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COMPARISON OF WIDAL TEST AND BLOOD CULTURE FOR DIAGNOSIS OF TYPHOID FEVER: CORRELATION WITH ANTIBIOTIC SUSCEPTIBILITY PATTERNS OF *SALMONELLA TYPHI*

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ABSTRACT

Typhoid fever is a systemic infection caused by *Salmonella enterica* serovar Typhi, a Gram-negative bacillus predominantly transmitted through contaminated food and water. It remains a significant public health concern in low- and middle-income countries due to poor sanitation and limited access to reliable diagnostics. Early and accurate diagnosis, along with effective antibiotic treatment, is crucial for controlling morbidity and mortality. This study aimed to evaluate the diagnostic accuracy of the Widal agglutination test in comparison with blood culture for the diagnosis of typhoid fever, and to assess the antibiotic susceptibility patterns of *Salmonella* isolates to guide effective treatment strategies. A total of 150 patients with clinically suspected typhoid fever were enrolled from various healthcare facilities in Abbottabad, Pakistan. Patients presented with common symptoms including prolonged fever, abdominal pain, rash, headache, weakness, loss of appetite, and constipation. Widal test was performed using standard agglutination methods, with titers 1:80 (anti-TO) and ≥1:160 (anti-TH) considered positive. Blood cultures were carried out to isolate *Salmonella Typhi* and *Paratyphi*. Antibiotic susceptibility testing of isolates was performed using the Kirby-Bauer disk diffusion method. Out of 150 samples, 90% tested positive by Widal test, while only 42% yielded positive results in blood cultures. Antibiotic susceptibility profiles revealed resistance to Azithromycin (33%), Ciprofloxacin (47%), and Moxifloxacin (54%). In contrast, high sensitivity was observed to Meropenem (98%) and Imipenem (96%). The study concluded that widal test showed limited sensitivity and specificity, with a high rate of false positives compared to blood culture, which remains the gold standard for typhoid diagnosis. The study also highlights levels of resistance to commonly used antibiotics, underlining the importance of culture-based

diagnosis and antibiotic stewardship in typhoid fever management.

1. INTRODUCTION

Salmonella enterica subspecies. *Enterica serotype. typhi*, a Gram-negative bacillus, causes typhoid disease. It is a leading source of disease and mortality globally, with an estimated 16.6 million new infections each year. If not treated, typhoid illness can cause cancer. The condition is peculiar to humans and is characterized by malaise, persistent fever, anorexia, nausea, exhaustion, headache, nausea, stomach pain, constipation or diarrhea, nonspecific abdominal discomfort, transitory rash, dry cough, and myalgia. These are followed by a coated tongue, sensitive belly, splenomegaly, hepatomegaly, bradycardia, and leukopenia. The most common consequences are intestinal bleeding and perforation. Infection affects people of all ages, with children having a greater frequency and more varied clinical presentation. Relapse and re-infection are common in typhoid, occurring in less than 10% of cases. Molecular typing is the sole way to distinguish between re-infection and relapse (Jahan *et al.*, 2021).

Typhoid fever is caused by the human-restricted *Salmonella enterica serovars typhi* and *Paratyphi* A, B, and C (*S. typhi* and *S. Paratyphi* A, B, and C). Different *Salmonella serovars*, including *S. typhi* and *S.paratyphi*, are characterized by a distinct set of their surface antigens: lipopolysaccharide O (somatic), flagellar H, and virulence-capsule antigens. Based on their host-specificity and disease outcomes, S. enterica are grouped into typhoidal and non-typhoidal *Salmonella* serovars (NTS). The majority of NTS serovars represented by *S. typhimurium* and *S. enteritidis* can infect humans and animals and cause a self-limiting gastrointestinal *Salmonella* in human body, with some exceptions of NTS causing invasive disease (Neupane *et al*., 2021).

The most often used serological test is Widal, which was established by Georges Fernand Widal in 1890 based on the development of agglutinating antibodies against the lipopolysaccharide (LPS; O) and flagella (H) antigens of *Salmonella typhi* and *paratyphi* A and B. This test has become outdated in many affluent nations due to its mediocre findings, low occurrence of typhoid fever, and the availability of more advanced diagnostic methods. The variable sensitivity and specificity of the widal test have been observed in several research, and its function as a diagnostic tool remains controversial. However, certain research done in Tanzania, Vietnam, Bangladesh, and India over time concluded that the widal test might be useful as a diagnostic tool as an alternative to blood culture. Other studies done in Pakistan, Nepal, South Africa, Tanzania, and Ethiopia found that the widal test alone may not be useful for diagnosing typhoid fever since it can yield false positive results. However, the situation is completely different in developing nations like Bangladesh, where widal tests are still commonly utilized since the ability to culture bacteria is limited to only tertiary care clinics, there is a shortage of microbiologist, and the cost of culture is prohibitively high when compared to serological tests (Akter *et al.*, 2020).

Widal tests are employed in endemic regions, although they have limited sensitivity and specificity. Widal is an agglutination test that is used to confirm the clinical diagnosis during the second week of illness. Furthermore, due to the Widal test's limited specificity, a fourfold increase in antibody titter within a one-week period for confirmation of typhoid fever is expected. As a result, relying just on a Widal

test for diagnosis is less reliable. However, it was advised to consider a greater antibody titer in a single test for a precise diagnosis of typhoid among infected individuals (Shahapur *et al*., 2021).

Culture of *S. typhi or S. Paratyphi* from sterile sites provides a definitive diagnosis of typhoid fever. In addition, culture permits antimicrobial susceptibility testing and molecular typing that guides treatment and informs public health and infection control activities. Blood is the preferred specimen type though stool or urine samples may yield positive results is about 30% of patients in the second week of illness. Theoretically culture bone marrow method is still now considered the gold standard as the sensitivity is higher, up to 90% and less affected by prior antimicrobial therapy, but bone marrow is not a practical sample to collect in virtually any clinical setting (Bamford, 2022).

Widal tests, however, are comparatively affordable, especially when compared to bacterial culture procedures, and are hence still commonly utilized. Though blood culture method had been used as gold standard method for accurate diagnosis of typhoid fever, it has limitation of time requirement, at least 3 days and positive results of only 30-70% even in well-equipped laboratory. Thus, a more quick, easy, and cost-effective diagnostic tool would be extremely beneficial, particularly in underdeveloped nations. To compare sensitivity and specificity of widal test in culture positive samples suspected for typhoid fever along with antibiotic resistance trend of isolated and determine multi drug resistant isolates (Chaudhary *et al.*, 2016).

The gold standard technique for diagnosing typhoid fever is to culture blood and bone marrow. Urine and feces cultures may also reveal growth in patients who acquired carriers either through clinical infection or contact with infected individuals. Limited facilities, labor, time, money, and resources impede culture-based diagnosis. As a result, different examinations such as immunochromatographic tests (ICTs) and Widal testing are utilized. Other technique include latex agglutination, co-agglutination, and the polymerase chain reaction (PCR) (Shahapur *et al*., 2021).

Antibiotic therapy of typhoid fever is impeded by growing multi-drug resistance bacteria that are less susceptible to the first-line medications chloramphenicol, trimethoprim, ampicillin, and, more recently, fluoroquinolones. Typhoid fever is believed to be fatal in up to 30% of cases if not treated with suitable antibiotics. Early diagnosis of typhoid fever is critical for recovery (Sauteur *et al.*, 2020).The worldwide spread of multidrug-resistant (MDR) Salmonella, as well as the introduction of extensively drug-resistant (XDR) Salmonella, highlight the need for improved diagnostic tests and innovative treatment regimens that offer alternatives to existing medications. Antibiotics are the major treatment choices for typhoid fever; however, Salmonella is constantly evolving to acquire plasmid, prophage, transposon, or chromosomal gene changes that enable drug resistance. A variety of publications have revealed the global spread of *S. typhi* and *S. paratyphi* strains that are resistant to all of the first-line antibiotics, ampicillin, chloramphenicol, and co-trimoxazole, together termed as multidrug resistance (MDR) salmonella (Crump *et a*l., 2015).

Multidrug-resistant (MDR) typhoid strains have been endemic in Pakistan for the last two decades. In November 2016, an epidemic of extensively drug-resistant (XDR) typhoid cases was detected in Pakistan's Sindh Province. These were caused by MDR S. Typhi, which had developed resistant to third-generation cephalosporins and ciprofloxacin but were usually still sensitive to azithromycin and carbapenems (Nizamuddin *et al*., 2021). Azithromycin and carbapenems are "last resort" medications for treating Salmonella infection, however azithromycin-resistant *S. typhi* strains and carbapenem-resistant invasive NTS have also been identified. These findings highlight the critical need for new diagnostic, preventive,

and treatment techniques to better manage drug-resistant *S. typhi* and *S. paratyphi*. (Neupane et al., 2021).

When a blood culture facility is unavailable, it is difficult to diagnose enteric fever accurately and early. As a result, the Widal test is still commonly employed in resource-limited environments.

Recently, fast immunochromatographic assays (ICT) have been devised for the rapid detection of enteric fever. We compared the sensitivity and specificity of an immunochromatography-based *Salmonella typhi* IgM/IgG test kit and Widal test to blood culture for the diagnosis of enteric fever (Akter *et al*., 2020).Therefore the present research aimed to evaluate the diagnostic efficacy of the Widal test in comparison to blood culture for the accurate diagnosis of typhoid fever and to identify effective antibiotic treatments against the infection. The study focused on assessing the sensitivity and specificity of the Widal test relative to blood culture, isolating *Salmonella typhi* from patients clinically suspected of having typhoid fever and evaluating the antibiotic resistance patterns of the isolates to determine the most effective therapeutic options.

2. MATERIALS AND METHODS

2.1 Study area and Collection of Sample

A total of 150 blood samples were collected from suspected typhoid patients across hospitals in District Abbottabad, Khyber Pakhtunkhwa (KPK). Following standard procedures, 3–5 ml of blood was drawn into EDTA bottles, labeled, and stored under refrigeration until processing. Widal tests were performed in the university laboratory, and only samples testing positive were further processed for blood culture. Negative Widal samples were excluded from further analysis.

2.2 Widal Test

The Widal test was performed using the slide agglutination method to detect O and H antigens in the blood serum. A drop of serum was mixed with equal drops of O and H antigen suspensions on a paper card, alongside positive and negative controls (Eleazar, 2020). The mixtures were gently stirred using separate applicator sticks and observed under adequate lighting after two minutes for visible agglutination, indicating a positive reaction. According to the manufacturer's guidelines, a titer of ≥1:80 for anti-TO and ≥1:160 for anti-TH antibodies was considered positive for recent typhoid infection (Mawazo *et al*., 2019).

2.3 Turbidity of Bacteria

Turbidity is the cloudiness or haziness of fluid caused by suspended insoluble particles. After widal test positive, the blood samples were then transfer to broth and kept in shaking incubator for 3-7 days then their turbidity appearance, once turbidity appears, the sample were culture initially on TSA and then on XLD and SS agar for confirm nature of strains.

2.4 Isolation and culturing

Bacterial growth was conducted in selective media such as Tryptic Soy Agar (TSA) and MacConkey agar. Clinical samples were streaked onto XLD or SS agar and incubated at 37°C for 24 to 48 hours. After 24 hours of incubation, plates were examined for characteristic growth of *Salmonella typhi*.

2.5 Bacterial Identification

Plates were examined for typical *Salmonella typhi* colony morphology and color following incubation. Further identification of the isolates was performed through Gram staining and a series of biochemical tests. Gram staining revealed the morphological and structural characteristics of the bacteria, distinguishing Gram-negative (pink) from Gram-positive (purple) cells under 100x oil immersion microscopy. For biochemical characterization, tests including oxidase, catalase, indole, and methyl red were conducted to confirm the identity of *Salmonella* strains. The oxidase test detected cytochrome oxidase activity, indicated

by a purple color change on filter paper. The catalase test confirmed the presence of catalase enzymes through visible bubble formation upon the addition of hydrogen peroxide. The indole test identified bacteria capable of degrading tryptophan, producing a pink ring after adding Kovac's reagent. The methyl red test assessed acid production from glucose fermentation, with a red color indicating a positive result. These tests collectively aided in confirming the identity and biochemical behavior of *Salmonella typhi* isolates.

2.6 Antibiotic Sensitivity testing

Antibiotic susceptibility testing (AST) was performed using the standard Kirby-Bauer disc diffusion method to determine the effectiveness of various antibiotics against *Salmonella typhi* isolates. Sterile cotton swabs were dipped into well-mixed saline bacterial suspensions, and excess liquid was removed by pressing against the inner wall of the tube. Mueller-Hinton agar plates were uniformly inoculated and left to dry for approximately five minutes before placing antibiotic discs on the surface. The discs were gently pressed to ensure contact with the agar, and plates were incubated in an inverted position at 37°C for 24 hours. Each isolate was tested for susceptibility to five different commercially available antibiotics (Rahman et al., 2019).

3. RESULTS

3.1 Sample collection

Samples were collected from various clinical setups in district Abbottabad, Hazara division Khyber Pakhtunkhwa (KPK) during August 2024 to September 2024. Total 150 Samples were collected from patients who have signs and symptoms of Salmonella typhi infection. Standard Operating Procedures (SOPs) were followed. Total 150 samples were collected, out of which only 63 samples show positive culture results, and 135 samples shows positive widal test (Table 1).

Table 1. Comparison positive culture and widal positive test

Total Samples	Widal positive	Culture positive
150	**135**	**63**
100%	**90%**	**42%**

3.2. Widal test

The widal test is a laboratory test to determine the antibodies that the body makes against the *salmonella* bacteria that causes typhoid fever. It looks for O and H antibodies in a patient's sample blood (serum). So that the collected samples of patients total 150 out of 87 samples were then used for further process of isolation of bacteria and identification and also check susceptibility of antibiotics.

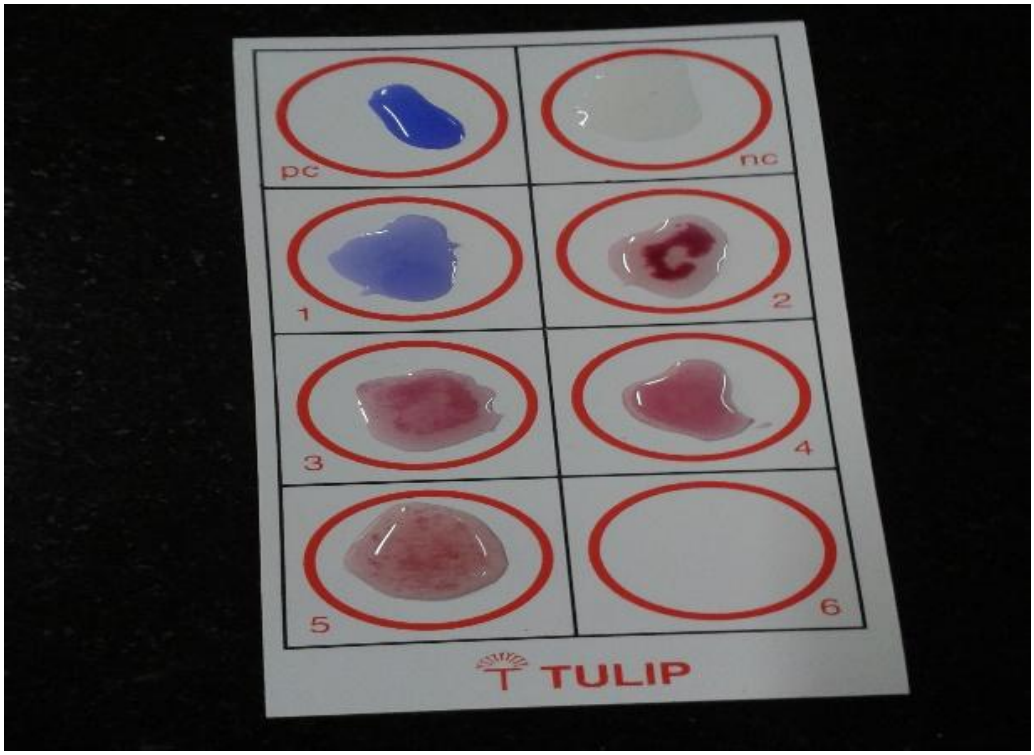

Figure 1. showing widal test agglutination titer

3.3. Sample Processing

The collected samples were streaked initially on the TSA. After 24hr incubation at 37^0c plate having the colonies were observed. Colonies on TSA were small, smooth, circular convex and milky, and on XLD media colonies were a black center and a slightly red colored translucent zone, while in SS agar media colonies were smooth and opaque or colorless.

3.4. Turbidity Checkups

Turbidity of bacteria growing in a liquid broth medium causes the culture to appear turbid. This is because a bacterial culture works as a colloidal suspension, blocking and reflecting light that passes through the culture broth (Figure 2).

Figure 2. Represented turbidity of bacteria broth culture

3.5. Morphology of *Salmonella typhi*

Salmonella typhi is gram-negative, non-motile rod shape that is 0.7–1.5 µm 2.0–5.0, *Salmonella typhi* colonies on TSA media are typically white, smooth, round, and 2–4 mm in diameter. It can ferment glucose but not lactose, and its oxidase negative, catalase positive, methyl red positive indole negative microorganism. On Blood agar these colonies are non-hemolytic, grey or whitish, and are about 2–3 mm in diameter were observed (figure 3).

Figure 3. Samples of growth colonies isolated from TSB media during study.

After 24 hours of incubation, salmonella typhi colonies on blood agar are grey/white, non-hemolytic, and non-swarming, measuring 2 to 3 mm in diameter. (figure 4).

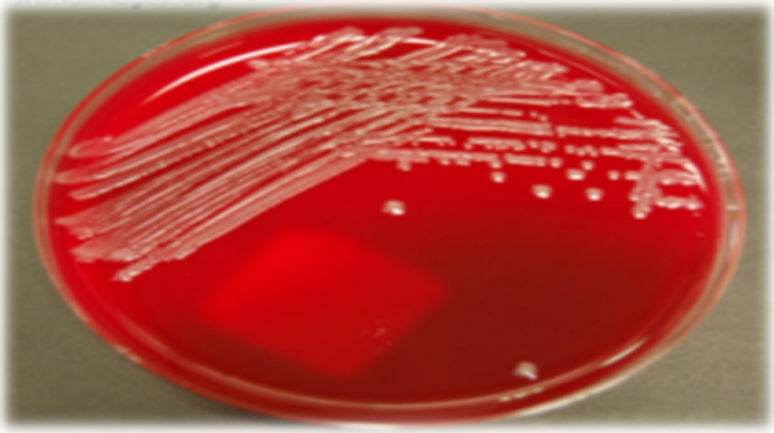

Figure 4. Representing *Salmonella typhi* colonies on blood agar media.

3.6 Bacterial identification

Biochemical tests were conducted to confirm the identity of *Salmonella typhi* isolates based on characteristic metabolic activities. The result show that the isolates were Gram-negative rods, catalase-positive, oxidase-negative, indole-negative, methyl red-positive, and urease-negative. This pattern is consistent with the standard biochemical profile of *Salmonella typhi*, supporting its identification in the tested clinical samples (Table 2).

Table 2. Tests for identification of *Salmonella typhi*

Bacteria	Gram stain	Morphology	Urease test	Methyl red test	Indol test	Catalase test	Oxidase test
Salmonella typhi	Negative	Rods	Negative	Positive	Negative	Positive	Negative

3.7. Antimicrobial susceptibility testing

After the identification of *Salmonella typhi* disc diffusion test were performed on MHA (Mueller–Hinton agar) media (Figure 5). To determine the antibiotic sensitivity pattern of isolated strains, sensitivity testing was performed.

Table 3. Antibiotics use for susceptibility test during study

Antibiotics	Microgram (µg)
Azithromycin	**15µg**
Meropenem	**10µg**
Ciprofloxacin	**10µg**
Moxifloxacin	**15µg**
Impienem	**10µg**

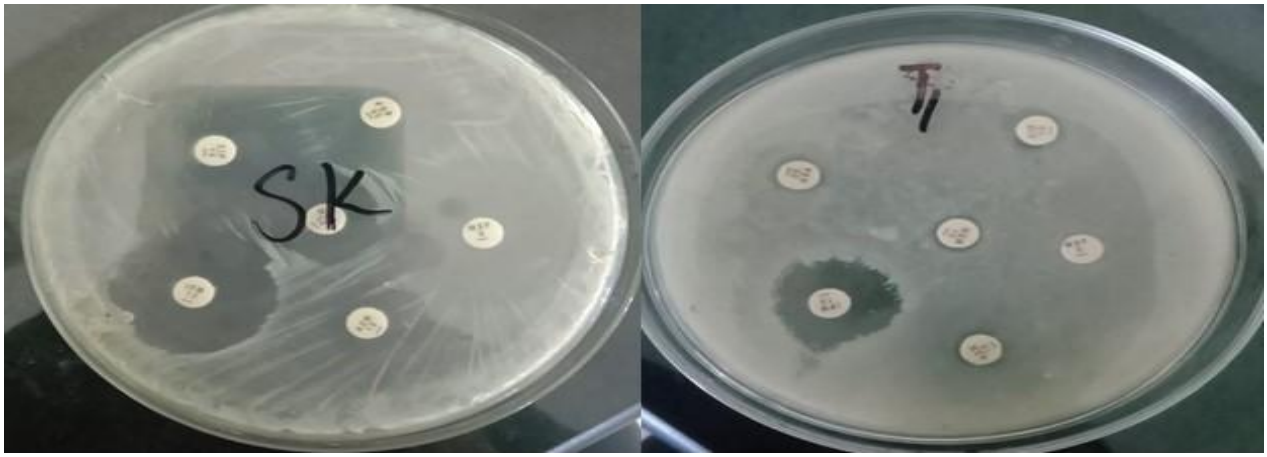

Figure 5. Kirby disc diffusion test results interpretation

During this study the Azithromycin shows 66.67% sensitive while the Moxifloxacin and Ciprofloxacin show 42.56% and 52.38% sensitive respectively, but Meropenem and Imipenem shows high sensitivity rate 95.24% and 90.44% this shows that these are highly effect drugs' in typhoid fever infection. In case of resistance the Moxifloxacin shows high resistance to Salmonella infection while other drugs such as Azithromycin, Ciprofloxacin, Meropenem and Imipenem shows that 33.33%,47.62%,4.76%,9.56 resistance respectively as shown in table 4.

Table: 4. Showing Resistance and sensitivity Pattern of antibiotics

Antibiotics	Resistance %	Sensitivity %
Azithromycin 15 µg	**33.33%**	**66.67%**
Meropenem 10 µg	**4.76%**	**95.24%**

Ciprofloxacin 10 µg	**47.62%**	**52.38%**
Moxifloxacin 15 µg	**57.14 %**	**42.86%**
Imipenem 10 µg	**9.56%**	**90.44%**

4. DISCUSSION

This study was based on the comparison diagnosis of Salmonella typhi between widal test and blood culture. Present study also revealed to see the antibiotics resistance and sensitivity pattern of current study isolates bacteria. So Typhoid fever is a systemic disease caused by Salmonella Enterica serotype typhi. It is a major source of morbidity and mortality globally. The blood samples are collected from Ayub teaching hospital who have sign and symptoms of typhoid fever or have past typhoid infection. Total 50 samples are collected for this study in which 45 samples are widal positive, and out of 45 samples only 21 samples are culture positive and isolates the bacteria. All samples were collected from hospital indoors and outdoors patients. TSA was used for enumeration of the pathogenic microbe in the sample collection. Gram's staining revealed some gram negative, rod's shape. After that biochemical tests were conducted for selected samples. Salmonella species was catalase and methyl red positive. It was found to be non-motile organism. Oxidase and indole were negative for the strain under study. For the conformation of resistance drugs to bacteria antibiotic susceptibility done. The usual laboratory measures employed, notably the widal test and blood culture, allow for a thorough comparison while considering recognized diagnostic methods. Aside from being the most often utilized, widal testing has been criticized for its low sensitivity and specificity. While regarded as a gold standard, blood culture is hampered by lengthier turnaround times and probable false negatives, especially in patients receiving previous antibiotic treatment.

This study among 540 samples analyzed, 114 were subsequently confirmed based on positive blood culture for *S. typhi*. 327 cases were positive as having significant titer of widal agglutination test. 99 cases had neither blood culture positive result nor significant titer on widal agglutination test but still were included due to clinically in favor of typhoid fever, it was observed that, in comparison between blood culture technique and widal agglutination test, widal agglutination test were less time consuming but gives more false positive reaction. In our study false positivity rate of Widal agglutination test was 213 (65.1%), which is very high. False positive results of Widal titer were so high in this study (Sangani and Toshniwal, 2018).

Another research of 160 samples, 106 Salmonella species, and revealed salmonella features after undergoing biochemical testing and an antibiotic sensitivity test using commercial antibiotics. The isolated Salmonella species demonstrated greater susceptibility to the medications Ciprofloxacin (100%), Ceftriaxone (91%), and Gentamycin (58%) (Bale Zone, 2020).

Sensitivity testing was done to ascertain the isolated isolates' pattern of antibiotic sensitivity. The antibiotics azithromycin, ciprofloxacin, meropenem, and moxifloxacin had the highest antimicrobial sensitivity. As like this study, two XDR cases of *S. typhi* were identified; both were sensitive to imipenem, a carbapenem antibiotic. One of these cases was tested for and found to be sensitive to azithromycin as well (Umair and Siddiqui, 2020).

Research on XDR S. typhi in Northern Pakistan found antibiotic sensitivity to just azithromycin and meropenem. Yousafzai et al. analyzed the

epidemic of ceftriaxone-resistant S. typhi in Hyderabad between 2016 and 2017 and identified a similar antibiotic sensitivity profile, that is, azithromycin, imipenem, (Yousafzai et al., 2019).

5. CONCLUSION

The findings of this study highlight the limitations of relying solely on the Widal test for diagnosing typhoid fever, as it showed a much higher positivity rate compared to blood culture, which is considered the gold standard. While the Widal test is easy and quick, its low specificity can lead to overdiagnosis. Blood culture, although more resource-intensive, provides a more accurate confirmation of Salmonella infections.

Furthermore, the antibiotic susceptibility results raise important concerns. Resistance to commonly prescribed antibiotics such as Azithromycin, Ciprofloxacin, and Moxifloxacin was notably high, which could compromise effective treatment in many patients. On the other hand, Meropenem and Imipenem showed strong effectiveness, suggesting they may still be reliable options for treating resistant cases.

Overall, this study emphasizes the need for improved diagnostic tools and more responsible use of antibiotics to manage typhoid fever, especially in regions like Abbottabad where the disease remains a significant public health challenge.

REFERENCES

AKTER, F., YEASMIN, M., ALAM, M. Z., HASAN, M. R., RAHMAN, F., KHANDKER, E., HOQUE, M. M., BARAI, L., MOHIUDDIN, M. & JILANI, M. S. A. 2020. Comparative evaluation of rapid Salmonella Typhi IgM/IgG and Widal test for the diagnosis of enteric fever. *IMC Journal of Medical Science,* 14**,** 18-25.

BALE ZONE, S. E. E. 2020. Prevalence and Antimicrobial Susceptibility Pattern of Salmonella Species Isolated from Human Blood Samples in Robe Hospital. *Prevalence,* 63.

BAMFORD, C. 2022. The laboratory diagnosis of typhoid fever. *Journal of the African Society for Paediatric Infectious Diseases,* 1**,** 1-9.

CHAUDHARY, P., SHARMA, V., CHAUDHARY, A., CHATURWEDI, S. & SHRESTHA, A. 2016. Comparative Study of Blood Culture and Widal Agglutination Test from the Patients Suspected of Enteric Fever. *British Microbiology Research Journal,* 16**,** 1-9.

CRUMP, J. A., SJÖLUND-KARLSSON, M., GORDON, M. A. & PARRY, C. M. 2015. Epidemiology, clinical presentation, laboratory diagnosis, antimicrobial resistance, and antimicrobial management of invasive Salmonella infections. *Clinical microbiology reviews,* 28**,** 901-937.

ELEAZAR, C. I. 2020. Comparison of Widal Screening Test with Cultural Isolation Method for Diagnosis of Salmonella typhi/paratyphi Infections. *Recent Progress in Microbiology and Biotechnology***,** 88.

JAHAN, N., KHATOON, R., MISHRA, P., MEHROTRA, S. & AHMAD, S. 2021. A comparative evaluation of rapid card and Widal Slide agglutination tests for rapid diagnosis of typhoid fever. *Medical Journal of Dr. DY Patil University,* 14**,** 409-414.

LÓPEZ-SEGURA, N., CORBERÓ-RIVALI, C., MALDONADO-FERNÁNDEZ, M. C., CALPE-FRAILE, S., PEYRA-ROS, J. & MARTÍNEZ-ROIG, A. 2019. Imported extensively drug resistant typhoid fever in a child travelling to Spain from Pakistan. *Journal of travel medicine,* 26**,** taz066.

MAWAZO, A., BWIRE, G. M. & MATEE, M. I. 2019. Performance of Widal test and stool culture in the diagnosis of typhoid fever among suspected patients in Dar es Salaam, Tanzania. *BMC research notes,* 12**,** 1-5.

NEUPANE, D. P., DULAL, H. P. & SONG, J. 2021. Enteric fever diagnosis: current challenges and future directions. *Pathogens,* 10**,** 410.

NIZAMUDDIN, S., CHING, C., KAMAL, R., ZAMAN, M. H. & SULTAN, F. 2021. Continued outbreak of ceftriaxone-resistant Salmonella enterica serotype Typhi across Pakistan and assessment of knowledge and practices among healthcare workers. *The*

American journal of tropical medicine and hygiene, 104**,** 1265.
RAHMAN, M. A., AHMAD, T., MAHMUD, S., BARMAN, N., HAQUE, M., UDDIN, M. & AHMED, R. 2019. Isolation, identification and antibiotic sensitivity pattern of Salmonella spp. from locally isolated egg samples. *Am. J. Pure Appl. Sci,* 1**,** 1-11.
SANGANI, S. & TOSHNIWAL, P. 2018. A COMPARATIVE STUDY OF WIDAL AGGLUTINATION TEST WITH BLOOD CULTURE TECHNIQUE IN THE DIAGNOSIS OF TYPHOID FEVER. *European Journal of Biomedical,* 5**,** 1017-1020.
SAUTEUR, P. M. M., STEVENS, M. J., PAIONI, P., WÜTHRICH, D., EGLI, A., STEPHAN, R., BERGER, C. & BLOEMBERG, G. V. 2020. Siblings with typhoid fever: An investigation of intrafamilial transmission, clonality, and antibiotic susceptibility. *Travel medicine and infectious disease,* 34**,** 101498.
SHAHAPUR, P. R., SHAHAPUR, R., NIMBAL, A., SUVVARI, T. K., SILVA, R. G. & KANDI, V. 2021. Traditional Widal agglutination test versus rapid immunochromatographic test in the diagnosis of enteric fever: A prospective study from South India. *Cureus,* 13.
UMAIR, M. & SIDDIQUI, S. A. 2020. Antibiotic susceptibility patterns of Salmonella typhi and Salmonella paratyphi in a tertiary care hospital in Islamabad. *Cureus,* 12.
YOUSAFZAI, M. T., QAMAR, F. N., SHAKOOR, S., SALEEM, K., LOHANA, H., KARIM, S., HOTWANI, A., QURESHI, S., MASOOD, N. & RAUF, M. 2019. Ceftriaxone-resistant Salmonella Typhi outbreak in Hyderabad City of Sindh, Pakistan: high time for the introduction of typhoid conjugate vaccine. *Clinical Infectious Diseases,* 68**,** S16-S21.