric fever in Bangladesh is estimated to be 3.9%. The present study was undertaken to detect multidrug resistant *S. Typhi* and *S. Paratyphi A* isolated from blood samples of enteric fever patients from a tertiary care hospital in Dhaka City. A total of 12,000 blood samples were collected from November 2022 to May 2023, from clinically suspected patients. Blood culture was positive for *Salmonella* in 313 samples. Out of 313 isolates, 50 were randomly selected and *Salmonella* was confirmed by PCR targeting the *InvA* gene. Among these 313, biochemical and serological tests detected 263 isolates as *S. Typhi* and 50 as *S. Paratyphi A*. Antimicrobial susceptibility was conducted by Kirby–Bauer method for chloramphenicol, ampicillin, co-amoxiclav, trimethoprim-sulfamethoxazole, ciprofloxacin, cefixime, cefepime, ceftriaxone, meropenem, azithromycin and colistin. MDR *Salmonella*, defined as a combined resistance against three first-line antimicrobial agents, ampicillin, chloramphenicol and trimethoprim-sulfamethoxazole were detected in 42 strains of *S. Typhi*. Moreover, all isolates except two *S. Typhi* and one *S. Paratyphi A* of *Salmonella* were resistant to nalidixic acid and ciprofloxacin. The emergence of MDR *Salmonella* Typhi with increased ciprofloxacin resistance is likely to further complicate the therapy of typhoid fever.

Key words: Multidrug resistant, enteric fever, *InvA*, Kirby–Bauer.

**Introduction:** The bacterial etiology of blood infections by *Salmonella* Typhi and Paratyphi A as the most frequently isolated organisms which have a high percentage of multidrug-resistant (MDR) strains. Multidrug resistant or MDR *Salmonella* has become prevalent in most of South Asia, with a frequency ranging from 50% to 80% of all *Salmonella* Typhi and *Salmonella*

Paratyphi A isolated from enteric fever patients. The positivity rate of enteric fever in Bangladesh is estimated to be 3.9%. The aim of this project is to detect multidrug resistant *S. Typhi* and *S. Paratyphi A* isolated from blood samples of enteric fever patients from a tertiary care hospital in Dhaka City.

**Materials and Methods:** A total of 12,000 blood samples were collected from November 2022 to May 2023, from clinically suspected patients. Blood culture was positive for *Salmonella* in 313 samples. These were identified by colony morphology, standard biochemical test and agglutination test. Biochemical tests such as, Triple Sugar Iron Test, Oxidase test, Motility Indole Urease test and Citrate Utilization Test etc. Slide agglutination tests were performed to differentiate *Salmonella Paratyphi A* from *Salmonella Typhi*. To identify the presence of genus specific InvA gene in *Salmonella Typhi* and *Salmonella Paratyphi A* conventional PCR was performed, InvR and InvF primers were included (53 degree Celsius as annealing temperature) and 284 band size was taken as the positive control band, in gel electrophoresis which was confirmed from published literature.

**Results:** The results of these biochemical tests were fermented glucose, produced acid/gas, didn't ferment lactose, red slant & yellow butt, motile, urea and indole negative (the reagent remained yellow) and the media remains green in citrate test which indicate the absence of citrate permease enzyme. Ultimately, among them 313 samples were *Salmonella* positive. Out of 313 isolates, *Salmonella Typhi* and *Paratyphi* were identified by performing slide agglutination test (O and H antigen). Among these 313, biochemical and serological tests detected 263 isolates as *S. Typhi* and 50 as *S. Paratyphi A*. Out of 313 isolates, 50 were randomly selected and *Salmonella* was confirmed by PCR targeting the InvA gene. Then, the presence of InvA gene was confirmed in 313 samples. Antimicrobial susceptibility was conducted by Kirby–Bauer method for chloramphenicol, ampicillin, co-amoxiclav, trimethoprim-sulfamethoxazole, ciprofloxacin, cefixime, cefepime, ceftriaxone, meropenem, azithromycin and colistin. MDR *Salmonella*, defined as a combined resistance against three first-line antimicrobial agents, ampicillin, chloramphenicol and trimethoprim-sulfamethoxazole were detected in 42 strains of *S. Typhi*. All isolates except two *S. Typhi* and one *S. Paratyphi A* of *Salmonella* were resistant to nalidixic acid and ciprofloxacin. Out of 313 Typhoidal *Salmonella*, 263 (84.03%) were identified as *Salmonella Typhi* and 50 (15.97%) were *Salmonella Paratyphi A*. 42 (13.42%) were detected as Multidrug

resistant *Salmonella* Typhi and 70.61% were detected as non- MDR. In case of, *Salmonella* Paratyphi A isolated, all strains were found sensitive to ampicillin, trimethoprim-sulfamethoxazole and chloramphenicol (100% non-MDR). Moreover, total 42 MDR *Salmonella* species were found where resistance to ampicillin, trimethoprim-sulfamethoxazole and chloramphenicol were 15.65%, 22.36% and 21.09% respectively.

**Discussion:** The present study is compared to a study which was conducted in BIRDEM hospital, Dhaka by Dr. Nasrin Sultana (2020), where 26.82% *Salmonella* isolates from were found MDR. After statistical analysis, comparing to Dr. Nasrin Sultana MDR samples, 2.7% of MDR *Salmonella* species were increased within 2 to 3 years. In Pakistan, Muhammad Zakir et al. (2021) revealed that multidrug resistance was 24.5 % in 2021 where Syed Asim Ali Shah et al. (2019) reported that multidrug resistance was 20 % in 2019. The rate of mdr *Salmonella* increased 4.5% within 2 years.

**Conclusion:** The emergence of MDR *Salmonella* Typhi with increased ciprofloxacin resistance is likely to further complicate the therapy of typhoid fever.