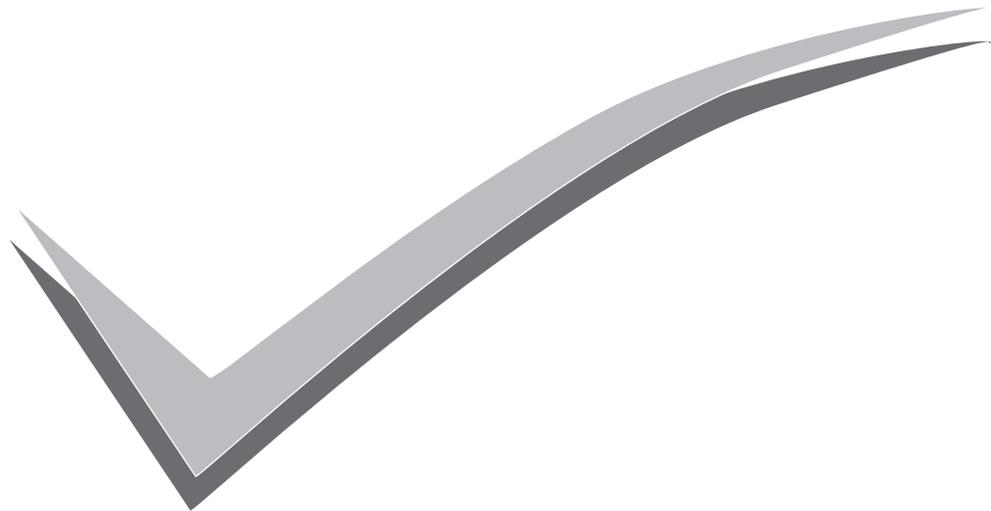




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Performance Evaluations



ENTEROCHECK[®]-WB

Rapid test for the detection of IgM antibodies to *S. typhi*

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Evaluation of Enterocheck WB[®] test in Diagnosis of Typhoid Fever among Egyptian Adults

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ABSTRACT

Background: Enteric fever is a potentially severe systemic febrile illness that is usually presented with non-specific symptoms and signs. Its diagnosis is usually confirmed by blood culture and stool culture besides the Widal agglutination test. The delayed results of microbiologic examination and the unreliable results of Widal test in endemic countries makes the search for a rapid and reliable method for diagnosis of typhoid fever more mandatory. **Objectives:** This study aimed to compare the performance of the Enterocheck WB[®] test as a rapid serological test to that of the Widal test for diagnosis of typhoid fever in acute stage taking blood culture as the gold standard method. **Methodology:** A total of 154 patients that had been clinically suspected as typhoid fever were investigated by the blood culture, Widal tube agglutination test and Enterocheck WB[®] test in addition to 46 healthy controls were investigated by Widal tube agglutination test and Enterocheck WB[®] test. **Results:** Enterocheck WB[®] test showed higher sensitivity and specificity, 86% and 89% than Widal tube agglutination test, 81% and 71% respectively. Enterocheck test had higher sensitivity when fever duration <5 days (100%). **Conclusion:** Enterocheck WB[®] test is a useful, rapid easy serological test for early diagnosis of typhoid fever especially for primary healthcare centers and outpatient clinics as well as hospitals.

Key words: Enterocheck test; Typhoid fever; Widal test

INTRODUCTION

Typhoid fever is a potentially severe systemic febrile illness that is caused by *Salmonella enteric serotype typhi* (*S. typhi*) and remains a significant health problem in many developing countries. Typhoid fever is considered an endemic disease in Mediterranean North African countries with estimated medium incidence of 10 to 100 cases per 100,000 persons. Egypt remains a country with intermediate incidence of one to 100 per 100,000 cases of enteric fever/year^[1,2]. Enteric fever is usually presented with non-specific symptoms and signs such as slow gradually progressive fever, nausea, vomiting, abdominal pain, malaise, headache, constipation then diarrhea and hepatosplenomegally. It can be complicated by serious complication such as intestinal hemorrhage, intestinal perforation in the distal ileum, septicemia, diffuse peritonitis, encephalitis and cholecystitis^[3]. With the unclear and inconclusive clinical picture of typhoid fever, many cases are usually missed or misdiagnosed on the clinical basis. Microbiologic culture of a blood sample is considered to be the gold test for the diagnosis of typhoid fever even though it takes up to seven days for isolation of the causative organism^[4]. Widal agglutination was introduced as a

serologic technique to aid in diagnosis of typhoid fever. The test is based on demonstrating the presence of agglutinin (antibody) in the serum of an infected patient, against the H (flagellar) and O (somatic) antigens of *Salmonella typhi*. Though the Widal test is extensively used, it cannot give reliable diagnostic results in endemic regions due to difficulty in establishing a steady-state baseline titer and cross reactivity with other organisms^[5,6]. Rapid diagnostic methods is required that should be reliable, easy to perform with comparable sensitivity and specificity. Numerous serological kits are available with variable reported sensitivities and specificities in diagnosis of typhoid fever as Typhidot M which is a dot enzyme immunoassay for detection of specific IgM antibodies to *S. typhi*^[7,8] and Tubex Idl that is a semi-quantitative calorimetric test for detection of anti-*Salmonella* 09 antibodies in serum^[9]. Enterocheck WB[®] is a rapid immunoassay which works on the principle of immunochromatography for the detection of IgM antibodies to lipopolysaccharide (LPS) specific to *S. typhi* in human serum, plasma or whole Blood. A study in India reported Enterocheck WB[®] as a very useful test for diagnosing typhoid fever in children and another study reported the sensitivity & specificity of the Enterocheck WB[®] test to be 89.47% and 96.87% respectively^[10]. This study compared the performance of the

Enterocheck WB[®] test to that of the Widal test taking blood culture as the gold standard method in the diagnosis of typhoid fever among Egyptian adults.

SUBJECTS & METHODS

This observational cross sectional study was conducted in Abassia fever hospital- Cairo-Egypt between September 2012 and March 2014. One hundred and fifty four febrile patients admitted to Abassia fever hospital with clinical suspicion of typhoid fever in acute stage (fever >38°C for less than 14 days, headache, constipation, diarrhea, hepatomegaly, and splenomegaly), and 46 healthy volunteers of matched age were enrolled in this study after informed consent.

Specimen collection Ten ml of blood samples were collected from each febrile patient. Five ml for blood culture and 5 ml were allowed to clot then centrifuged, sera collected and divided into 2 aliquots, one used for Widal tube agglutination test, and the other had been stored at -70°C for Enterocheck WB[®] test. Regarding the control group, 5 ml of blood were collected, allowed to clot, centrifuged, and sera stored in aliquots at -70°C for Widal test and Enterocheck WB[®] test.

Blood culture Biphasic blood culture bottles (Biolife – Egypt) had been inoculated with blood samples and incubated aerobically at 37°C. Bottles were checked for growth on the solid agar every 24 hours. Subsequent sub-cultures were made on MacConkey's agar, Salmonella-Shigella (S.S) agar and blood agar media. Final sub-culture was made from fluid phase on the seventh day - in cases with negative bacterial growth on solid phase till seventh day-before reporting a negative blood culture result. The growth of *S. typhi* was identified by the standard biochemical tests and it was confirmed

by agglutination with the Salmonella polyvalent 'O', 09 and the H:d antisera (Remel-England)^[11].

Widal tube agglutination test The Widal tube agglutination test was performed for all cases enrolled in the study. According to the manufacturer's instructions (Atlas medical, Cambridge, UK), One drop of each antigens specifically for *S.typhi* serotyping (O:9 and H:d antigens from Atlas Medical-Cambridge: Atlas febrile antigens, lot: 12090917) was added to each test tube contains patient serum in double folded serial dilution (1/40, 1/80, 1/160, 1/320, and 1/640) in 2 rows one for O antigen and second row for H antigen. Additional saline control tube was added to each raw. According to *Ministry of Health and Population, Egypt*^[12] after incubation for 24 hours at 37° C, the test is considered positive if O titer is $\geq 1/160$ and H titer is ≥ 160 .

Enterocheck WB[®] test Enterocheck-WB[®] (Zephyr Biomedicals, Tulip Group, Goa India) is a rapid, qualitative, sandwich immunoassay for the detection of IgM antibodies to *S. typhi* in human serum/plasma or whole blood specimen using the principle of immunochromatography. In this study, the Enterocheck WB[®] test was conducted on serum samples according to kit instructions. The foil containing the testing device and the sample loop was brought to room temperature, the testing device was labeled and placed horizontally and 5 μ l of serum was dispensed into specimen port A using the loop provided. Five drops of sample running buffer were added to buffer port B kept at room temperature for 15 min. Samples negative for IgM to LPS of *S. typhi* showed only one coloured control band in the control (C) window, while positive samples yielded two coloured bands in the test (T) and control (C) window^[13] (**figure 1**)

Statistical analysis Statistical presentation and analysis of this study was conducted, using the mean, standard deviation, Chi-square ROC curve by SPSS V17.

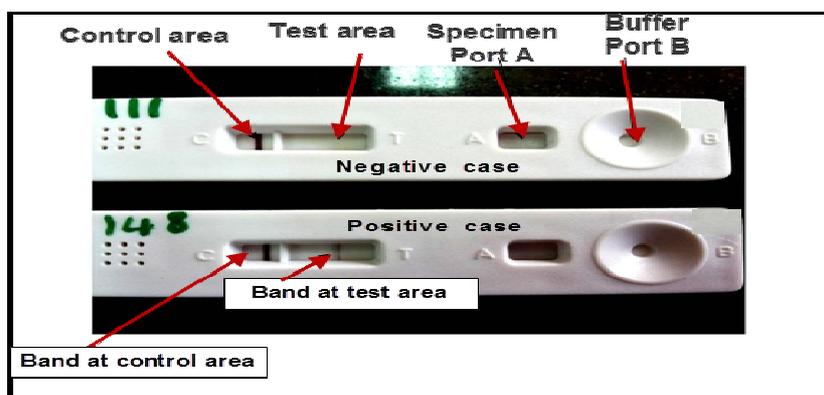


Figure 1: Interpretation of Enterocheck WB[®] test results. (Negative and positive cases)

RESULTS

This study included 154 patients and 46 controls of matched age and sex.

Blood culture results were positive for *S.typhi* in 21/154 (13.6%) of the patients, 4/154 (2.6%) of blood cultures showed growth of non typhi serotypes of *S. enteric (as Salmonella paratyphi A and B)*, 6/154 (3.9%) yielded coagulase negative staphylococci species and 123/154 (79.9%) had negative blood culture results (the total cases with negative results for *S.typhi* were 133/154; 86.4%). The clinical manifestations among patients are summarized in **table 1**, 70/154 patients (48.2%) complained of fever for less than 5 days while 84/154 patients (57.9%) had fever duration \geq 5 days.

Out of 21 cases of positive blood culture for *S.typhi*, Widal test was positive in 17(81%) of the cases while Enterocheck test was positive in 18 cases (85.8%). The individual positive results for Widal test and Enterocheck was recorded in 39 (29.3%) and 14 (10.5%) of the 133 cases with negative blood culture results for *S.typhi*, respectively, only 12/133 (9%) were positive for

both Widal test and Enterocheck test. None of the 46 controls included in the study was positive for Enterocheck test while 3 of them were positive for the Widal test (6.5%).

Regarding fever duration, only two patients of 70 patients with fever duration < 5 days had positive blood culture results, these 2 patients were positive for Enterocheck test (100%) and only 1 case was positive for Widal test (50%). For the other 68 patients with negative blood culture results and fever for less than 5 day, only 1 case (1.5%) showed positive results for Enterocheck test and 11 cases (16%) were positive for Widal test (**Table 3**). Out of the 84 patients with fever duration of 5-13 days, 19 patients gave positive blood culture results. 18/19 patients (95%) were positive for Enterocheck test and 16/19 of patients (84%) were positive for widal test. 11/65 patient (17%) with negative blood culture and fever duration \geq 5 days and positive for Enterocheck test while Widal test was positive in 28 patients out of the 65 (43%) cases with negative blood culture and fever duration \geq 5 days (**Table 4**).

Table 1: Clinical manifestations among patients

	Positive blood culture N = 21	Negative blood culture N = 133	Total N = 154	Chi-square	
				X ²	P-value
Fever					
< 5 days	2 (9.5%)	68 (51.1%)	70 (48.2%)	11.039	0.009 *Hs
\geq 5 days	19 (90.5%)	65 (48.9%)	84(57.9)		
Abdominal pain	18 (85.7%)	83 (62.4%)	110 (55%)	3.394	0.065 **NS
Diarrhea	7 (33.3%)	55 (41.4%)	62 (40.3%)	0.209	0.647 **NS
Constipation	14 (66.7%)	78 (58.6%)	92 (59.7%)	0.008	0.930 **NS

Taking blood culture results for isolation of *S.typhi* as a gold standard test, sensitivity, specificity, the positive predictive value (PPV) and the negative predictive value (NPV) of Widal test were 81%, 71%, 34%, and 96%

respectively and of Enterocheck test were 86%, 89%, 56%, and 98% respectively . Agreement between blood culture results and Widal test was 72% while for Enterocheck test was 82% (**Table 2**)

Table 2: Comparative evaluation of the diagnostic tests

Blood culture for <i>S.typhi</i>	Widal test		Enterocheck WB test	
	Positive	Negative	Positive	Negative
Positive (n= 21)	17	4	18	3
Negative (n=133)	39	94	14	119
Sensitivity	81%		86%	
specificity	71%		89%	
PPV	34%		56%	
NPV	96%		98%	
Agreement	72%		82%	

NPV: the negative predictive value, PPV: the positive predictive value

The relationship between time interval since onset of fever and reaction positivity was studied (table 3 and 4). In patients with fever for <5 days, Enterocheck test had sensitivity of 100% and in those with fever for 5-13 days the recorded sensitivity was 95%. While the Widal test recorded sensitivity 50% for fever <5 days that increases to 84% for duration of 5-13 days.

Table 3: Comparative evaluation of the diagnostic tests for fever duration <5 days

Blood culture	Enterocheck		Widal	
	Positive	Negative	Positive	Negative
Positive n=2	2 (100%)	0 (0%)	1 (50%)	1 (50%)
Negative n= 68	1 (1.5%)	67 (98.5%)	11 (16%)	57 (84%)
Sensitivity	100%		50%	
Specificity	99%		84%	

Table 4: Comparative evaluation of the diagnostic tests for fever duration from 5 to 13 days

Blood culture	Enterocheck		Widal	
	Positive	Negative	Positive	Negative
Positive n=19	18 (95%)	1 (5%)	16 (84%)	3 (16%)
Negative n= 65	11 (17%)	54 (83%)	28 (43%)	37 (57%)
Sensitivity	95%		84%	
Specificity	83%		83%	

DISCUSSION

Typhoid fever is a systemic illness with a significant morbidity and mortality in developing countries. Poor sanitation, overcrowding, low standard of living, lack of medical facilities, and indiscriminate use of antibiotics lead to endemicity of typhoid fever^[14]. Blood culture is the gold standard for diagnosis of typhoid fever. However in this study the isolation rate for *S.typhi* was relatively low (13.6%). **Anusha et al.^[13] in India reported** similar results as the positive cases of blood culture for *S.typhi* was only 12%. Different isolation rates for diagnosis of typhoid fever is common between different countries and even between different areas particularly in Egypt and can be explained by the different rates of empiric use of antimicrobial and the degree of application of antibiotics stewardship^[15]. Using automated blood culture systems increases the isolation rate and sensitivity of blood culture as in the study done by **Fadeel et al.^[16] in Egypt** that included 332

Patients <18 years using automated Bact/ Alert system and higher isolation rate (22.89%) was reported.

In this study the sensitivity and specificity of Widal test were 81% and 71% respectively with 34% PPV and 96% NPV. Similar results had been reported by **Rahman et al.^[17] in Bangladesh**, the sensitivity and specificity of the Widal test were 81% and 71% respectively. **Dutta et al.^[18] in India** showed much lower sensitivity (37%) than the current study but with comparable specificity (95%). **Siba et al.^[8] in Papua New Guinea** had the same low sensitivity of Widal test the sensitivity (40%) but with specificity 97.5%, 71.4% PPV and 91.2% NPV. The interpretation of the Widal test remains problematic to this day, with a great number of articles reporting different cut-offs and the test has lost some popularity in recent years as technical skills are required for its performance and interpretation, different sensitivity and specificity rates are obtained even in same region^[19]. Increased false positive results that affects the specificity of Widal test is usually encountered due to previous immunisation with *Salmonella* antigen, cross-reaction with non-typhoidal *Salmonella*, variability and poorly standardized commercial antigen preparation, infection with malaria or other enterobacteriaceae or other diseases such as dengue fever or due to the high background rates of circulating antibodies to serotype Typhi or other *Salmonella* serotypes^[5,20]. **Bakr et al.^[15]** stated that different reports of sensitivity and specificity of widal test are usually present due to different commercial antigens brands used. The 3 positive results of Widal test among healthy controls may be due to repeated exposures to *S. typhi* in endemic regions or cross-reactivity with other non-*Salmonella* organisms^[5].

Enterocheck test showed high sensitivity (86%) and specificity (89%) with a positive predictive value 56% and negative predictive value 98%. **Anusha et al.^[13] in India**, reported similar results; sensitivity and specificity of (85.45%) and (88.6%) respectively and positive predictive value 51.1%. Higher sensitivity (89.47%) was reported by **Anagha et al.^[10] in India** that could be due to the lower number of cases enrolled in the study (83) compared to our study (154). The 3 false negative cases can be explained by the test's failure to detect low levels of antibodies that are below its detection level^[20] while the 14 positive results with negative blood culture might be due to prior treatment with antimicrobials or low bacterial counts in blood that affects blood culture results or the presence of background levels of agglutinins and

antibodies to *S. typhi* in endemic areas, the prevalence of these antibodies has been shown to increase with age^[13].

In this study the Enterocheck test showed higher agreement with blood culture (82%) than Widal test (72%). This was also detected by *Anusha et al.*^[13] as agreement of Enterocheck with blood culture has been calculated as (88%). Enterocheck WB had a higher sensitivity (100%) when fever duration < 5 days, this indicates that the Enterocheck test detects IgM seroresponse for *S. typhi*-infected subjects in early phase of illness that helps in early diagnosis and early therapeutic intervention^[21].

CONCLUSION

Taking blood culture results for *S.typhi* isolation as the golden standard in this study, the Enterocheck test showed higher sensitivity and specificity than Widal test, besides its simplicity and rapid turn round time (15 minutes), it can be effectively used as useful diagnostic tool for typhoid fever in different health care settings especially where blood culture facilities are not available preferably in combination with Widal test not as an alternative diagnostic test till more detailed studies are available to assess its performance taking other diagnostic methodologies as a golden standard . As it detects IgM antibodies it is of value as a marker of recent infection offering better chance for early diagnosis and treatment for patients when typhoid fever is clinically suspected. Large scale studies taking other serological tests as ELISA test for *S.typhi* IgM or molecular diagnostic methods for diagnosis of detection of *S.typhi* infections as golden standards are highly recommended to overcome the drawbacks of blood culture low isolation rates that affects the sensitivity and specificity of the studied Enterocheck test. This helps to evaluate Enterocheck test more accurately compared to the traditional Widal test.

REFERENCES

1. **Affi S, Earhart K, Azab MA, Youssef FG, El Sakka H, Wasfy M, Mansour H, Oun S, Rakha M and Mahoney F (2005):** Hospital-based surveillance for acute febrile illness in Egypt: a focus on community-acquired bloodstream infections. *Am J Trop Med Hyg* 73: 392-399.
2. **Connor BA and Schwartz E (2005):** Typhoid and paratyphoid fever in travelers. *Lancet Infect Dis* 5: 623-628.
3. **Clark TW, Daneshvar C, Pareek M, Perera N and Stephenson I (2010):** Enteric fever in a UK regional infectious diseases unit: a 10 year retrospective review. *J Infect*; 60:91–98.
4. **Parry CM, Hien TT, Dougan G, White NJ and Farrar JJ. (2002):** Typhoid fever. *N Engl J Med.* 28; 347(22):1770-82.
5. **Olopoenia LA and King AL. (2000):** Widal agglutination test – 100 years later: still plagued by controversy. *Postgrad Med J.*76:80-4.
6. **Yang J, Acosta CJ, Si GA, Zeng J, Li CY, Liang DB, Ochiai RL, Page AL, Danovaro-Holliday MC, Zhang J, Zhou BD, Liao HZ, Wang ML, Tan DM, Tang ZZ, Gong J, Park JK, Ali M, Ivanoff B, Liang GC, Yang HH, Pang T, Xu ZY, Donner A, Galindo CM, Dong BQ and Clemens JD (2005):** A mass vaccination campaign targeting adults and children to prevent typhoid fever in Hechi; expanding the use of Vi polysaccharide vaccine in southeast China: a cluster-randomized trial. *BMC Public Health.*; 18: 49.
7. **Beig FK, Ahmad F, Ekram M and Shukla I(2010):** Typhidot M and Diazo test vis-à-vis blood culture and Widal test in the early diagnosis of typhoid fever in children in a resource poor setting. *Braz J Infect Dis.*; 14(6):589-93.
8. **Siba V, Horwood PF, Vanuga K, Wapling J, Sehuko R, Siba P and Greenhilla AR (2012):** Evaluation of Serological Diagnostic Tests for Typhoid Fever in Papua New Guinea Using a Composite Reference Standard. *Clin Vaccine Immunol.*; 19(11):1833-7.
9. **Ley B, Thriemer K, Ame SM, Mtove GM, Seidlein LV, Amos B, Ilse CE, Hendriksen, Mwambuli A, Aikande Shoo A, Deok R Kim DR, Leon R, Ochiai LR, Favorov M, Clemens JD, Wilfing H, Deen JL and Ali SM (2011):** Assessment and comparative analysis of a rapid diagnostic test Tubex(R) for the diagnosis of typhoid fever among hospitalized children in rural Tanzania. *BMC Infect Dis.*;11:147.
10. **Anagha K, Deepika B, Shahriar R and Sanjeev K (2012):** The Easy and Early Diagnosis of Typhoid Fever. *Journal of Clinical and Diagnostic Research.* 6(2): 198-199.
11. **Collee JG, Duguid JP, Fraser AG, Marmion BP, and Simmons A (1996):** Laboratory strategy in the diagnosis of infective syndromes. In Mackie and

- McCartney Practical Medical Microbiology, Colle JG, Fraser AG, Marmion BP and Simmons A, 14th edition, New York, Churchill Livingstone, p53-94.
12. **Ministry of Health and Population, Egypt:** Enhanced Surveillance for Communicable Diseases. Annual Summary January – December 2000. Introduction. [cited 2008 Feb 16]. Available from: <http://www.geis.fhp.osd.mil/GEIS/Training/EgyptSurv2000.htm>
13. **Anusha R, Ganesh R and Lalitha J (2011):** Comparison of a rapid commercial test, Enterocheck WB (®), with automated blood culture for diagnosis of typhoid fever. *Ann Trop Paediatr*; 31(3): 231-234.
14. **Sherwal BL, Dhamija RK, Randhawa VS, Jais M, Kaintura A and Kumar M (2004):** A Comparative Study of Typhidot and Widal Test in Patients of Typhoid Fever. *Infect Immun.*; 5(3): 244-6
15. **Bakr W, El Attar L, Ashour M and El Toukhy A (2011):** The dilemma of widal test- which brand to use? A study of four different widal brands: a cross sectional comparative study. *Ann Clin Microbiol Antimicrob.* 8;10:7.
16. **Fadeel M, House B, Wasfy M, Klena J, Habshy E, Said M, Maksoud M, Rahman B and Pimentel G (2012):** Evaluation of a newly developed ELISA against Widal, TUBEX-TF and Typhidot for typhoid fever surveillance. *J Infect Dev Ctries.* 21;5(3):169-75
17. **Rahman M, Siddique A K, Tam FCH, Sharmin S, Rashid H and Iqbal A (2007):** Rapid detection of early typhoid fever in endemic community children by the Tubex O9-antibody test. *Dig Microbiol Infect Dis*, 58, 275-81.
18. **Dutta S, Sur D, Manna B, Sen B, Deb AK, Deen JL, Wain J, Von Seidlein L, Ochiai L, Clemens JD and Kumar Bhattacharya S (2006):** Evaluation of new generation serologic test for the diagnosis of typhoid fever. *Diag Microbiol and Infect Dis*, 56, 359-65.
19. **Wain J and Hosoglu S (2008):** The laboratory diagnosis of enteric fever. *J Infect Developing Countries* 2008; 2(6):421-425.
20. **Olsen SJ, Pruckler J, Bibb W, Thanh NT, Trinh T, Minh NT, Sivapalasingam S, Gupta A, Phuong P, Chinh NT, Chau NV, Cam PD, and Mintz ED (2004):** Evaluation of Rapid Diagnostic Tests for Typhoid Fever. *J Clin Microbiol.*; 42(5):1885-9.
21. **Caumes E, Ehya N, Nguyen J, Bricaire F, Caumes E and Ehya N (2001):** Typhoid and paratyphoid fever: a 10-year retrospective study of 41 cases in a Parisian hospital. *J Travel Med.*; 8(6):293-7.

تقييم اختبار الانتروشك في تشخيص المصريين البالغين المصابين بحمى التيفويد

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المقدمة: حمى التيفويد من الأمراض الخطيرة ذات الاعراض الاكلينيكية الغير مميزة. يتأكد تشخيص حمى التيفويد باجراء المزارع الدمويه للدم و البراز بجانب اختبار الفيديال. لان التأكيد الميكروبيولوجي يستغرق الكثير من الوقت و عدم القدرة على الاعتماد على نتائج اختبار الفيديال في المناطق المتوطنه بالنسبه لحمى التيفويد، فقد أصبح من الضروري التوصل لاختبارات سريعة.

الهدف من البحث: المقارنه بين اختبار الانتروشك و اختبار الفيديال في تشخيص حمى التيفويد باستخدام مزرعة الدم كاختبار معيارى .

المرضى و طرق البحث: تم اختبار عينات الدم التي تم جمعها من 104 مريض متوقع اكلينيكيًا تشخيصه كحالة من حالات الإصابة حمى التيفويد باستخدام المزارع الدمويه، اختبار الفيديال، و اختبار الانتروشك. كما تم اختبار 46 عينة دم من المتطوعين الأصحاء باستخدام اختبار الفيديال، و اختبار الانتروشك.

النتائج: اظهر اختبار الانتروشك قيم اعلى من حيث درجة الحساسية 86% و نسبة التخصص الايجابية 89% بالمقارنه مع اختبار الفيديال الذي سجل درجة حساسيه 81% و نسبة تخصص ايجابية 71%. كما كانت درجة حساسية اختبار الانتروشك اعلى في حالات الحمى التي مر عليها اقل من 5 أيام (100%).

الخلاصة: يعتبر اختبار الانتروشك احد الاختبارات السيرولوجيه السريعه و السهله الاستخدام للتشخيص المبكر لحالات حمى التيفويد خاصة في مراكز الرعاية الأولية، العيادات الخارجية الملحقه بالمستشفيات بالاضافه الى المستشفيات.

The Easy and Early Diagnosis of Typhoid Fever

KINIKAR ANAGHA, BHALERAO DEEPIKA, ROUSHANI SHAHRIAR, KULKARNI SANJEEV

ABSTRACT

Background and Objective: Typhoid fever is endemic in India. Its diagnosis is usually confirmed by blood culture, clot culture, stool culture and the Widal agglutination test. In the present study, we assessed the usefulness of the Enterocheck WB test in detecting the IgM antibodies in typhoid fever.

Materials and Methods: A total of 83 cases of clinically suspected typhoid fever were included in the study. A blood culture and the Widal test were done by using the standard methods. The Enterocheck WB test was done according to the manufacturer's instructions.

Results: Of the 83 cases, the blood culture was positive for *Salmonella typhi* in 19 (22.89%) cases, among which the Widal test was positive in 12 (14.45%) cases and the Enterocheck

WB test was positive in 17 (20.48%) cases. The sensitivity & specificity of the Enterocheck WB test were found to be 89.47% and 96.87% respectively.

Interpretation and Conclusion: We compared the sensitivity and the specificity of the Enterocheck WB test and the Widal test to those of the blood culture as the gold standard method in the diagnosis of typhoid fever. We found that the Enterocheck WB test for the detection of the IgM antibodies was easy to perform and that no special equipment or prior training of the para-medical staff were required for the testing and the interpretation of the results. Hence, it can be concluded that the Enterocheck WB test can be used as a complimentary test as compared to the blood culture and the Widal test for the diagnosis of typhoid fever.

Key Words: *S. typhi*, Blood culture, Typhoid fever, Widal test, Enterocheck WB test, Early diagnosis

INTRODUCTION

Typhoid fever is a systemic infection which is caused by the bacterium, *Salmonella enterica*, serotype typhi. This highly adapted, human specific pathogen has evolved remarkable mechanisms for its persistence in its host that help the organism to ensure its survival and transmission [1]. The socio-economic impact of the disease is huge, because the typhoid survivors may take several months to recover and to resume work [2]. An early and accurate diagnosis is necessary for a prompt and effective treatment. One has to rely on serological diagnosis, since many diagnostic laboratories in the developing countries, especially in the rural areas, do not have facilities for blood culture and thus the serological diagnosis becomes an important diagnostic tool. The Widal agglutination test usually detects the IgM and the IgG antibodies to *S. typhi* in the patient's serum from the second week of the onset of the symptoms of typhoid fever. The early rising antibodies to the lipopolysaccharide (LPS) O are predominantly IgM in nature. The *S. typhi* specific IgM antibodies can be used as an early marker to detect a recent infection. So, this study was undertaken to evaluate the sensitivity and the specificity of the Enterocheck WB test for the diagnosis of typhoid fever.

MATERIALS AND METHODS

This study was conducted in Department of Microbiology, Rural Medical College, Loni, India during January 2010- July 2011. The study included patients with fever for > four days and symptoms and signs which were suggestive of typhoid fever. An informed consent was taken from the patients before the sample collection. The blood samples were collected for blood culture, Widal test and the Enterocheck WB test. The blood culture was done by using

brain heart infusion (BHI) broth and it was incubated. Subsequent sub-cultures were made on Mac Conkey's agar and blood agar medias after 24, 48 and 96 hours and the final sub-culture was made on the seventh day. The growth of *S. typhi* was identified by the standard biochemical tests and it was confirmed by agglutination with the *Salmonella* polyvalent 'O', 09 and the H:d antisera. The Widal test was done by using the standard procedure [3]. The Widal test was confirmed by the tube agglutination method and it was considered as positive when a titer of equal to or more than 1:160 was observed. The Enterocheck WB test (manufactured by Zephyr Biomedicals) is a rapid, qualitative sandwich immunoassay for the detection of the IgM antibodies to *S. typhi* in human serum / plasma or whole blood specimens. It detects the presence of the IgM class of antibodies to a lipopolysaccharide (LPS) which is specific to *S. typhi* in the specimens.

RESULTS

Among the 83 cases, *S. typhi* was isolated from the blood culture in 19 cases (22.89%) and the remaining 64 cases were blood culture negative. The Widal test was positive in 12 cases (14.45%) and the Enterocheck WB test was positive in 17 cases (20.48%). The sensitivity and the specificity of the Enterocheck WB® test and the Widal test were compared [Table/Fig-1].

DISCUSSION

In the present study, the sensitivity & specificity of the Enterocheck WB test were found to be 89.47% and 96.87% respectively. This was comparable to those of other studies for the rapid diagnosis of typhoid fever. D. Narayanappa et al reported the sensitivity of Typhidot – M as 92.6 per cent [4]. Mary Jesudason

Blood Culture	Enterocheck WB		Widal Test	
	Positive	Negative	Positive	Negative
Positive (n=19)	17	2	12	7
Negative (n=64)	2	62	22	40

[Table/Fig-1]: Comparison of results of Enterocheck WB and Widal test with blood culture

*Sensitivity of Enterocheck WB - 89.47%
 †Specificity of Enterocheck WB - 96.87%
 ‡Sensitivity of Widal test - 63.15%
 §Specificity of Widal test - 62.5%

et al reported Typhidot with 100% sensitivity and 80% specificity when bacteraemic patients were analyzed. A further prospective evaluation of the same test by this author on a large sample size showed better results [5,6]. Sonja J Olsen et al reported the sensitivity of the Tubex test to be 78% and its specificity to be 89% [7]. Anush R compared the Enterocheck WB® test to automated blood culture for typhoid fever and found its sensitivity and specificity as 85.5% and 88.6% respectively [8].

Our study also focused on the utility of the Widal test. In the present study, its sensitivity (63.15%) and specificity (62.5%) were satisfactory. But R Duthie et al reported a higher sensitivity (78%) and specificity (99%) of the Widal test [9]. In a developing country like India, the Widal test has been used extensively in the serodiagnosis of typhoid fever. However, Lateef A et al reviewed the significance of the Widal agglutination test and concluded that its use should not be encouraged in endemic areas [10]. Ideally, in the Widal test, a fourfold rise of the antibody titer in paired sera is considered as diagnostic of typhoid fever. However, paired sera are often difficult to obtain and as a specific chemotherapy, it has to be instituted on the basis of a single Widal test only. Kulkarni M et al and Rasaily R et al revealed that a single Widal test, in association with relevant clinical findings, can still be used as a useful diagnostic tool for typhoid fever, [11,12].

CONCLUSION

The Widal test showed 63.15% sensitivity and 62.5% specificity in the blood culture positive cases of typhoid fever. The Enterocheck WB test, with its higher sensitivity and specificity, can be effectively

used in the rural set-up, wherein blood culture facilities are not available. The early detection of the IgM antibodies by the Enterocheck WB test will serve as a marker of recent infection. The Widal test can be used as a complimentary serological diagnostic tool as and when it is required. However, the importance of the blood culture in the typhoid cases cannot be ignored, as the antibiotic susceptibility testing of the isolated strains is equally important.

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REFERENCES

- [1] Parry CM, Hien TT, Dougan T, White NJ, Farrar JJ. Typhoid Fever. *The New England Journal of Medicine* Nov 2002; 347(22): 22 1770-81.
- [2] Park K. Textbook of Preventive and Social Medicine. In Chapter 5 Epidemiology of communicable diseases. 21st Edition. *Banarsidas Bhanot Publishers, Jabalpur*. 2011; 212-16.
- [3] Collee JG, Digid JP, Fraser A G. Mackie and McCartney Practical Medical Microbiology. 14th Edition Churchill Livingstone, Edinburgh, 1996.
- [4] Narayanappa D, Sripathi R, Jagdishkumar K, Rajani HS. Comparative study of the dot enzyme immunoassay (Typhidot-M) and the Widal test in the diagnosis of typhoid fever. *Indian Paediatrics*. 2010; 47: 331-33.
- [5] Jesudason M, Esther E, Mathai E. The Typhidot test to detect the IgM and IgM antibodies in typhoid fever. *Indian Journal Medical Research* 2002; 116: 70-72.
- [6] Jesudason M, Sivkumar S. Prospective evaluation of a rapid diagnostic test, Typhidot ©, for typhoid fever. *Indian Journal Medical Research*; 2006; 123: 513-16.
- [7] Oslon SJ, Pruckler J, Bibb W, Nguyen TMT, Tran MT, Sivapalasingam S, et al. Evaluation of the rapid diagnostic tests for typhoid fever. *Journal of Clinical Microbiology*. 2004; 42(5):1885-89.
- [8] Anush R, Ganesh R, Lalitha J . Comparison of the rapid commercial test and the Enterocheck WB® test to automated blood culture for the diagnosis of typhoid fever. *Annals of Tropical Paediatrics*.2011; 31(3):231-34.
- [9] Duthie R, French GL. Comparison of the methods for the diagnosis of typhoid fever. *Journal of Clinical Pathology*.1990; 43:863- 65.
- [10] Lateef AO, King AL. The Widal agglutination test-100 years later: still plagued by controversy. *Postgraduate Medical Journal* 2000;76: 80-84.
- [11] Kulkarni ML, Rego SJ. Value of a single Widal test in the diagnosis of typhoid fever. *Indian Paediatrics* 1994; 31:1373-77.
- [12] Rasaily R, Dutta P, Saha MR, Mitra U, Bhattacharya SK, Manna B, et al. Value of a single Widal test in the diagnosis of typhoid fever. *Indian Journal Medical Research*.1993; 97: 104-07.

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Evaluation of Enterocheck WB - a rapid test for diagnosis of typhoid fever

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Abstract: Blood culture although continues to be the gold standard for diagnosis of typhoid fever, its applications for routine diagnosis of typhoid fever is restricted because of several limitations. Widal test has been used extensively as a laboratory tool for diagnosis of typhoid fever in most laboratories, but it is laborious, time consuming and may not be positive in early stages and is to be interpreted judiciously. In the recent times, several other serological tests other than Widal test have been described and evaluated. In the present study, we evaluated Enterocheck WB, an immunochromatographic rapid test for diagnosis of typhoid fever, in Widal positive cases. This test detected IgM antibodies and was found to have a sensitivity of 90% and a specificity of 94.6%. The positive and negative predictive values of Enterocheck WB was 84.9% and 96.5% respectively. The test serves as an useful alternative to other conventional tests for rapid diagnosis of typhoid fever.

Key words: Enteric fever, Enterocheck, Rapid test, Typhoid fever

I. Introduction

Typhoid fever continues to be prevalent in several countries around the world. It poses an important public health problem in many parts of India where it is endemic, accounting for considerable morbidity and mortality [1]. Incidence of typhoid fever has been estimated as approximately 17 million cases; with 600000 associated deaths occurring annually [2-3]. The disease presents a clinical dilemma with varied manifestations, closely resembling several other clinical conditions presenting with febrile illnesses, such as vector borne malaria, dengue fever and rickettsiosis as well as environmentally transmitted leptospirosis and melioidosis [4-8]. According to World Health Organization (WHO) a confirmed case of typhoid fever is defined as a patient with fever ($>38^{\circ}$) that has lasted for at least three days, with a laboratory confirmed positive culture of *S. typhi* [9]. Probable case of typhoid fever is a patient with fever ($>38^{\circ}$) that has lasted for 3 days, with positive serodiagnosis or antigen detection test but without *S. typhi* isolation [9]. A chronic carrier is determined by excretion of *S. typhi* in stools or urine for longer than one year after the onset of acute typhoid fever.

Laboratory diagnosis of typhoid fever is based on isolation of bacilli from patient's blood and demonstration of antibodies in the serum. Blood culture and Widal test are the routinely employed investigations for diagnosis of typhoid fever in clinical settings. Blood culture although confirmatory, requires cultural processing and identification of the causative agent, which is labor intensive and relatively costly. Widal test has been used in diagnosis of typhoid illness for a long time in the country, but it remains a serological test with moderate sensitivity and specificity [10]. There is a need for rapid, reliable and easy to perform test with high degree of sensitivity and specificity, which will assist the clinicians in early diagnosis and prompt treatment.

With this objective, we evaluated the efficiency of Enterocheck WB immunochromatographic card test as a predictor of typhoid fever and compared its performance with standard culture and Widal test.

II. Materials and methods

This study was conducted in the Department of Microbiology, Goa Medical College, Bambolim Goa. Widal tube agglutination test was done using standard stained antigens supplied by Tulip Diagnostics (P) Ltd Bambolim Goa, on all clinically suspected cases of enteric fever. The clinical response to specific therapy for typhoid fever in all cases was also noted.

Fifty suspected cases of typhoid fever which were Widal positive were subjected to immunochromatographic card test (Enterocheck WB) supplied by Zephyr Biomedicals, Verna Goa.

Enterocheck WB is a rapid qualitative immunochromatographic sandwich immunoassay for detection of IgM antibodies to *S. typhi* in serum / plasma / whole blood. It detects antibodies specifically directed against lipopolysaccharide (LPS) of *S. typhi*. The test employs nitrocellulose membrane conjugate pad containing anti human IgM conjugated to colloid gold and rabbit IgG conjugated to colloid gold. In positive samples the *S. typhi* specific serum IgM forms complex with anti IgM colloid gold and then moves along the membrane to the test region where it is immobilized by the *S. typhi* specific LPS antigen coated on the membrane, giving a pink

purple band. In negative cases only the rabbit IgG gold conjugate moves further on the membrane to the control region where it is immobilized by anti rabbit antibodies, giving a pink purple band. (see figure 1)

This test was performed as per the technical instructions provided by the manufacturer, using 5 μ l of patient's serum and charging it into the sample port 'A' with the loop provided with the kit. 5 drops of sample running buffer was later added to the port 'B'. The test was readable after 15 minutes.

Blood cultures were also done on all the 50 cases, simultaneously using Glucose Broth and Hartley's broth employing standard culture techniques.

Samples from hundred antenatal women attending the antenatal clinics of this Institution and fifty healthy blood donors were taken as controls and their serum samples were subjected to Enterocheck WB and Widal test.

III. Results

Amongst 50 suspected cases of typhoid fever, blood culture was positive in only 8 (16%) cases. Widal test was found to be positive in all 50 cases, while Enterocheck WB was positive in 45 cases (90%) only. Widal test detected five more positive cases as compared to Enterocheck WB, but none of the Widal test negative cases were positive by Enterocheck WB. In the control groups, Enterocheck WB was positive in 5 (5%) antenatal samples and 3 (6%) blood donors' samples. Similarly, Widal test was positive in 5 (5%) antenatal and 3 (6%) blood donors' samples (Refer Table 1).

Considering Widal positivity and clinical response to therapy in suspected cases as a marker of typhoid fever, all Widal positive cases that responded to therapy, were considered as probable cases of typhoid fever. We evaluated usefulness of Enterocheck WB for diagnosis of typhoid fever based on this parameter.

The sensitivity and specificity of Enterocheck was found to be 90% and 94.6% respectively. Further, the positive predictive value for Enterocheck WB was 84.9%, while negative predictive value was 96.5%. The Enterocheck WB showed an acceptable positive likelihood ratio of 16.6 (acceptable limit > 10), and an acceptable negative likelihood ratio of 0.10 (acceptable limit < 1).

IV. Discussion

Typhoid fever remains an important public health problem in developing countries. The definitive diagnosis is based on the isolation of the causative agent *Salmonella typhi* from blood, faeces, urine or other clinical samples. Although blood culture remains the gold standard test in diagnosis of typhoid fever, its utility in diagnosis is restricted to early phase of illness and is not useful for rapid diagnosis [10].

Although blood culture is considered as gold standard for diagnosis of typhoid fever, in the present study, only 8 out of 50 (16%) suspected cases were culture positive. This could be attributed to the fact that most of these patients were presenting after first week of illness and had received some antibiotic therapy before being referred to tertiary care centre.

In developing countries, facilities for isolation and culture are often not available, especially in smaller hospitals and diagnosis relies upon the clinical features of the disease and agglutinating antibodies to *S. typhi* [11]. Widal test has been used very extensively in serodiagnosis of typhoid fever and in developing countries particularly, remains the only practical test available [11]. However interpretation of Widal test is difficult in areas where *S. typhi* is endemic when basal titre of the population is not known. Further, in areas where fever due to infection is a common occurrence, the possibility exists that false positive reactions may occur as a result of non-typhoidal fevers [11].

There is a need for a rapid and reliable diagnostic test to overcome these problems. Recently, numerous tests other than culture isolation and Widal test are made available to support the diagnosis of typhoid fever. These include amongst others, Multi-test Dip-S-Ticks, TyphiDot and TUBEX to detect IgG, IgG and IgM and IgM, respectively [12, 13]. TyphiDot, an ELISA based assay in dot test format has been tried in Malaysia, Singapore, Pakistan and India with satisfactory results [14]. However, some of them are either expensive or labor intensive.

In the present study, we evaluated the usefulness of Enterocheck WB in cases where presumptive diagnosis of typhoid fever was based on clinical presentation, a positive Widal test and response to therapy. The test had a high positive predictive value of 84.9% and a negative predictive value of 96.5%, besides having sensitivity and specificity of 90% and 94.6% respectively. The test was comparable to Widal test, but had a superior interpretative value, as it detected IgM antibodies denoting recency of infection. The other advantage of this test was that it is less laborious, rapid and results are obtained within 15 minutes. This will help the clinicians to initiate antimicrobial therapy at an early stage of the disease, thereby decreasing morbidity.

The test is however designed to detect only cases of enteric fever caused by *S. typhi* and not those caused by *S. paratyphi* A, B or C. The test can well serve as a substitute to Widal test, which is time consuming and laborious. Further evaluations need to be done to ascertain its comparative value with other rapid tests available commercially.

V. Figures and tables

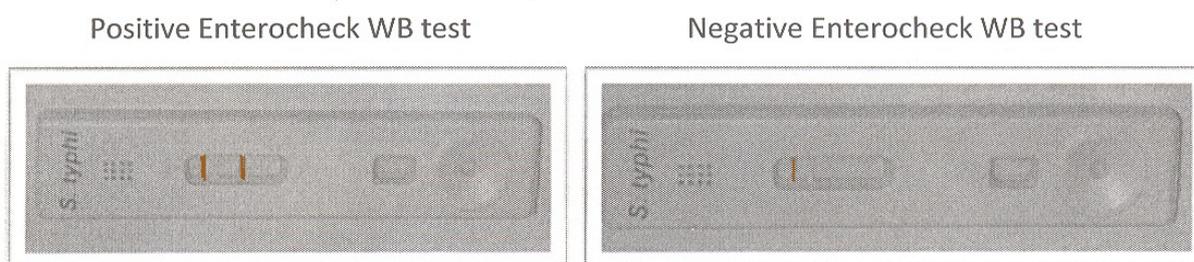


Figure1. Positive and negative Enterocheck WB test

Table 1:
Result of Enterocheck WB, Widal and Blood Culture

Category	Enterocheck WB Positive	Enterocheck WB Negative	Widal Positive	Widal Negative	Blood Culture Positive	Blood Culture Negative
Suspected typhoid cases (n= 50)	45	5	50	-	8	42
Blood Bank donors (n=50)	3	47	3	47	-	-
Antenatal cases (n=100)	5	95	5	95	-	-
Total (n=200)	53	147	58	142	8	42

VI. Conclusion

Diagnosis of enteric fever often comes under scrutiny, as blood cultures may not be rewarding specially when patient is partially treated or when the patient presents a week or later after onset of acute illness. Widal test which is commonly done for diagnosis of enteric fever serves as the widely used test especially in developing countries, although it has a moderate sensitivity. Hence there is a need to opt for any better test that would give reliable test results.

In the past many rapid tests like Multi-test Dip-S-Ticks, TyphiDot an ELISA based assay in dot test format and TUBEX to detect IgG, IgG and IgM and IgM, respectively have been tried with fairly satisfactory results. In the current, we compared Enterocheck WB rapid test for diagnosis of typhoid fever with blood cultures and widal test and found it to be comparable to widal test. More over this test detects IgM antibodies and is indigenously prepared. This test should be subjected to extensive use to further ascertain its diagnostic utility.

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References

- [1] Bhaskaram P, Sahay BK, Rao NSP. Specific immune responses in typhoid fever and after TAB vaccination. *Indian J Med Res* 1990; 91: 115-118.
- [2] Eelman R, Levine MM. Summary of an international workshop on typhoid fever. *Rev. Infect Dis* 1986; 8:329-47.
- [3] Ivanoff BN, Levine MM, Lambert PH. Vaccination against typhoid fever: present status. *Bull world Health Organ.* 1994; 72: 957-71.
- [4] Choudhury DS. Malaria in India: Past, present and future. *Indian J Paediatr* 1985; 52: 243-8.
- [5] Jacob S. Dengue: Emergence as a global public health problem and prospectus for control. *Trans R Soc Trop Med Hyg* 200; 94: 7-8.
- [6] Mathai E, Lloyd G, Cherian T, Abraham OC, Cherian AM. Serological evidence for continued presence of human Rickettsiosis in Southern India. *Ann Trop Med Parasitol* 2001; 95: 395-8.
- [7] Bharati AR, Nally JE, Ricaldi JN, Matthais MA, Diaz MM, Lovett MA, et al. Leptospirosis: a zoonotic disease of global importance. *Lancet Infect Dis* 2003; 3:757-71.
- [8] Jesudason MV, Anbarasu A, John TJ. Septicaemic meliodosis in a tertiary care hospital in south India. *Indian J Med Res* 2003; 117: 119-21.
- [9] WHO. Background document: The diagnosis, treatment and prevention of typhoid fever. WHO/V&B/03.07. 2003 Page 4.
- [10] Sherwal BL, Dhamija RK, Randhawa VS, Jais M, Kaintura A, Kumar M. A comparative study of typhiDot and Widal test in patients of Typhoid fever. *J Indian Acad Clin Med* 2004; 5(3): 244-6.

- [11] Pang T, Puthucheary SD. Significance and value of Widal test in the diagnosis of typhoid fever in an endemic area. *J Clin Pathol* 1983; 36:471-475.
- [12] Oslen SJ, Pruckler J, Bibb W, Thanh NTM, et al. Evaluation of rapid diagnostic tests for typhoid fever. *J Clin Microbiol* 2004; 42(5): 1885-1889.
- [13] Keddy KH, Sooka A, Letsoalo ME, et al. Evaluation Sensitivity and specificity of typhoid fever rapid antibody tests for laboratory diagnosis at two sub-Saharan African sites. *Organ* 2011; 89: 640-7
- [14] Jesudason MV, Sivakumar S. Prospective evaluation of a rapid diagnostic test Typhidot for typhoid fever. *Indian J Med Res* 2006;123: 513-517.

A comparative clinical study of efficacy of microimmuno assay with WIDAL-test in enteric fever in children

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Abstract

The diagnosis of typhoid fever in young children is also a dilemma because of its manifestations and typical presentation may not be seen in all cases. Antibodies to *Salmonella typhi* antigen are developed in the human body, which can be detected as a diagnostic test for the enteric fever. **Objective:** This study was undertaken to compare the efficacy of WIDAL-test with micro-immunoassay (dot enzyme immunosorbent assay). **Method:** 40 cases of clinically suspected enteric fever cases were included in this study. **Result:** In the present study, nearly 92% were positive for micro immunoassay (dot-enzyme immunosorbent assay) by Enterocheck-WB kit, 80% were positive for WIDAL and only 15% were culture positive. Immunoassay positive, but WIDAL negative cases were 20%, whereas WIDAL positive and immunoassay negative cases were only 7.5%. The positive predictivity of micro-immunoassay in diagnosing enteric fever is better than WIDAL both in 1st and 2nd week of illness. Micro-immunoassay done in the study was rapid in diagnosing the case. **Conclusion:** It is concluded from the present study that the micro-immunoassay (Enterocheck-WB) is better than WIDAL-test in the diagnosis of enteric fever in children.

Key words: Enteric fever, Enterocheck-WB kit, micro-immunoassay, WIDAL-test

INTRODUCTION

Enteric fever or typhoid fever is a severe systemic disease that is found mainly in developing countries, but it is encountered world-wide because of international travel. The diagnosis of typhoid fever in young children is also a dilemma because of its manifestations and typical presentation may not be seen in all cases. Isolation of *Salmonella typhi* organisms either by blood culture or cultures of bone marrow, stool and urine or bile juice is considered as the gold standard. Due to parent's urgency and enthusiastic doctors, children receive antibiotics prior to diagnosis which makes the isolation of the *S. typhi* organisms difficult. Antibodies

to *S. typhi* antigen are developed in the human body, which can be detected as a diagnostic test for the enteric fever.

In WIDAL-test the agglutination titer will depend on the stage of disease. Agglutinins will usually appear by the end of the 1st week, so that blood taken earlier may give a negative result. The titer increases steadily until the third or the 4th week, after which it declines gradually.^[1] Complexity and higher costs of molecular tests hinder its application in routine use.^[2]

A case or carrier is infectious as long as bacilli appears in the stool or urine.^[3] Because of intermittent and low level bacteremia, repeated blood cultures should be obtained in suspected cases.^[4] Complications can be reduced with timely accurate diagnosis and with adequate chemotherapy^[5,6] Clinical suspicion is necessary before the laboratory assistance can be mobilized.^[7]

This study was undertaken to compare the efficacy of WIDAL-test with micro-immunoassay (dot enzyme immunosorbent assay).

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Objective

The objective of this study is to compare the micro immunoassay-IgM detection (dot enzyme immunosorbent assay) with WIDAL-test.

MATERIALS AND METHODS

Source of data

The present study was conducted at Kempegowda Institute of Medical Sciences Hospital over a period of 1½ year (June 2009 to December 2010). In this prospective clinical study, suspected cases of enteric fever up to fifteen were admitted to the pediatric ward.

Inclusion criteria

Clinically suspected cases of enteric fever up to the age of 15 years.

Exclusion criteria

1. Patients with enteric fever above the age of 15 years.
2. Patients who have other associated conditions.

On admission, these subjects were enrolled in to the study. A detailed history was taken, clinical examination was done. A detailed questionnaire was recorded as per the proforma. These subjects were investigated for Hb%, total count, differential count, erythrocyte sedimentation rate, blood culture, WIDAL-test, micro immunoassay-IgM detection, stool routine, urine routine.

Blood sample for culture was drawn and inoculated in to blood culture bottles (before starting antibiotics) and sent to the laboratory, where the culture was done using biphasic brain heart infusion agar.

WIDAL-test by slide and tube agglutination technique, which was expressed in dilution titers of 1/20, 1/40, 1/80, 1/160, 1/320 and 1/640. Another serological test, micro immunoassay (dot enzyme immunosorbent assay) was done with Enterocheck-WB kit to detect IgM class of antibodies directed to lipopolysaccharide (LPS) 'O' of *S. typhi* organism and expressed as present or absent.

The dot-enzyme immunoassay (EIA) is a relatively newer serologic test based upon the presence of specific IgG and IgM antibodies to a specific 50-kD outer membrane protein (OMP) antigen on the *S. typhi* strain. The test incorporates nitrocellulose strips impregnated with the OMP antigen and separately identifies IgG and IgM antibodies.^[1]

The dot enzyme immunosorbent assay allows separate visual assessment of the presence of specific IgG and IgM antibodies to the OMP in a standard 1:100 dilution of serum by a characteristic color change and has been reported to be at least as specific and sensitive as WIDAL-test in children with typhoid fever.^[8] The cases, which were negative for blood cultures, WIDAL and micro immunoassay (dot-enzyme immunosorbent assay by Enterocheck-WB) even in the 2nd week of illness, were excluded from the study.

Sample size: 40 cases.

RESULTS

In the present study, fever was the symptom present in all the cases (100%).

In the present study, 11 cases out of 40 cases (27.5%) presented in the 1st week, 28 cases presented in the 2nd week (70%), only one case presented in the 3rd week of fever (2.5%).

Vomiting was noticed in 11 cases (27.5%).

Pain abdomen was the symptom in 15% of the cases.

Loose stools were present in 2 cases (5%).

Cough was present in 7 cases i.e., 17.5%.

In the present study 37 cases out of 40 (92%) were positive for micro immunoassay by Enterocheck-WB kit, 32 cases (80%) were positive for WIDAL and 6 cases (15%) were culture positive. Out of 11 cases admitted in the 1st week of fever immunoassay + and WIDAL – were 6 cases, immunoassay + and WIDAL + were 3 cases, immunoassay – and WIDAL + were 2 cases, blood culture + and immunoassay + were 5 cases, blood culture + WIDAL + were 2 cases, blood culture +, WIDAL + and immunoassay + were 2 cases. Out of 28 cases admitted in the 2nd week of fever immunoassay + and WIDAL – were 2 cases, immunoassay + and WIDAL + were 25 cases, immunoassay – and WIDAL + were 1 case, blood culture + and immunoassay + were 1 case, blood culture + WIDAL + were 1 case, blood culture +, WIDAL + and immunoassay + were 1 case as shown in Graph 1 and Graph 2.

Only one case was admitted in the 3rd week of fever, where both immunoassay and WIDAL was positive.

Table 1 shows, in the present study, out of 11 cases presented in the 1st week of fever: 6 cases were positive by immunoassay and were negative for WIDAL, 2 cases were WIDAL positive an immunoassay negative and in 3 cases both immunoassay and WIDAL were positive. Out of 28 cases presented in the 2nd week of fever: 2 cases

were positive by immuno assay and were negative for WIDAL, 1 case was WIDAL positive and immunoassay negative and in 25 cases both immunoassay and WIDAL were positive. Only one case presented in the 3rd week of fever that was positive by both WIDAL and micro immunoassay.

The sensitivity of WIDAL-test is 78.4% and the sensitivity of micro immunoassay is 90.6% in the present study [Table 2 and Graph 3].

DISCUSSION

In the present study, 37 cases out of 40 (92%) were positive for micro immunoassay (dot-enzyme immunosorbent assay) by Enterocheck-WB kit, 32 cases (80%) were positive for WIDAL and 6 cases (15%) were culture positive. Zulfiqar Ahmed Bhutta *et al.* reported 52 cases out of 71 (73%) in the entire cohort and 39 cases out of 46 cases (84%) among culture proven typhoid fever cases were positive by EIA (Typhidot-M). 39 cases (54%) in the entire cohort and 29 (63%) cases among culture proven typhoid fever cases were positive for WIDAL-test.^[8]

Deborah House *et al.* reported serological assays based on the detection of IgM antibodies against either serotype Typhi LPS (ELISA) or whole bacteria (dipstick) had a significantly higher sensitivity than the WIDAL TO test when used with a single acute phase serum sample

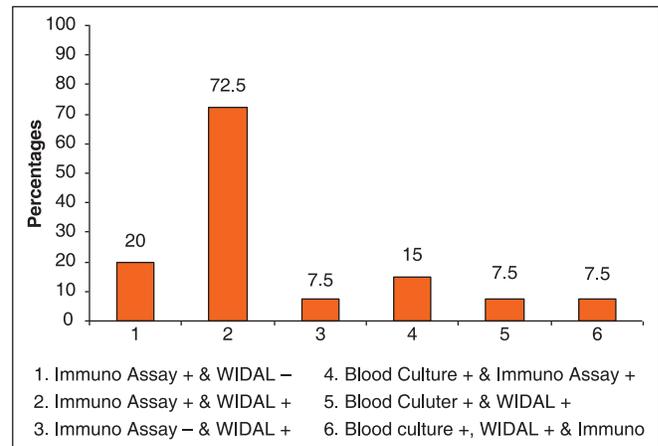
Table 1: Comparison of investigation methods (WIDAL and micro immunoassay)^[9]

Duration of fever	Week 1	Week 2	Week 3	Total
Total number of cases	11	28	1	40
1. Immuno Assay + & WIDAL -	6 (54.5%)	2 (7.1%)	-	8 (20.0%)
2. Immuno Assay + & WIDAL +	3 (27.3%)	25 (89.3%)	1 (100.0%)	29 (72.5%)
3. Immuno Assay - & WIDAL +	2 (18.2%)	1 (3.6%)	-	3 (7.5%)

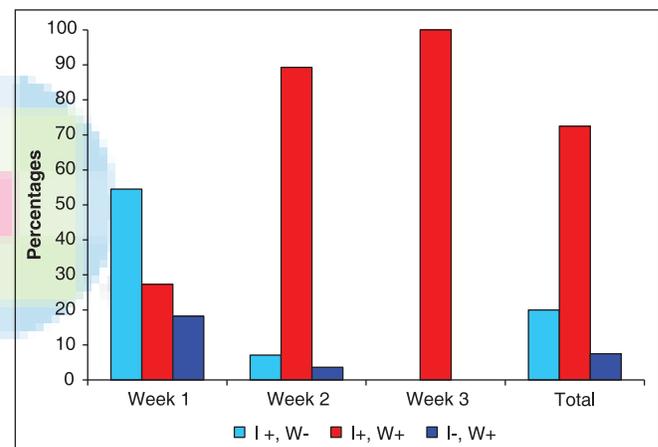
Table 2: Vise-versa comparison of positive with negative tests of WIDAL and micro immunoassay^[9]

Comparison of WIDAL and IMMUNO ASSAY	Number
WIDAL +: IMMUNO ASSAY +	29
WIDAL +: IMMUNO ASSAY -	3
WIDAL -: IMMUNO ASSAY +	8
WIDAL -: IMMUNO ASSAY -	0
Total	40
Sensitivity of WIDAL	78.4%
Sensitivity of IMMUNO ASSAY	90.6%

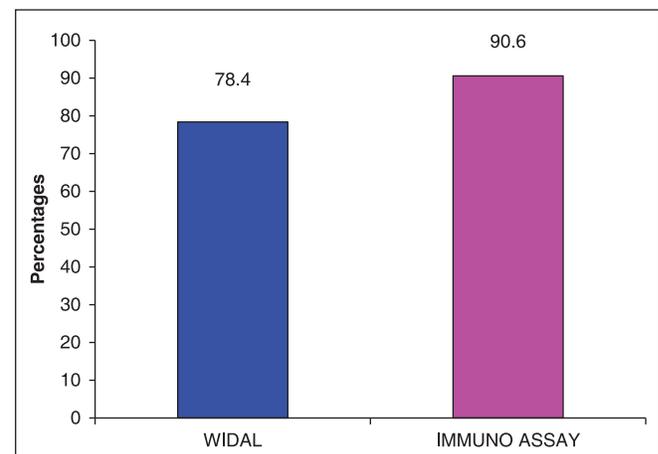
($p < 0.007$).^[10] Mary V Jesudasan *et al.* reported that detection of anti LPS antibodies by ELISA had a sensitivity of 89.4% and specificity of 94.9% in their study.^[11]



Graph 1: Comparison of investigation methods (WIDAL and micro immunoassay)^[9]



Graph 2: Vise-versa comparison of positive with negative tests of WIDAL and micro immunoassay^[9]



Graph 3: Comparison of sensitivity in % of WIDAL and micro immunoassay^[9]

CONCLUSION

In the present study, 92% were positive for micro immunoassay (dot-enzyme immunosorbent assay) by Enterocheck-WB kit, 80% were positive for WIDAL and only 15% were culture positive. Immunoassay positive but WIDAL negative cases were 20% whereas WIDAL positive and immunoassay negative cases were only 7.5%. The positive predictivity of micro-immunoassay in diagnosing enteric fever is better than WIDAL both in 1st and 2nd week of illness.

Its concluded from the present study that the micro immunoassay (by Enterocheck-WB) is better than WIDAL-test in the diagnosis of enteric fever in children. Micro-immunoassay done in the study was rapid in diagnosing the case, comparatively economical and can be done on the out-patient basis with a small quantity of serum or whole blood (5 µl).

REFERENCES

1. Balakrishna TP, Sumathi S, Anuradha K, Venkatesh D, Krishna S, *et al.* A comparative study of typhidot and Widal test in the diagnosis of typhoid fever. *J Evol Med Dent Sci* 2013;2:3721.
2. Pastoor R, Hatta M, Abdoel TH, Smits HL. Simple, rapid, and affordable point-of-care test for the serodiagnosis of typhoid fever. *Diagn Microbiol Infect Dis* 2008;61:129-34.

3. Park K. Park's Text Book of Preventive and Social Medicine. 18th ed. Jabalpur: M/S Banarsidas Bhanot; 2005. p. 195-8.
4. Behrman RE, Kliegman RM, Jenson HB. Nelson Text Book of Pediatrics. 17th ed. Philadelphia: WB Saunders Company; 2004. p. 916-8.
5. Collier L, Balow A, Sussman M, Duerden BI. Topley and Willson's Microbiology and Microbial Infections. Systemic Bacteriology. 9th ed. Vol. 2. London: Arnod; 1998. p. 969-95.
6. Rator MY. Typhoid fever. *Indian J Pract Doct* 2004;1:1.
7. Olsen SJ, Pruckler J, Bibb W, Nguyen TM, Tran MT, Nguyen TM, *et al.* Evaluation of rapid diagnostic tests for typhoid fever. *J Clin Microbiol* 2004;42:1885-9.
8. Sherwal BL, Dhamija RK, Radhawa VS, Jais M, Kaintura A, Kumar M. A comparative study of typhidot and Widal test in patients of typhoid fever. *J Indian Acad Clin Med* 2004;5:224-6.
9. Madhu GN. "A comparative clinical study of efficacy of short course ofloxacin therapy with ceftriaxone in uncomplicated enteric fever in children and efficacy of micro immuno assay with widal test." Unpublished data of Rajiv Gandhi University of Health Sciences (RGUHS).
10. Agrawal PK, Gogia A, Gupta RK. Typhoid fever. *J Indian Acad Clin Med* 2004;5:60-4.
11. Dutta S, Sur D, Manna B, Sen B, Deb AK, Deen JL, *et al.* Evaluation of new-generation serologic tests for the diagnosis of typhoid fever: Data from a community-based surveillance in Calcutta, India. *Diagn Microbiol Infect Dis* 2006;56:359-65.

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PubMed

Abstract

Ann Trop Paediatr. 2011;31(3):231-4. doi: 10.1179/1465328111Y.0000000030.

Comparison of a rapid commercial test, Enterocheck WB(®), with automated blood culture for diagnosis of typhoid fever.

Anusha R¹, Ganesh R, Lalitha J.

Author information

Abstract

BACKGROUND: Rapid diagnosis of typhoid fever is very important in the timely initiation of therapy.

OBJECTIVES: To evaluate the sensitivity and specificity of the Enterocheck WB(®) test.

METHODS: In a prospective study, the sensitivity and specificity of Enterocheck WB(®) that detects IgM antibodies to *Salmonella typhi* were compared with blood culture using BacT/Alert.

RESULTS: Four hundred and fifty patients with suspected typhoid fever were assessed both by blood culture and Enterocheck WB(®); 100 of them were either Enterocheck WB(®)-positive or blood culture-positive or both for *S. typhi*. Both tests were positive in 47 of the 100 patients. In 45 patients, Enterocheck WB(®) was positive but blood culture was negative. In eight cases, Enterocheck was negative but blood culture was positive. Using blood culture as the gold standard, the sensitivity of Enterocheck WB(®) was 85.5%, specificity 88.6%, negative predictive value 97.7% and positive predictive value 51.1%.

CONCLUSION: The Enterocheck WB(®) test can be performed at the bedside in 15 minutes and is a useful rapid method for diagnosis of typhoid fever.

PMID: 21781418 [PubMed - indexed for MEDLINE]

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Rapid Immunochromatographic Assay of IgM Antibodies to Salmonella Typhi in Diagnosis of Typhoid Fever in Adults /

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المؤلف

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Microbiology and Immunology

الفهرس



Introduction

I- INTRODUCTION

Typhoid fever is caused by Salmonella enteric serotype typhi (*S. typhi*) and remains a significant health problem in many developing countries. Estimates suggest an incidence rate of more than 21.5 million cases with 200,000 deaths worldwide annually. The estimated incidence of typhoid fever in Egypt ranges between 13 and 59 per 100,000 persons per year (*Crump et al; 2004 and Srikantiah et al; 2006*).

Enteric fever is a potentially severe systemic febrile illness that is usually presented with non-specific symptoms and signs such as slow gradually progressive fever, nausea, vomiting, abdominal pain, malaise, headache, constipation then diarrhea and hepatosplenomegally. It can be complicated by serious complication such as intestinal hemorrhage, intestinal perforation in the distal ileum, septicemia, diffuse peritonitis, encephalitis and cholecystitis (*Clark et al; 2010*).

As the clinical picture of typhoid fever usually is misdiagnosed, microbiologic culture of a blood sample is considered to be the gold test for the diagnosis of typhoid

1

from 121

Abstract

Enteric fever is a potentially severe systemic febrile illness that is usually presented with non-specific symptoms and signs such as slow gradually progressive fever, nausea, vomiting, abdominal pain, malaise, headache, constipation then bloody diarrhea and hepatosplenomegally (Clark et al; 2010).

Microbiologic culture of a blood sample is considered to be the golden test for the diagnosis of typhoid fever even though it takes up to seven days and requires a well equipped laboratory (Parry et al; 2002).

Serological diagnosis using Widal test provides a cost and time efficient alternative for blood culture, however its performance remains unsatisfying with sensitivity reported from Egypt of 77% using blood culture as a gold standard test and with the need for establishment of a local cut off titer prior to use which makes its interpretation more complicated (Wilke et al; 2002).

Assays that detect IgM antibodies to *S. Typhi* which develop early in acute typhoid fever suggesting current infection are more sensitive and specific than the Widal test, and can be performed more rapidly. In the absence of culture facilities, IgM antibody tests are more useful than Widal test in diagnosis of typhoid fever in endemic areas (Cheesbrough; 2006). The aim of the present study was to Evaluate the Enterocheck WB® test (a rapid, qualitative, Immunochromatographic assay for the detection of IgM antibodies to *S. typhi* in human serum/plasma or whole blood specimen), in comparison to Widal test as a rapid diagnostic tool for typhoid fever in adults.

The present study was conducted on 154 patients clinically suspected as typhoid and 46 healthy controls. The patients were divided according to blood culture results into 2 groups; Group I comprised 21/200 (14%) patients with positive blood culture for *S.typhi*. Group II included 133/200 (86%) patients with negative blood culture results. Blood samples were collected from the patients to perform blood cultures, Widal test and Enterocheck test as well as from controls to perform Widal test and Enterocheck test.

Blood culture showed only 21/200 positive cases (14%). When considering O and H titer ≥ 160 as positive, Widal test showed 59/200 (72.2%) positive cases, the sensitivity and specificity of Widal were 80.95% and 70.67% respectively.

Eighteen cases out of 200 representing 95% of the studied subjects were positive using Enterocheck test and the sensitivity and specificity of this test were 86% and 89%, respectively.

Comparing the two serological tests; Widal and Enterocheck, the present study found Enterocheck test to be more sensitive than Widal test for acute typhoid diagnosis. Furthermore, a clear advantage of the Enterocheck test is the rapid availability of the results which is essential in clinical work.

Performance Evaluations



ISO 9001: 2008
EN ISO 13485: 2012

OTHER EVALUATIONS

ENTEROCHECK[®]-WB

Rapid test for the detection of IgM antibodies to *S. typhi*



Tulip Diagnostics

From: "Shamala Devi Sekaran" <SHAMALA@ummc.edu.my>
To: "tulip" <tulip@sanchamnet.in>
Sent: Tuesday, April 20, 2004 12:57 PM
Attach: Entercheck evaluation.doc
Subject: Entercheck evaluation

I attach the evaluation carried out by us. Would appreciate if you let me know before you publish anything with this.

Shamala

Shamala Devi
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shamalamy@yahoo.com

-----Original Message-----

From: tulip [mailto:tulip@sanchamnet.in]
Sent: Wednesday, October 08, 2003 7:55 PM
To: Shamala Devi Sekaran
Subject: Thanks

Dear Dr. Shamala Devi,

It was indeed a pleasure to meet you in your department during my visit to Kuala Lumpur.

Thanks are also due for providing me an opportunity to interact with the faculty and students from your department.

We are reviewing the data provided to us from the evaluation and prima-facie we are of the opinion that it is indeed possible to reduce the non-specificity of the test while maintaining the sensitivity.

We feel that this change can be done in 15 days time.

We would like to send you another 100 tests for your final evaluation if you can kindly consent to the same.

Thanks & best regards,
Linda Dias
For D.G. Tripathi

Typhoid kit Evaluation Record

- + — Clear band
- ± — Faint band
- — No band
- * — Result of first evaluation
- ** — Result of second evaluation

No.	LN	Blood Culture	Widal Titer			Enterocheck-WB	
			TO	AO	BO	Region T*	Region T**
1	42301	S. typhi +ve	≥1280	≥1280	≥1280	+	+
2	43483	S. typhi +ve	≥1280	≥1280	<160	+	+
3	41589	S. typhi +ve	≥1280	320	<160	+	+
4	28926	S. typhi +ve	≥1280	160	<160	+	+
5	43558	S. typhi +ve	≥1280	160	<160	±	+
6	21729	S. typhi +ve	≥1280	≥1280	640	+	+
7	46821	S. typhi +ve	≥1280	≥1280	<160	+	+
8	30906	S. typhi +ve	≥1280	≥1280	<160	+	+
9	30907	S. typhi +ve	≥1280	≥1280	320	+	+
10	39969	S. typhi +ve	640	160	<160	±	+
11	23153	S. typhi +ve	640	640	<160	±	+
12	37291	S. typhi +ve	<160	160	<160	±	+
13	30974	S. typhi +ve	640	640	<160	±	+
14	33060	S. typhi +ve	≥1280	≥1280	<160	±	+
15	57922	S. typhi +ve	640	160	<160	±	+
16	20424	S. typhi +ve	160	<160	<160	±	+
17	20063	S. typhi +ve	320	160	160	±	+
18	74263	S. typhi +ve	640	320	<160	±	+
19	37556	S. typhi +ve	640	320	<160	±	+
20	26394	S. typhi +ve	160	160	<160	±	+
21	61378	S. typhi +ve	<160	<160	320	-	-

22	63200	S. typhi +ve	<160	<160	<160	-	-
23	56413	S. typhi +ve	160	160	<160	±	-
24	52804	S. typhi +ve	320	<160	<160	±	+
25	37861	S. typhi +ve	≥1280	160	<160	±	+
26	30336	S. typhi +ve	<160	<160	<160	-	-
27	51733	S. typhi +ve	<160	<160	<160	-	-
28	45324	S. typhi +ve	640	320	<160	±	+
29	53132	S. typhi +ve	160	<160	<160	±	-

No.	LN	Blood culture	Widal Titer			Enterocheck-WB	
			TO	AO	BO	Region T*	Region T**
30	12233	S. typhi -ve	<160	<160	<160	±	-
31	00664	S. typhi -ve	<160	<160	<160	±	-
32	01205	S. typhi -ve	<160	<160	<160	±	-
33	08888	S. typhi -ve	<160	<160	<160	±	+
34	01203	S. typhi -ve	160	160	<160	±	-
35	01176	S. typhi -ve	<160	<160	<160	-	-
36	00967	S. typhi -ve	<160	<160	<160	-	-
37	00750	S. typhi -ve	<160	<160	<160	-	-
38	00746	S. typhi -ve	<160	<160	<160	-	-
39	84008	S. typhi -ve	160	<160	<160	±	-

No.	LN	Group of Sera	Widal Titer			Enterocheck-WB	
			TO	AO	BO	Region T*	Region T**
40	20216	Lepto. +ve	160	<160	<160	±	+
41	18445	Lepto. +ve	<160	160	<160	±	+
42	16235	Lepto. +ve	<160	<160	<160	-	-
43	38254	Lepto. +ve	<160	<160	<160	-	-
44	02966	Lepto. +ve	<160	<160	<160	-	-
45	42202	ST +ve	≥1280	<160	<160	+	-
46	75896	ST +ve	<160	<160	<160	±	-
47	37893	ST +ve	<160	<160	<160	-	-
48	21802	ST +ve	320	320	<160	±	-
49	26441	ST +ve	160	160	<160	±	-
50	35363	ST +ve	<160	<160	<160	-	-
51	86792	ST +ve	<160	<160	<160	±	-
52	50739	ET +ve	<160	<160	<160	-	-
53	67001	ET +ve	<160	<160	<160	±	-
54	03356	ET +ve	<160	<160	<160	±	-
55	02595	ET +ve	<160	<160	<160	-	-
56	72245	ET +ve	640	<160	<160	±	-
57	DF 29134	Den +ve	<160	160	<160	±	-
58	DF 29136	Den +ve	<160	<160	<160	-	-
59	DF 29137	Den +ve	160	<160	<160	±	-
60	DF 29138	Den +ve	160	<160	<160	±	+
61	DF 29139	Den +ve	160	<160	<160	±	-
62	DF 29141	Den +ve	<160	<160	<160	±	-
63	DF 29145	Den +ve	<160	<160	<160	±	-
64	M 13192	M +ve	<160	<160	<160	±	-
65	M 13160	M +ve	160	160	<160	±	-
66	M 12774	M +ve	160	160	<160	±	-
67	M12730	M +ve	160	<160	<160	±	-
68	M 12584	M +ve	160	<160	<160	±	-
69	M 12437	M +ve	160	<160	<160	±	-

70	M 10683	M +ve	<160	320	<160	±	-
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No.	RN	Group of Sera	Widal Titer			Remark	Enterocheck-WB*	
			TO	AO	BO		Region T*	Region T**
71	17733413	Widal +	640	<160	<160		+	-
72	17631297	Widal +	320	<160	<160		±	+
73	17678790	Widal +	640	<160	<160		+	+
74	17777453	Widal +	160	<160	<160		+	-
75	17669103	Widal +	320	<160	<160		+	+
76	12773988	Widal +	320	<160	<160		±	+
77	17641381	Widal +	320	320	<160		±	-
78	14680430	Widal +	160	320	<160		+	-
79	17739616	Widal +	320	<160	<160	AH640	-	-
80	17783720	Widal +	<160	<160	<160	AH640	±	-
81	17553466	Widal +	640	160	320		+	+
82	01200348	Widal +	<160	<160	<160	BH320	±	-
83	17498503	Widal +	≥1280	<160	<160		+	-
84	17494176	Widal +	320	<160	<160		±	+
85	17523699	Widal +	320	<160	<160		±	-
86	04470346	Widal +	<160	<160	<160	TH320	-	-
87	17818036	Widal +	640	<160	<160		+	-
88	17800420	Widal +	<160	<160	<160		-	-
89	11879227	Widal +	640	160	160		+	+
90	04148436	Widal +	640	640	<160		+	+
91	17696453	Widal +	320	<160	<160		±	+
92	LN1011703	Widal +	320	320	<160	TH1280	+	-
93	17657091	Widal +	640	320	160	TH1280	±	+
94	07522789	Widal +	1280	160	<160		-	+
95	14488296	Widal +	320	<160	<160		+	+
96	17766460	Widal +	<160	<160	<160	AH640	+	+

97	Iyngkaran	Widal +	160	320	<160		±	-
98	17567947	Widal +	640	1280	<160		+	+
99	15389801	Widal +	<160	320	<160		-	-
100	Rahmat B. Bayudi 16707427	Widal +	<160	<160	160		±	+

Result:

	No. of positive test	No. of negative test	Total
1. Blood culture positive	23	6	29
2. Normal serum	1	9	10
3. serum positive for other infectious agents	3	28	31
4. Widal positive	15	15	30
			100

$$\begin{aligned} \text{Sensitivity} &= \text{No detected by kit} / \text{No of true positives} \times 100\% \\ &= 23/29 \times 100\% \\ &= 79.3\% \end{aligned}$$

$$\begin{aligned} \text{Specificity} &= \text{No detected by kit} / \text{No of true negatives} \times 100\% \\ &= (9 + 28) / 41 \times 100\% \\ &= 90.2\% \end{aligned}$$

$$\begin{aligned} \text{Efficiency} &= \text{No of true negatives} + \text{No of true positives} / \text{Total} \\ &= (23 + 9 + 28) / 70 \times 100\% \end{aligned}$$

For further information contact :



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Micropress



Viola



TULIP GROUP OF COMPANIES

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