

TMSS Medical College Journal (TMCJ)

Volume 12, No. 01, January 2019

Contents

Editorial

- ▶ Learning environment and its impact on students' learning. 5-6
Dr. Hafiza Arjuman, Dr. Md. Azfarul Habib.

Original Articles

- ▶ Comparison of widal test and blood culture in the diagnosis of enteric fever in febrile patients. 7-13
Dr. Abu Bakar Siddique, Dr. Ripa Kundu, Dr. Md. Abdul Motaleb, Dr. Alok Kumar Sorker, Dr. Mirza Ruhul Amin, Dr. Shamima Akhter.
- ▶ Plasmid detection in uropathogenic Escherichia coli of urine of suspected urinary tract infection cases and its association with drug resistance. 14-21
Dr. Mst. Jeneya Afrin, Dr. Md. Abdullah Siddique, Dr. Arefa Akter, Dr. Taslima Yasmin, Dr. Farzana Afroz, Dr. Shamima Akhter.
- ▶ Pattern of chest X-ray in smear positive tuberculosis patient. 22-28
Dr. A.K.M. Towfiqul Alam, Dr. Debashis Kumar Gupta, Dr. Md. Moshir Rahman, Dr. A.K.M. Ahsan Habib, Dr. Md. Rezaul Alam, Dr. A.K.M Anowar Hossain.
- ▶ Subcutaneous antiseptic wick drain to prevent surgical site infections following acute & complicated appendectomy. 29-32
Dr. Md. Sayeedur Rahman, Dr. Md. Shariful Alam Khan, Dr. Md. Shaharul Alam Mondol, Dr. Iftexhar Md. Kudrat-e-Khuda.
- ▶ Association between duration of hypertension, level of systolic blood Pressure and ECG. 33-36
Dr. Md. Mahfuzul Islam, Dr. Mohammad Mahbub-ur Rahim, Dr. Md. Kamrul Islam, Dr. Md. Yunus Ali.
- ▶ Role of myringoplasty in hearing Improvement. 37-41
Dr. Md. Saiduzaman, Dr. Md. Nurul Islam, Dr. Md. Hasan Ali.

Case Report

- ▶ Cystic hygroma of breast in a child. 42-43
Dr. Md. Rezaul Islam



An Official Publication of TMSS Medical College



TMSS Medical College Journal (TMCJ)

January 2019

An Official Publication
of
TMSS Medical College, Bogura, Bangladesh

TMSS Medical College Journal (TMCJ)

Vol. 12, No. 1, January 2019

An Official Publication of TMSS Medical College, Bogura, Bangladesh

Editorial Board

Chairman : Prof. Dr. Md. Abdus Shukur
Editor in Chief : Prof. Dr. Md. Azfarul Habib
Assistant Editor : Dr. Md. Abdul Mazid
Advisory Editor : Prof. Dr. Moudud Hossain Alamgir
Managing Editor: Dr. Md. Matiur Rahman

Members

Dr. ASM Barkatullah
Prof. Dr. Md. Shahjahan Ali Sarker
Prof. Dr. Anup Rahman Choudhury
Prof. Dr. AKM Masudur Rahman
Prof. Dr. AKM Ahsan Habib
Dr. Md. Amirul Islam

Advisory Board

Prof. Dr. Hosne-Ara Begum
Prof. Dr. M Afzal Hossain
Prof. Dr. AKM Ahsan Habib
Prof. Dr. Md. Jawadul Haque
Prof. Dr. Sayed Mainul Hasan
Prof. Dr. Hafiza Arzuman
Shahzadi Begum

The TMSS Medical College Journal is published two times a year (January and July). It accepts original articles, review articles and case reports. While every effort is always made by the Editorial Board and the members of the Journal Committee to avoid inaccurate or misleading information appearing in the TMSS Medical College Journal, information within the individual article are the responsibility of its author(s). The TMSS Medical College Journal, its Editorial Board and Journal Committee accept no liability whatsoever for the consequences of any such inaccurate and misleading information, opinion or statement.

Printed by

TMSS Printing Press
Bogura, Bangladesh
e-mail: tmssprintingpress@gmail.com

Address of Correspondence

Executive Editor, TMSS Medical College Journal, TMSS Medical College
Rangpur Road, Thengamara, Bogura, Bangladesh.
Phone: 051-61830, Fax : 051-61830
e-mail: editortmcjournal@gmail.com, Web: www.tmssmedicalcollege.com

Original Article

Comparison of widal test and blood culture in the diagnosis of enteric fever in febrile patients

Siddique AB^{1*}, Kundu R², Motaleb MA³, Sorker AK⁴, Amin MR⁵, Akhter S⁶

1. Dr. Abu Bakar Siddique, Assistant Professor, Department of Medicine, Shaheed Ziaur Rahman Medical College, Bogura.
2. Dr. Ripa Kundu, Indoor Medical Officer, Shaheed Ziaur Rahman Medical College Hospital, Bogura.
3. Dr. Md. Abdul Motaleb, Associate Professor, Department of Medicine, Shaheed Ziaur Rahman Medical College, Bogura.
4. Dr. Alok Kumar Sorker, OSD, DG Health, Dhaka.
5. Dr. Mirza Ruhul Amin, Assistant Registrar, Shaheed Ziaur Rahman Medical College Hospital, Bogura.
6. Dr. Shamima Akhter, Associate Professor, Department of Pathology, TMSS Medical College, Bogura.

* Corresponding author

Abstract

Background: The enteric fever, caused by *Salmonella typhi*, is endemic in the sub-continent. The disease can occur in all age group with highest incidence among children. It is transmitted usually by close person to person contact with infected individuals or chronic carriers and rarely from person to person through faeco-oral route. **Objective:** To compare between blood culture and widal test for lab diagnosis of enteric fever. **Materials & Methods:** A comparative study was carried out Department of Medicine in Shaheed Ziaur Rahman Medical College Hospital, Bogura during July 2017 to December 2017. Hospitalized 100 patients with acute febrile illness were included in this study and fever for other causes of febrile illness or known disease and those who took antibiotic for the fever 1 week before admission were excluded from this study. **Results:** Widel test positive was found 61(61.0%) and negative 39(39.0%). Regarding blood culture 72(72.0%) patients was found culture positive and their organisms were salmonella typhi. Regarding bacteria of blood culture, it was observed that *S.thphi* was found 41(41.0%), *S.paratyphi* was 19(19.0%), non typhoidal salmonella 8(8.0%), other 4(4.0%) and negative blood culture 28(28.0%). The sensitivity (81.9%), specificity (92.9%), accuracy (85.0%), positive predictive value (96.7%), and negative predictive value (66.7%) of widal tests for diagnosis of enteric fever from febrile patients respectively. **Conclusion:** Widal test has a low sensitivity, specificity and positive predictive value, but it has good negative predictive value which indicates that negative widal test result have a good indication for the absence of the disease.

Key words: Enteric fever, Blood culture, Widel test, Febrile patients.

Introduction

The enteric fever, caused by *Salmonella typhi*, is endemic in the sub-continent. The disease can occur in all age group with highest incidence among children.¹ Typhoid fever causes significant morbidity and mortality in developing countries. Disease burden assessment is limited by lack of a high degree of sensitivity and specificity by many current rapid diagnostic tests.² Widal agglutination test which is the main laboratory procedure for diagnosis of typhoid fever in our rural and urban settings has received cautious acceptability and often outright rejection in

other places.³ Widal test is routinely used as diagnostic tool to rule out the disease. The diagnostic ability of Widal test is debatable as the test method has a low sensitivity, specificity and positive predictive value (PPV). Therefore, reviewing articles across the world regarding the diagnostic value of Widal test is necessary.⁴ This study was carried out to compare the value of a single acute phase widal test result by blood culture for the diagnosis of enteric fever and to assess how much the widal test is diagnostic for enteric fever where blood culture facilities is not available.

Materials and Methods

A comparative study was carried out in the Department of Medicine in Shaheed Ziaur Rahman Medical College Hospital, Bogura during July 2017 to December 2017. Hospitalized patients with acute febrile illness were included in this study and fever for other causes of febrile illness or known disease and those who took antibiotic for the fever 01 week before admission were excluded in this study. Enteric fever known a systemic febrile illness of prolonged duration that includes typhoid and paratyphoid fevers marked by step ladder fever, Diffuse abdominal pain, frontal headache, delirium, splenomegaly, hepatomegaly and many other systemic manifestations. Data was processed and analyzed by using computer based software SPSS- 23 (Statistical Package for Social Science). Qualitative variables were expressed as frequency and percentage and were compared between groups by Chi-square test or Fisher's Exact test as applicable.

Widal test

Qualitative slide agglutination and semi quantitative tube agglutination (titration) were performed using febrile antigen kits of Salmonella typhi (Chromatest Febrile Antigens kits, Linear chemicals, Barcelona, Spain). The slide agglutination test is used as a screening test for the presence of anti TO and anti TH antibodies in the patient's serum. For the slide agglutination test a drop of Salmonella typhi O and H antigens are added on a drop of serum on card and rotated at 100 rpm for one minute and reported as reactive or non reactive. For those slide agglutinations whose results are reactive and weakly reactive titer was determined. In the tube agglutination test (titration), serum sample was serially diluted by using fresh 0.95% saline preparation from 1:20 to 1:640 for anti TO and anti TH separately in 12 test tubes. Then a drop of O antigens and H antigens are added in the test tubes, equal amount in all. Based on the manufacturer manual, an antibody titer of 1:80 and higher for anti TO and 1:160 and higher for anti TH antibodies were taken as a cut of value to indicate recent infection of typhoid fever.

Blood Culture (FAN Method)

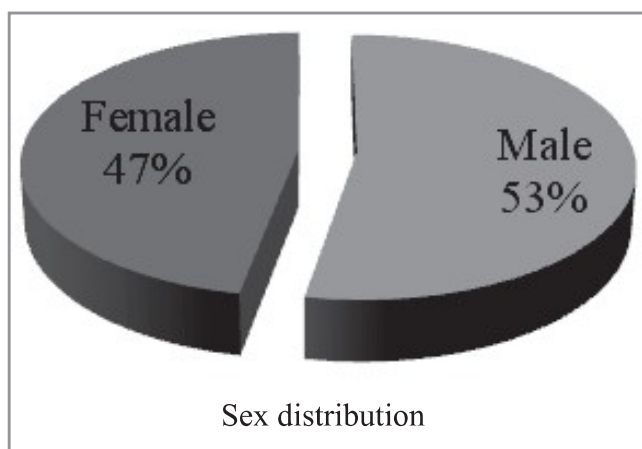
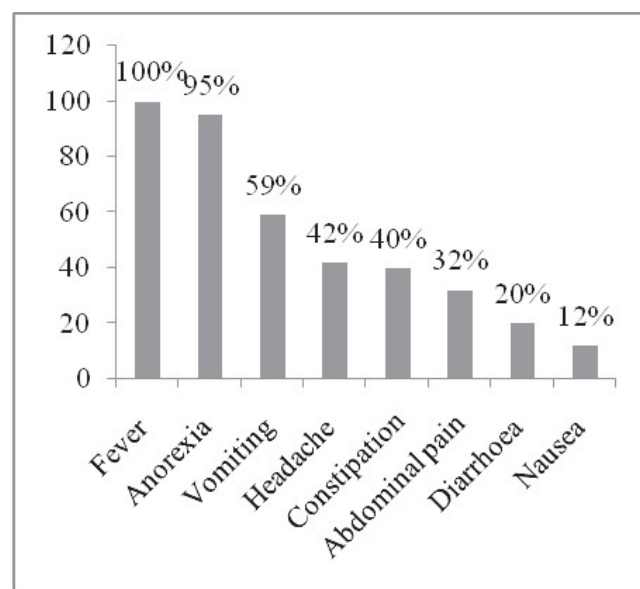
Using a sterile syringe and needle about 8-10 ml of blood from each adult study specimen and 3-5 ml of blood from each young child is to be collected and culture is done by BD (Becton, Dickinson) BACTEC™ FX 40 method.

Results

Regarding age of the study patients, it was observed that majority (59.0%) patients belonged to age 21-30 years. The mean age was found 29.3 ± 5.7 years with range from 15 to 45 years (I). In this study shows sex of the study patients, it was observed that more than half (53.0%) patients were male and 47(47.0%) patients were female. Male female ratio was 1.1:1 (Figure 1). Figure 2 shows symptoms of the study patients, it was observed that all (100.0%) patients had fever followed by 95(95.0%) had anorexia, 59(59.0%) had vomiting, 42(42.0%) had headache and 40(40.0%) had constipation. Table II shows widal test of the study patients, it was observed that 25(25.0%) cases was found in both (TO and TH)=1:160, followed by 17(17.0%) in TO=1:320 and TH=1:160, 14(14.0%) found TO=1:160 and TH=1:320, 03(3.0%) in AO > 1:160 and AH=1:320, 02(2.0%) in BO>1:160 and BH =1:320 and 39(39.0%) in negative. Figure 3 shows blood culture of the study patients, it was observed that 72(72.0%) patients was found blood culture positive and their organisms were salmonella typhi. Regarding bacteria of blood culture, it was observed that *S.typhi* was found 41(41.0%), *S.paratyphi* was 19(19.0%), non typhoidal salmonella 8(8.0%), other 4(4.0%) and negative blood culture 28(28.0%) (Table III). Present study showed association between widal test and blood culture, it was observed that 61 patients had widal test positive among them 59 were blood culture positive and 02 were negative. Thirty nine (39) patients had widal test negative among them 13 were blood culture positive and 26 were negative. The difference was statistically significant ($p < 0.05$) between two groups (Table IV). The sensitivity (81.9%), specificity (92.9%), accuracy (85.0%), positive predictive value (96.7%), and negative predictive value (66.7%) of widal tests for diagnosis of enteric fever from febrile patients respectively (Table V).

Table I: Distribution of the study patients by age (n=100)

Age (in year)	Number of patients	Percentage
≤ 20	15	15.0
21-30	59	59.0
>30	26	26.0
Mean ±SD	29.3±5.7	
Range (min-max)	15-45 years	

**Figure 1: Distribution of the study patients according to sex (n=100)****Figure 2: Distribution of the study patients according to symptoms (n=100)****Table II: Distribution of the study patients according to widal test (n=100)**

Widal test	Number of patients	Percentage
Both (TO and TH)=1:160	25	25.0
TO=1:320 and TH=1:160	17	17.0
TO=1:160 and TH=1:320	14	14.0
AO > 1:160 and AH=1:320	03	03.0
BO>1:160 and BH =1:320	02	02.0
Negative	39	39.0

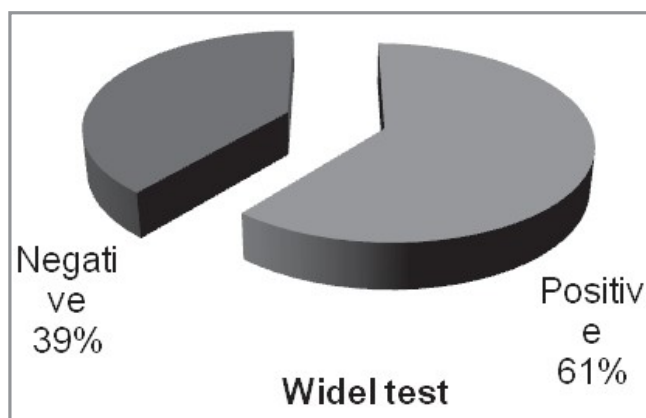
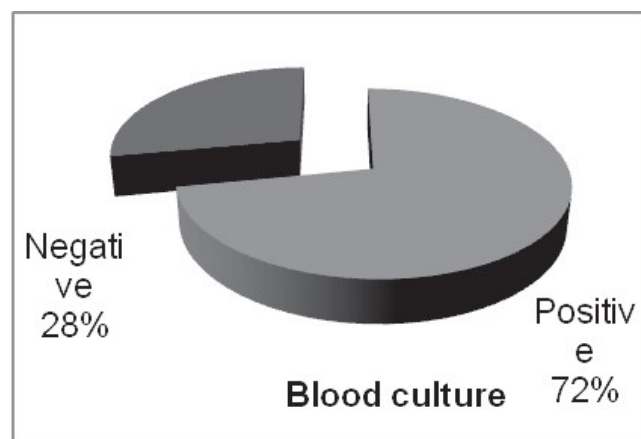
**Figure 3: Distribution of the study patients according to widal test (n=100)****Figure 4: Distribution of the study patients according to blood culture (n=100)**

Table III: The distribution of blood culture results of febrile patients suspected of typhoid fever (n=100)

Bacteria	Number of patients	Percentage
<i>S. typhi</i>	41	41.0
<i>S. paratyphi</i>	19	19.0
Non typhoidal salmonella	8	8.0
Other bacteria	4	4.0
Negative	28	28.0

Table IV: Association between widal test and blood culture (n=100)

Widal test	Blood culture		Total	p value
	Positive	Negative		
Positive	59	02	61	0.001 ^s
Negative	13	26	39	
Total	72	28	100	

s= significant

P value reached from Chi square test

Table V: The sensitivity, specificity, accuracy, PPV and NPV of widal tests for diagnosis of enteric fever from febrile patients

Validity of test	Percentage (%)
Sensitivity	81.9
Specificity	92.9
Accuracy	85.0
Positive predictive value (PPV)	96.7
Negative predictive value (NPV)	66.7

Discussion

In this study showed that majority (59.0%) patients belonged to age 21-30 years. The mean age was found 29.3 ± 5.7 years. In study of Anduallem et al.⁵ showed their study age ranged from 8–80 years ($M = 35.82 \pm 12.4$ [SD]) and most of them were 15–19 years (94.3%). In another study Nsutebu et al.⁶ observed Patients' ages ranged from 4 to 62 years [mean 23.1 ± 17.8 years (SD)].

In this study it was observed that more than half (53.0%) patients were male and 47(47.0%) patients were female. In study of Anduallem et al.⁵ showed one hundred and eighty six (68.9%) participants were females and eighty four (31.1%) were males. Nsutebu et al.⁶ observed that eighty-seven (43.5%) of the patients were male while 113 (56.5%) were female.

In current study was observed that all (100.0%) patients had fever followed by 95(95.0%) had anorexia, 59(59.0%) had vomiting, 42(42.0%) had headache and 40(40.0%) had constipation. Shah et al.⁷ common symptoms were fever (100%), Common symptoms were fever (100%), vomiting 37 (31.09%), diarrhea 27 (22.68%), abdominal pain 31 (26%) cough 24 (20%), and constipation six (5%). Gosai et al.⁸ Predominant symptoms were fever, abdominal pain, vomiting and headache. Joshi et al.⁹ All (100.0%) patients had fever followed by 52.5% had headache, 22.5% had abdominal pain, 20% had vomiting, 15.0% had constipation and 10.0% had diarrhea. Modi¹⁰ study showed fever was the chief presenting complaint present in all the patients (100%). The other most common symptoms were abdominal pain (57.14%), vomiting (50%), anorexia (30.61%), and cough (13.26%). Other complaints such as body ache (4.08%), headache (3.06%), constipation (2.04%), irritability (1.02%), aphasia (1.02%), and convulsions (1.02%) were present in small number of patients. Nsutebu et al.⁶ study showed fever was the most common symptom (100%), followed by fatigue (38.5%), headache (32.5%) and anorexia (32.5%), abdominal pain (23.5%), vomiting (23.0%), diarrhoea (14.5%).

In present study, observed that 25(25.0%) cases was found in both (TO and TH)=1:160, followed by 17(17.0%) in TO=1:320 and TH=1:160, 14(14.0%) found TO=1:160 and TH=1:320, 03(3.0%) in AO > 1:160 and AH=1:320, 02(2.0%) in BO>1:160 and BH =1:320 and 39(39.0%) in negative. Kakaria et al.¹¹ A single estimation of significant titre of widal test was suggestive of Typhoid fever in 80% cases. Among the widal positive cases blood culture was positive in 18(36%). Hyder et al.¹² study diagnosis was made on

clinical impression along with positive widal test in all 40 cases (including the 4 blood culture positive cases). High positivity of widal test has been reported by some authors.¹³⁻¹⁴ *Salmonella typhi* is a major cause of enteric fever in Bangladesh. Of the paratyphoid fevers, those cause by *Salmonella paratyphi A* is more common.¹⁵ Jeeyani et al.¹⁶ widal test was positive in 33 of 62 patients (53.2 %). Gopalakrishnan et al.¹⁷ of the 144 samples, 121 were widal positive.

In this study revealed that blood culture of the study patients was observed that 72(72.0%) patients was found blood culture positive and their organisms were *salmonella typhi*. Jeeyani et al.¹⁶ the blood culture of 34 of the 62 study patients (89.5%) were positive for *Salmonella typhi* and 4 (10.5%) were positive for *Salmonellaparatyphi A*. Kakaria et al.¹¹ twenty four cases was found blood culture positive and 26 were blood culture negative. Shah et al.⁷ among 119 positive blood culture, 104 (87.39%) were *Salmonella typhi* and 15 (12.6%) were *Salmonella Paratyphi A*. Hyder et al.¹² showed only 4(15.3%) cases were blood culture positive. In this study the high positive result of blood culture is probably due to FAN Method.

Regarding bacteria of blood culture, it was observed that *S.thphi* was found 41(41.0%), *S. Paratyphi* was 19(19.0%), non typhoidal salmonella 8(8.0%), other 4(4.0%) and negative blood culture 28(28.0%). In study of Anduaem et al.⁵ among 5/7 (71.4%) of culture confirmed typhoid cases by *S.typhi* as compared with 2/4 (50%) of *S. paratyphi* and 3/7 (42.9%) of non typhoidal salmonella. In addition to *S.typhi* and *S.paratyphi* other bacteria were identified from blood culture of the febrile patients who had positive or negative widal titer. In the current study seven (2.6%) cases of non typhoidal salmonella and 51 (18.8%) cases of other bacteria were identified from blood culture. The result of non typhoidal salmonella (2.6%) was similar to a study done in Tanzania; the study identified 2.9% of non typhoidal salmonella from blood culture.¹⁸⁻¹⁹ Positive widal titers were also seen in cases of nontyphoidal salmonella and in blood culture positive cases for other bacteria. Three out of 7 (42.9%) of nontyphoidal salmonella cases and 17 of the 51 (33.3%) other bacteria positive cultures had a positive titer of anti TO.²⁰

In current study observed that 61 patients had widal test positive among them 59 were blood culture positive and 02 were negative. Thirty nine (39) patients had widal test negative among them 13 were blood culture positive and 26 were negative. Anduaem et al.⁵ observed that two cases among seven culture confirmed typhoid fever cases had a negative titer. The false negative widal test results were there probably because blood was collected too early in the disease processes, or inoculated bacterial load is inadequate to induce the antibody production.²¹ Previous antibiotic treatment may also contribute to negative widal agglutination test but there was no patient who explained taking antibiotic within two weeks before coming for the diagnosis during this study.

In this study revealed that the sensitivity (81.9%), specificity (92.9%), accuracy (85.0%), positive predictive value (96.7%), and negative predictive value (66.7%) of widal tests for diagnosis of enteric fever from febrile patients respectively. Anduaem et al.⁵ study observed that the sensitivity, specificity, Positive predictive Value and Negative predictive Value of widal test were 71.4%, 68.44%, 5.7% and 98.9% respectively. Wilke et al.²² the second widal test for both group 1 and group 3 has a sensitivity and specificity of over 90%, a PPV of 88%, and an NPV of 93%, with 90% acuity. Gopalakrishnan et al.¹⁷ the sensitivity and specificity of widal were calculated to be 100% and 21.2% respectively, with a PPV of 40.3%, NPV of 100% and efficiency of test of 48.6%. Studies conducted by Jackson et al.²³ showed a higher specificity (91%), sensitivity (90%), PPV (76%) and NPV (97%), but the study group included only 21 samples in Typhoid group compared to 144 samples in our study. When compared with the studies by Karamat et al.²⁴, sensitivity (92%) and specificity (96%) were higher, but here the study group consisted of only 25 samples.

Conclusion

Clinical presentation, signs and symptoms of typhoid fever patients are varying. Widal test has a low sensitivity, specificity and positive predictive value,

but it has good negative predictive value which indicates that negative widal test result have a good indication for the absence of the disease. Nevertheless, using widal test as the only laboratory test for the diagnosis of typhoid fever will result in misleading diagnosis. Therefore, it is very essential to use blood culture to diagnose enteric fever.

References

1. Sangani S and Toshniwal P. A comparative study of widal agglutination test with blood culture technique in the diagnosis of typhoid fever. *European Journal of Biomedical and Pharmaceutical Sciences*, 2018;5(4):1017-1020
2. Ajibola O, Mshelia MB, Gulumbe BH and Eze AA. Typhoid Fever Diagnosis in Endemic Countries: A Clog in the Wheel of Progress? *Medicina* 2018;54(23):2-12.
3. Jombo GT, Mbaawuaga EM, Ahmed U et.al. Patterns of Widal test results among patients suspected to have typhoid fever attending Bsouth Makurdi Nigeria. *Int J Health Sci Res*. 2019; 9(1):6-12.
4. Mengist HM and Tilahun K. Diagnostic Value of Widal Test in the Diagnosis of Typhoid Fever: A Systematic Review. *J Med Microb Diagn* 2017, 6:1:2-4
5. Andualem G, Abebe T, Kebede N, Gebre-Selassie S, Mihret A and Alemayehu H. A comparative study of widal test with blood culture in the diagnosis of typhoid fever in febrile patients. *BMC Research Notes* 2014, 7:653
6. Nsutebu EF, Martins P, Adiogo D. Prevalence of typhoid fever in febrile patients with symptoms clinically compatible with typhoid fever in Cameroon. *Trop Med Int Health*. 2003; 8 (6): 575-578.
7. Shah G, Ghimire A, Shrestha S. Clinical profile of culture proven enteric fever in children at university teaching hospital, Nepal. *Journal of Patan Academy of Health Sciences* 2014; 1(2):42-45.
8. Gosai MM, Hareshwaree HB, Purohit PH, Momin A G. A Study of Clinical Profile of Multidrug Resistant Typhoid Fever In Children. *NJIRM* 2011; 2(3): 87-90.
9. Joshi BG, Keyal K, Pandey R, Shrestha BM. Clinical Profile and Sensitivity Pattern of Salmonella Serotypes in Children: A Hospital Based Study. *J Nepal Paediatr Soc* 2011; 31(3): 180-183.
10. Modi R, Clinical profile and treatment outcome of typhoid fever in children at a teaching hospital, Ahmedabad, Gujarat, India. *International Journal of Medical Science and Public Health* 2016; 5(02): 212-216.
11. Kakaria A, Asgaonkar D, Narkhede M. Clinical profile of enteric fever: a prospective study of fifty enteric fever patients. *Int J Res Med Sci*. 2014 2(4): 1620-1625.
12. Hyder RT, Yasmeen BHN, Ahmed S, Islam MS, Islam F. Clinical profile and Outcome of Enteric Fever in hospitalized children aged 6 months to 2 years. *Northern International Medical College Journal* 2013; 5(1): 301-305.
13. Kulkarni ML, Rego SJ. Value of single widal test in the diagnosis of typhoid fever. *Indian Pediatr* 1994 ; 31 : 1373-77.
14. Pang T, Puthuchear SD. Significance and value of the widal test in the diagnosis of typhoid fever in an endemic area. *J Clin Pathol*, 1983; 36 : 471-75.
15. Chowdhury MAY, Haque MG, Karim AMMR. Value of widal Test in the Diagnosis of Typhoid Fever. *Orginal article* 2015; 27(02), pp. 38-32.
16. Jeeyani HN, Prajapati BS, Bloch A. Enteric Fever in Children-Clinical Profile, Sensitivity Patterns and Response to Antimicrobials. *GCSMC J Med Sci*, Vol (IV) No (I) 2015, 40-43.
17. Gopalakrishnan V, Sekhar WY, Soo EH, Vinsent RA, Devi S. Typhoid fever in Kuala Lumpur and a

- comparative evaluation of two commercial diagnostic kits for the detection of antibodies to salmonella typhi. Singapore Med J. 2002; 43 (7): 354-358.
18. Mamo Y, Belachew T, Abebe W, Gebre-Selassie S, Jira C: Pattern of widal agglutination reaction in apparently healthy population of Jimma town, southwest Ethiopia. Ethiop Med J 2007, 45(1):69–77.
 19. Feasey NA, Archer BN, Heyderman RS, Sooka A, Dennis B, Gordon MA, Keddy KH: Typhoid fever and invasive nontyphoid Salmonellosis, Malawi and South Africa. Emerg Infect Dis 2010, In press article.
 20. Aftab R, Khurshid R: Widal agglutination titre: a rapid serological diagnosis of typhoid fever in developing countries. Pak J Physiol 2009, 5(1) :65–67.
 21. Olopoenia LA, King AL. Widal agglutination test - 100 years later: still plagued by controversy. Postgrad Med J. 2000; 76: 80-84.
 22. Willke A, Ergonul O, Bayar B. Widal test in diagnosis of typhoid fever in turkey. Clin Diagn Lab Immunol. 2002; 9 (4): 938-941.
 23. Jackson AA, Ismail A, Ibrahim TAT, Kader ZS and Nawi NM. Retrospective review of dot enzyme immunoassay test for typhoid fever in an endemic area. Southeast Asian J Trop Med Public Health 1995; 26(4): 625-30.
 24. Karamat KA, Usman J, Ahmed TA, Abbas SH. Detection of serum IgM against *Salmonella typhi*: A rapid diagnostic technique. J College Physicians Surgeons Pakistan 1998; 8(4):170-3.