

Prevalence and Antimicrobial Resistance of Nontyphoidal Salmonellosis in a Tertiary Care Hospital in South India



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ABSTRACT

Introduction: Nontyphoidal *Salmonella* (NTS) is a significant cause of food- and water-transmitted illness in the world, with the burden amplified in low-socioeconomic countries such as India. While most infections present as self-limiting gastroenteritis, vulnerable populations may develop invasive nontyphoidal salmonellosis (iNTS). This study aimed to determine the prevalence, clinical spectrum, serotype distribution, and antimicrobial resistance patterns of NTS isolates in a tertiary care hospital.

Materials and methods: A total of 61 NTS isolates were recovered from clinical samples, including stool, blood, pus, and urine. Identification was performed using the VITEK-2 system, and serotyping of fifteen isolates was done at the National *Salmonella* and *Escherichia* Center, Central Research Institute (CRI), Kasauli, Himachal Pradesh. Antimicrobial susceptibility testing was performed using the VITEK-2 compact system.

Results: The prevalence of NTS among 1,08,468 samples was 0.056%. Most isolates were from stool samples, followed by blood, pus, and urine. Most patients presented with acute gastroenteritis (56%), often associated with outside food consumption. *S. Enteritidis* and *S. Typhimurium* were the most common serovars isolated. Extraintestinal manifestations included diabetic cellulitis, sepsis, and a rare case of generalized lymphadenopathy in a child, later diagnosed with Mendelian susceptibility to mycobacterial diseases (MSMD), and a case of neonatal meningitis in a 15-day-old infant. All isolates were susceptible to piperacillin-tazobactam, imipenem, and meropenem (100%). However, resistance to ceftriaxone was 22.9%, ciprofloxacin 31.1%, ampicillin 14.7% and trimethoprim-sulfamethoxazole 3.27%.

Conclusion: Nontyphoidal *Salmonella* continues to pose a public health threat in India, particularly with the rise of antimicrobial resistance. These findings underscore the importance of prudent antibiotic usage, robust surveillance systems, and public health interventions focused on food safety, sanitation, and targeted education.

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INTRODUCTION

Nontyphoidal *Salmonella* (NTS) is a significant cause of food- and water-transmitted illness in the world, with the burden amplified in low-socioeconomic countries, such as India. NTS are ubiquitous in nature and commonly associated with spoiled food products of animal origin, especially poultry, eggs, beef, as well as unpasteurized dairy.¹ Among the 2,600 identified *Salmonella* serovars, only a limited group of nontyphoidal types are responsible for the main cause of human infections.² NTS predominantly causes self-limiting gastroenteritis characterized by diarrhea, abdominal cramps, fever, and vomiting. However, in certain populations, particularly infants, the elderly, and immunocompromised individuals, NTS can breach the intestinal mucosa and cause invasive nontyphoidal salmonellosis (iNTS), leading to bacteremia, septicemia, and focal infections with high disease burden. NTS infection's incidence is estimated at over 90 million cases, with approximately 155,000 deaths annually. In Africa and some parts of

Asia, the burden of iNTS disease is alarmingly high, often linked to comorbid conditions such as HIV, malaria, and malnutrition.^{3,4} *S. Enteritidis* and *S. Typhimurium* are the most often isolated serovars associated with human disease.⁵ The increasing trend in antimicrobial resistance among nontyphoidal salmonellae isolates leads to a big challenge in clinical management, as empirical treatment becomes increasingly ineffective against multidrug-resistant (MDR) strains. Resistance to first-line antimicrobials such as chloramphenicol, trimethoprim-sulfamethoxazole, ampicillin, fluoroquinolones, and third-generation cephalosporins has been frequently reported, often related to the spread of resistance genes via plasmids and mobile genetic elements.⁶ The development of AMR in NTS is largely attributed to indiscriminate antibiotic use in human medicine, agriculture, and animal husbandry. In many regions, antibiotics are used not only for treatment but also as growth promoters in livestock, increasing the emergence of resistant strains,

which may enter the food chain and pose a risk to humans.⁷ Given the emerging threat of antimicrobial resistance, continuous surveillance of NTS prevalence and its resistance patterns is vital. This study aims to determine the prevalence of NTS infections in a tertiary care hospital in Madurai district and to evaluate the antimicrobial resistance patterns of the isolated strains.

MATERIALS AND METHODS

Study Design and Setting

This study was conducted in the Department of Microbiology, Velammal Medical College Hospital and Research Institute, Madurai, Tamil Nadu. This retrospective study was conducted from January 2019 to January 2025. The objective was to determine the prevalence and antimicrobial resistance patterns of NTS isolates obtained from clinical samples during the study period.

Sample Collection and Isolation

The study included 61 NTS isolates. These isolates were obtained from various clinical specimens received in the microbiology laboratory for routine culture and sensitivity testing. The medical records of the corresponding patients were reviewed to gather information regarding age, sex, clinical presentation, and any underlying or predisposing illnesses such as immunosuppression, diabetes mellitus, or recent hospitalization.

Culture and Identification

All clinical specimens were processed using standard microbiological procedures.

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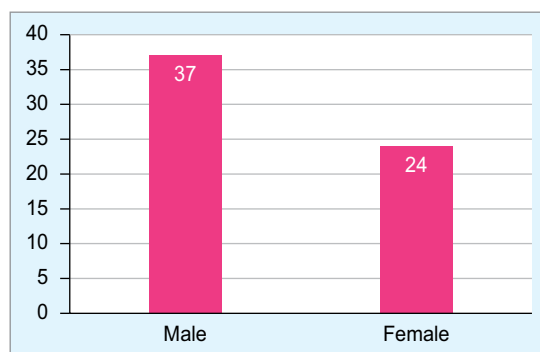


Fig. 1: Gender distribution (n = 61)

Samples were inoculated on the following culture media: 5% sheep blood agar, cystine lactose electrolyte deficient (CLED) agar (for urine), MacConkey agar, and deoxycholate citrate agar (DCA) (HiMedia, India). Plates were incubated at 37°C for 18–24 hours under aerobic conditions. Colonies suspected to be *Salmonella* were subjected to automated identification using the VITEK-2 compact system (bioMérieux, France). 61 isolates were identified as *Salmonella enterica* belonging to nontyphoidal serovars.

Serotyping

The serovar identification and antigenic profiling of 15 isolates were done at the National *Salmonella* and *Escherichia* Center, Central Research Institute (CRI), Kasauli, Himachal Pradesh, using standard slide agglutination with specific antisera based on the Kauffmann–White classification scheme. The serotype of 46 isolates could not be serotyped due to logistical limitations.

Antimicrobial Susceptibility Testing

Antimicrobial susceptibility testing (AST) of all 61 NTS isolates was performed using the VITEK-2 compact system according to the Clinical and Laboratory Standards Institute (CLSI) M100 guidelines. The following classes of antibiotics were tested: β -lactams (ampicillin, ceftriaxone, cefepime, imipenem, meropenem, piperacillin/tazobactam, cefoperazone/sulbactam, fluoroquinolones (ciprofloxacin), and sulfonamides (trimethoprim-sulfamethoxazole). Isolates were classified as susceptible, intermediate, or resistant based on minimum inhibitory concentration (MIC) breakpoints defined by CLSI M100 (2024) standards.

Data Analysis

Descriptive analysis was used to summarize the distribution of isolates by specimen type, serovars, and resistance patterns by using tables and charts. Statistical analysis was done using Microsoft Excel.

RESULTS

Among 1,08,468 samples received, a total of 61 NTS isolates were identified. Patient's median age was 48 years, with an age range of 15 days to 80 years. Of the 61 patients, 37 were male (including two children) and 24 were female (including one child), indicating males accounted for a higher proportion of cases compared to females (Fig. 1).

The majority of isolates (56%) were obtained from stool samples, followed by blood, pus, and urine. The most common clinical manifestation among the patients was diarrhea. A total of 34 patients presented with acute gastroenteritis and were managed with intravenous antibiotic therapy. Of these, 16 patients had a documented history of consuming food from outside sources, suggesting a potential foodborne origin. None of the patients exhibited features of enteric fever, and Widal tests were nonreactive in all cases, supporting the diagnosis of nontyphoidal salmonellosis. In addition, two patients presented with diabetic cellulitis, associated with soft tissue infection, which was treated with surgical debridement in conjunction with intravenous antibiotics. Nine patients developed sepsis, with systemic symptoms and positive blood cultures for *Salmonella*. All cases in the study showed complete clinical recovery within 2 weeks of initiating appropriate treatment, and no mortality was reported.

A notable case involved a 4-year-old male child with a prior history of pulmonary tuberculosis, who presented with generalized lymphadenopathy. Culture of a lymph node biopsy yielded *Salmonella* Typhimurium. Given the unusual site of infection and his previous history, further immunological workup was performed, which revealed a diagnosis of Mendelian susceptibility to mycobacterial diseases (MSMD), an uncommon genetic condition involving T-cell dysfunction. The child was treated with intravenous ceftriaxone and was subsequently planned for bone

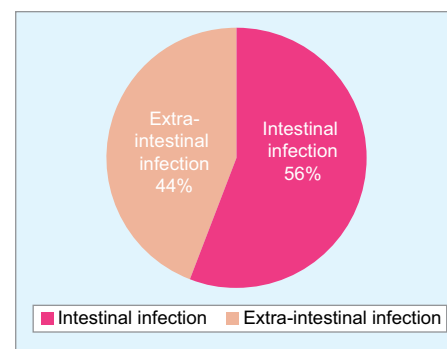


Fig. 2: Distribution of intestinal and extraintestinal infection (n = 61)

marrow transplantation as part of definitive therapy. A rare case of a 15-day-old neonate, delivered via lower-segment cesarean section (LSCS) due to breech presentation, presented with signs of meningitis. Blood and cerebrospinal fluid (CSF) cultures were positive for NTS, confirming the diagnosis of NTS meningitis. Investigations to identify the source of infection revealed negative cultures for NTS from the mother's breast milk and stool, suggesting that vertical transmission was unlikely. Given the mode of delivery and absence of maternal colonization, a postnatal or nosocomial source of infection is strongly suspected (Fig. 2).

Fifteen isolates were successfully serotyped at the National *Salmonella* and *Escherichia* Center. Among these, *S. Typhimurium* was identified in five cases, with the antigenic formula 4,12:1:1. *S. Enteritidis* was identified in nine cases with antigenic formula 9,12:gm. *S. Kentucky* was isolated in one case with antigenic formula 8:i:z6 (Fig. 3).

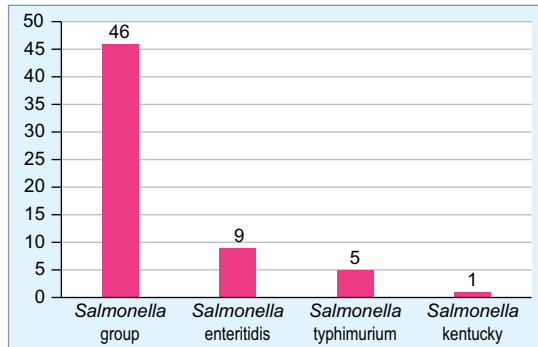
High susceptibility rates were observed with piperacillin/tazobactam, imipenem, and meropenem, with 100% of isolates being susceptible. Cefoperazone/sulbactam also demonstrated excellent activity, with 98.3% susceptibility and only one isolate (1.6%) showing intermediate resistance. Among the cephalosporins, cefepime and ceftriaxone showed susceptibility rates of 86.8% and 77%, respectively. Ampicillin showed reduced efficacy, with 85.2% of isolates susceptible and 14.7% resistant. Resistance was notably higher with ciprofloxacin, where only 60.6% of isolates were susceptible, 8.1% intermediate, and 31.1% resistant. Trimethoprim/sulfamethoxazole remained effective in 95% of isolates, with minimal resistance (3.27%) (Table 1).

DISCUSSION

In this study, the prevalence of NTS is found to be 0.056%, which is significantly lower

Table 1: Antibiotic susceptibility rates of NTS

Antibiotics	Susceptible	Intermediate	Resistant
Ampicillin	52 (85.2%)	0	9 (14.7%)
Piperacillin/tazobactam	61 (100%)	0	0
Ceftriaxone	47 (77%)	0	14 (22.9%)
Cefoperazone/sulbactam	60 (98.3%)	1 (1.63%)	0
Cefepime	53 (86.8%)	0	8 (13.1%)
Imipenem	61 (100%)	0	0
Meropenem	61 (100%)	0	0
Ciprofloxacin	37 (60.6%)	5 (8.1%)	19 (31.1%)
Trimethoprim/sulfamethoxazole	58 (95%)	1 (1.6%)	2 (3.27%)

**Fig. 3:** Distribution of nontyphoidal *Salmonella* serovars (n = 61)

compared to the pooled proportion estimate for noninvasive NTS (2.1%) and for invasive NTS (0.3%).³ The majority of NTS isolates in our study were recovered from stool specimens, with 56% of isolates obtained from patients presenting with acute gastroenteritis. This finding aligns with global epidemiological patterns, where NTS is predominantly associated with foodborne diarrheal illness.⁸ Approximately, 14.7% of isolates were recovered from blood cultures, indicating a potential for invasive NTS infections, which is lower compared to the study conducted in Africa by Feasey et al.⁹ Extraintestinal manifestations were observed, with NTS isolated from pus samples in diabetic foot infections and wound infections, as well as from a urine specimen in a patient with sepsis. These findings further emphasize the diverse clinical presentations of NTS beyond gastrointestinal illness, particularly in patients with comorbidities such as diabetes.¹⁰ The serotyping results revealed *S. Typhimurium*, *S. Enteritidis*, and *S. Kentucky*, which is consistent with other studies from India and globally, where *S. Typhimurium* and *S. Enteritidis* are recognized as the most common NTS serovars implicated in human disease.^{11–14} In contrast to our study, where *Salmonella enterica* serovar Kentucky was isolated from only one case of wound infection, a hospital-based study reported *S. Kentucky* as the most prevalent NTS serovar, accounting for 48% of isolates. That study also highlighted significant antimicrobial resistance in

S. Kentucky, showing resistance to ampicillin, nalidixic acid, and ciprofloxacin. In the present study, *S. Kentucky* isolate was resistant to cephalosporins and ciprofloxacin. Although we did not encounter a high prevalence of *S. Kentucky* or detailed resistance patterns in our limited sample, its emerging role as a resistant NTS serovar emphasizes the need for continuous surveillance.¹⁵ A study conducted in India showed that NTS isolates were sensitive to all antibiotics except ciprofloxacin and nalidixic acid.¹⁶ In the present study, 22.9% of isolates were resistant to ceftriaxone, which is significantly higher than a study conducted in South India, where 5% resistance to ceftriaxone was reported.¹⁷ 31.1% of the isolates are resistant to ciprofloxacin, which creates limited treatment options in invasive NTS.

LIMITATIONS

The study is limited by its small sample size. Furthermore, serotyping could not be performed for all isolates, which restricts a complete understanding of circulating serovars in the region. Molecular characterization and resistance gene profiling were also not undertaken, which could have provided additional insights into the resistance mechanisms.

CONCLUSION

In India, NTS infections pose a significant public health threat, primarily transmitted

via the fecal-oral route. This often results from the consumption of contaminated food, polluted water, and fertilizers. Poor sanitation practices further worsen the situation. With the growing concern of antimicrobial resistance, it is crucial to handle food and water safely and improve public health infrastructure. Effective prevention strategies should include better sanitation, public awareness, strong surveillance systems, and an effective infection control policy.

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