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Original Research Article

Antibiotic resistance trends and diagnostic accuracy of Typhi Dot and Widal in enteric fever: a 2023-24 single-centre study from North India

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ABSTRACT

Background: Antimicrobial resistance (AMR) in enteric fever is an emerging public health challenge in India. Early and accurate diagnosis, along with appropriate antibiotic therapy, is essential for reducing disease-related morbidity.
Methods: This retrospective, single-center observational study included all culture-confirmed enteric fever cases diagnosed at Moolchand K.R. Hospital, New Delhi, between August 2023 and September 2024. A total sample size of 45 patients with positive blood cultures for *Salmonella Typhi* or *Salmonella Paratyphi* was analyzed. Antimicrobial susceptibility testing was performed using the VITEK 2 compact system with GN and AST-N cards. Diagnostic performance of WIDAL and Typhi Dot tests was evaluated descriptively using blood culture as the reference standard.
Results: Forty-five culture-confirmed cases were analyzed (mean age 22.6±14.5 years; 51.1% female). *S. Typhi* accounted for 75.6% (34/45) isolates, while *S. Paratyphi* represented 24.4% (11/45). WIDAL was positive in 20.0% (9/45) cases and demonstrated a sensitivity of 20.6% (7/34), missing 79.4% of confirmed *S. Typhi* infections. Typhi Dot testing was performed in 25 patients, and all results were negative. Third-generation cephalosporins showed high susceptibility (97.8%), whereas resistance to fluoroquinolones was common. Multidrug resistance was identified in 2.2% (1/45) of isolates.
Conclusions: *S. Typhi* remained the predominant pathogen. Third-generation cephalosporins continue to be effective, although emerging multidrug resistance warrants ongoing surveillance and antimicrobial stewardship. Blood culture remains the diagnostic gold standard, while WIDAL and Typhi Dot demonstrated limited clinical utility.

Keywords: Blood culture, Enteric fever, Salmonella typhi, Typhi dot, WIDAL test

INTRODUCTION

Typhoid fever remains a major public health problem in South Asia despite improved sanitation and vaccine availability. India accounts for one of the highest burdens globally, with an estimated 9 million cases and 110,000 deaths annually (WHO).

The emergence of multidrug-resistant (MDR) and extensively drug-resistant (XDR) *Salmonella Typhi* threatens effective therapy and elevates case fatality rates. At the same time, the front-line laboratory tests used in many centers principally the WIDAL slide agglutination assay and Typhi Dot IgM provide variable accuracy, leading to both under- and overtreatment.[1]

Our center routinely maintains an electronic microbiology database, allowing contemporary assessment of resistance trends and diagnostic performance. We therefore undertook a retrospective analysis to describe the antibiogram of *Salmonella* spp. isolated from August 2023 to September 2024, and to compare WIDAL and Typhi Dot results with blood culture, the accepted gold standard.

METHODS

A retrospective, single-center, observational study was conducted at Moolchand K.R. Hospital, a tertiary care hospital in New Delhi, India.

The study included a total sample size of 45 patients with culture-confirmed enteric fever diagnosed during the study period. All eligible cases meeting the inclusion criteria were included by consecutive sampling.

Case definition and inclusion criteria

We included all patients (any age, either sex) with one positive blood culture isolate of *S. Typhi* or *S. Para typhi* between August 2023 and September 2024. Only the first isolate per patient episode was analyzed.

Data collection

Demographic variables, age, sex, WIDAL titers, Typhi Dot results, and full antibiotic susceptibility panels were extracted from the laboratory information system.

Microbiological procedures

Widal test

Slide agglutination was performed, and the presence of visible agglutination within one minute was considered a positive result. The interpretation of the test results was carried out according to the manufacturer's instructions.[2]

Typhi Dot test

Rapid immunoassay detecting IgM antibodies against *S. Typhi* using a lateral flow device. A visible pink/purple line within 15 minutes was positive.[3]

Blood culture

Samples were inoculated in aerobic bottles and incubated in Bac-T/ALERT 3D system. Positive samples were sub-cultured and identified by morphology, gram stain, biochemical tests, and serotyping.[4]

Antimicrobial susceptibility

Tested via VITEK 2 Compact with GN and AST-N cards, interpreted per CLSI guidelines. MDR is defined as resistance to chloramphenicol, ampicillin, and trimethoprim-sulfamethoxazole; XDR is defined as resistance to first-line agents plus fluoroquinolones and third-generation cephalosporins.[2]

Statistical analysis

Data compiled in Excel and analyzed using SPSS v25. Normality was assessed by the Shapiro-Wilk test, histograms, and Q-Q plots. Non-parametric tests were used due to the non-normal distribution. Descriptive stats reported as mean±SD, median with interquartile range (IQR), or frequency (%). Diagnostic performance of WIDAL and Typhi Dot was descriptively assessed against blood culture as the reference standard. The Mann-Whitney U test compared continuous variables. Significance set at $p<0.05$.

RESULTS

A total of 45 culture-confirmed enteric fever cases were included. Mean age was 22.6 with a standard deviation of 14.47 years, reflecting a broad age spectrum. A notable segment of 11 people was under 10 years of age (24.4%), while the majority of 28 people fell within the 10 to 39-year range (62.2%), indicating that enteric fever primarily impacts children and young adults. The gender ratio was nearly equal, with 48.9% of participants identifying as male and 51.1% as female, suggesting an absence of significant gender bias within the sample.

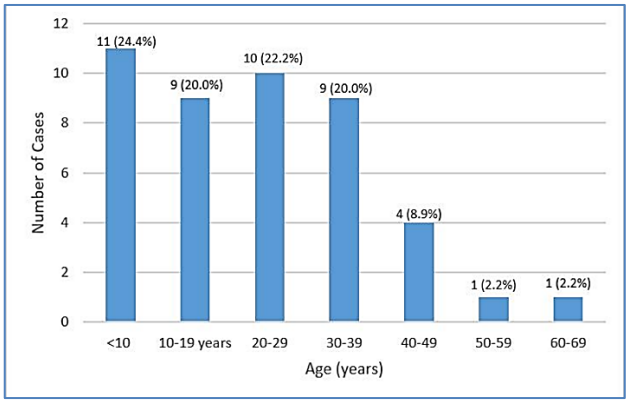

Figure 1: Age distribution of patients with culture-confirmed enteric fever (n=45).

Most cases were reported in the 10-39-year age group (62.2%), followed by children under 10 years (24.4%), indicating a higher burden among adolescents and young adults.

Widal test performance

Of the 45 culture-confirmed enteric fever cases, 20.0% (9/45) were WIDAL positive and 80.0% (36/45) were WIDAL negative. The sensitivity of the WIDAL test was 20.6% (7/34; 95% CI: 8.7–37.9%). The false-negative rate was 79.4% (27/34), indicating that the WIDAL test missed the majority of culture-confirmed Salmonella Typhi cases. WIDAL-negative patients had a significantly higher median age compared to WIDAL-positive patients (24 vs. 14 years; Mann–Whitney U test, p=0.028).

Typhi Dot test performance

Typhi Dot testing was performed in 55.6% (25/45) of patients and not performed in 44.4% (20/45). Among those

tested, 100% (25/25) were negative. This corresponded to a sensitivity and detection rate of 0% (0/25), with a false negative rate and missed diagnosis rate of 100% (25/25) among tested patients.

Species distribution

The predominant causative agent of the cases was *Salmonella typhi* (34 cases), accounting for 75.6%, while 11 cases of *Salmonella Para typhi* represented 24.4%, demonstrating that *S. typhi* is the leading species responsible for typhoid fever in this population.

Antimicrobial susceptibility

The data indicate that most participants exhibited sensitivity to commonly used antibiotics such as amoxicillin, piperacillin, ertapenem, cefixime, ceftazidime, ceftriaxone, and trimethoprim-sulfamethoxazole, suggesting that these medications continue to be largely effective. First- and second-generation cephalosporins (cefalotin, cefoxitin) exhibited 100% resistance. Significant resistance was observed against amikacin and gentamicin, with 44 out of 45 cases found to be resistant (97.8%). Quinolone resistance was high: nalidixic acid 100% resistant; ciprofloxacin 64.4% (29/45) resistant; ofloxacin 64.4% (29/45) resistant. Results indicate limited utility for empirical therapy in this setting. The detection of resistance to fluoroquinolones, including ciprofloxacin and ofloxacin, raises concerns and emphasizes the necessity for ongoing surveillance of resistance patterns.

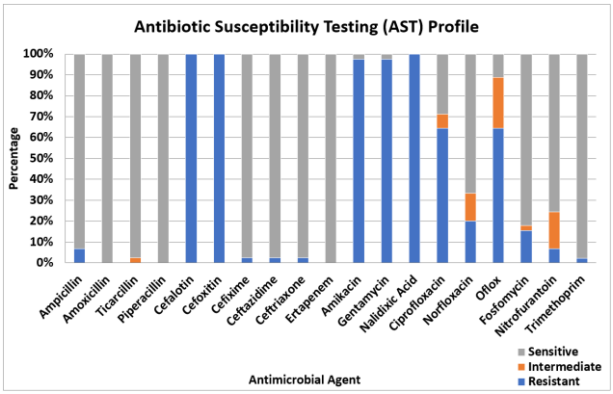

Figure 2: Antimicrobial susceptibility profile of isolates showing high susceptibility to third-generation cephalosporins (97.8%), with a clinically important 2.2% resistance observed. Marked resistance was noted against first- and second-generation cephalosporins, aminoglycosides and quinolones.

The overall low resistance rate of 2% (1/45) to third-generation cephalosporins suggests that these antibiotics remain predominantly effective against the infections studied, but the presence of even limited resistance is clinically concerning, as it implies the possibility of diminishing effectiveness over time if resistance continues to proliferate.[2] Therefore, continuous monitoring of antibiotic resistance trends is essential to ensure the sustained efficacy of these treatment options. Multidrug resistance rate was found to be 2.2% (1/45).

DISCUSSION

At our tertiary care center, the antimicrobial resistance (AMR) profile of enteric fever has shifted markedly over the last decade. This 12-month study quantifies that change while providing regional context. The 2% MDR rate we observed is lower than the 12% pooled estimate reported by the 28-site ICMR-AMR network in 2022 and the 10% MDR prevalence documented by the surveillance for enteric fever in India (SEFI) consortium in 2021.[6] Continuous genomic surveillance is therefore warranted. Third-generation cephalosporins remain highly active (98% susceptible), contrasting with the sharp decline to 87% observed by Aziz and colleagues in Lahore but matching the 96% susceptibility noted across Indian SEFI sites.[7]

Diagnostic accuracy analysis reinforces this stewardship imperative. In our cohort, WIDAL detected only 20% of culture-confirmed cases, comparable to the ~35% pooled sensitivity reported in the Cochrane review.[8] Two studies have similarly reported low sensitivity and specificity of the WIDAL test, aligning with our findings of poor accuracy and low predictive values, confirming WIDAL's limited reliability for typhoid diagnosis.[9,10]

To halt further resistance, we recommend three coordinated actions: First, enforce culture-guided therapy under robust hospital stewardship programs.[11] Second, rapidly expand the WHO prequalified typhoid conjugate vaccination to cut disease incidence and antibiotic exposure.[12] Given the emerging resistance to fluoroquinolones and the first signs of ceftriaxone resistance observed in our study, promoting widespread use of the Typhoid conjugate vaccine (TCV) is essential to reduce typhoid cases, curb antibiotic consumption, and prevent the rise of multidrug-resistant and extensively drug-resistant strains in the region.[13] Third, integrate routine laboratory AMR data with real-time genomic surveillance to flag high-risk clones early. Such a multipronged approach offers the best chance to preserve current antibiotics and curb typhoid morbidity in endemic settings.[14,15] Strengths include uniform laboratory methodology, adult and pediatric representation, and concurrent evaluation of diagnostic and resistance data. Limitations are the single-center, retrospective design and absence of genotyping. This single-center retrospective study with a small sample size (n=45) limits generalizability and statistical power. The absence of molecular typing restricts insight into resistance mechanisms. The absence of Typhi Dot testing in 44% of patients limited comprehensive assessment of its diagnostic performance, rendering sensitivity estimates subject to cautious interpretation.

Limitations

This study has several limitations. First, its retrospective single-center design and relatively small sample size (n=45) may limit the generalizability of the findings to other settings. Second, the absence of molecular characterization and genotyping precluded detailed evaluation of resistance mechanisms and circulating strains. Finally, Typhi Dot testing was not performed in all patients, which limited a comprehensive assessment of its diagnostic accuracy and may have affected estimates of test performance.

CONCLUSION

Our 12-month surveillance shows that 2.2% of *Salmonella* isolates are now multidrug-resistant, while third-generation cephalosporins remain effective in over 95% of cases. However, rising resistance to fluoroquinolones, cefoxitin, and aminoglycosides indicates a narrowing therapeutic window. WIDAL detects only a minority of culture-confirmed cases, and Typhi Dot has limited current relevance, reaffirming blood culture as the diagnostic standard for clinical decision-making and antimicrobial stewardship. To prevent MDR emergence and preserve existing therapies, enhanced typhoid conjugate vaccine deployment, pharmacy-linked stewardship, and multicentric genomic AMR surveillance are urgently needed.

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Ethical approval: The study was approved by the Institutional Ethics Committee

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