DIAGNOSTIC ACCURACY OF TYPHIDOT IN PATIENTS OF TYPHOID FEVER

Karamat Hussain, Hammad Jamal, Tariq Bashir, Sanaullah, Umair Ijaz, Ismail Khoso

Combined Military Hospital Kohat/National University of Medical Sciences (NUMS) Pakistan

ABSTRACT

Objective: To assess the diagnostic accuracy of Typhidot test in patients with acute febrile illness taking blood culture as gold standard.

Study Design: Cross-sectional validation study.

Place and Duration of Study: The study was conducted at Combined Military Hospital Kohat, from Mar 2016 to Oct 2016.

Material and Methods: In this study 211 patients with acute febrile illness were included. All patients had Typhidot IgM test done along with blood cultures, blood counts, chemistries and relevant diagnostic tests. Patients were divided into two groups based on blood culture results and both groups were compared in terms of positivity for Typhidot. Sensitivity, specificity, positive predictive value and negative predictive value were calculated using SPSS v 20. Chi square was applied to assess the association between Typhidot and blood culture results.

Results: Out of total 211patients, 49 patients had typhoid fever (culture positive) and 162 had non-typhoidal illnesses (culture negative). Typhidot IgM was positive in 47 (95.9%) cases of typhoid fever and in 155 (73.5%) cases of non-typhoidal fevers. The sensitivity of Typhidot for diagnosis of typhoid fever was 95.9% and specificity was 26.5%.

Conclusion: Our study reveals that Typhidot IgM has high sensitivity for typhoid fever but specificity and diagnostic accuracy are very low. Nevertheless, a high negative predictive value means it can help rule out the disease in suspected cases.

Keywords: Blood culture, Diagnostic accuracy, Typhidot, Typhoid.

INTRODUCTION

Typhoid fever remains a common cause of morbidity and mortality in developing countries like Pakistan. Around 22 million cases of typhoid fever occur annually worldwide with children bearing the major brunt of the disease[1]. Typhoid fever is estimated to cause 600,000 deaths every year, mostly in Asia[2]. Despite being a common disease, the actual incidence can not be accurately assessed owing to overlap of clinical features with other febrile illnesses and lack of diagnostic facilities in most of the heavily afflicted areas[3]. The gold standard of diagnosis has been culture of organism from blood or other body fluids[4]. Nevertheless a number of rapid tests have been designed that require minimum technical training and financial input, results are available much earlier than culture[5]. These include the well renowned Widal test, Typhidot, TUBEX and many others. The current study aims to determine the diagnostic accuracy of Typhidot test keeping in view the fact that it is commonly being utilized in our setup. The indiscriminate use of this test often leads to erroneous results, protracted course of antibiotics and financial implications for the patients and health system.

MATERIAL AND METHODS

This study was conducted at Combined Military Hospital Kohat from Mar 2016 to Oct 2016. It was a cross sectional validation study designed to assess the diagnostic accuracy of Typhidot test against blood culture taken as gold standard for the diagnosis of Typhoid fever. The initial sample consisted of 290 patients who were admitted in the medical ward. A non-probability convenience sampling technique was used. All patients underwent thorough history

Correspondence: Dr Hammad Javed, Department of Medicine, Combined Military Hospital Rawalpindi Pakistan
Email: hammad2286@gmail.com
Received: 09 Jun 2017; revised received: 15 Mar 2018; accepted: 21 Mar 2018

and physical examination. Blood samples were drawn for blood counts, liver function tests, urea, creatinine, electrolytes, thick and thin smear for malarial parasite, Typhidot IgM and blood cultures. Urine was sent for routine examination and cultures were advised in those with abnormal urinalysis. Chest x-rays were done in those with respiratory symptoms. About 89 patients having inconclusive clinical or laboratory diagnosis after thorough assessment and investigations were excluded from the study.

All patients presenting with fever for more than three days were included in the study. All patients who did not have a definitive diagnosis based upon predefined criteria were excluded from the study. Those with positive blood culture for Salmonella typhi were diagnosed as having typhoid fever. Malaria was diagnosed on the basis of ICT rapid diagnostic test or positive peripheral smear for malarial parasite. Acute viral hepatitis (AVH) was diagnosed on basis of clinical features and raised liver enzymes at least 10 times in a hepatocellular fashion. Dengue fever was diagnosed on basis of IgM positive test on ICT based rapid diagnostic method. Tuberculosis was diagnosed based on clinical features, chest x-ray and sputum analysis. A patient with clinical features, abnormal urinalysis and positive urine culture was diagnosed as having urinary tract infection. Respiratory tract infection/pneumonia was diagnosed on basis of consistent clinical features, raised TLC and/or presence of infiltrates on chest x-ray that responded to antibiotic therapy.

The Typhidot Test

The Typhidot assay (Reszon Diagnostics International Sdn. Bhd. Malaysia) was used in our study. It is a test very commonly employed for diagnosis of Typhoid fever in acutely febrile patients in our setup[6]. It is a simple, rapid and easy to perform test which does not require specialized training of staff or sophisticated equipment. It is done using an ELISA test strip detecting antibodies (both IgG and IgM) directed against outer membrane protein (OMP) of Salmonella typhi[7]. It usually becomes positive within 48 to 72 hours of the onset of fever. IgM is considered to represent recent infection and IgG can be positive in cases with previous infection. Both IgM and IgG positive results are considered to represent acute infection. About 0.05 ml of patient's serum was applied to the test pad and appearance of color band against IgM area was considered to be positive.

Blood Cultures

Blood culture remains the gold standard for the diagnosis of typhoid fever. It has a specificity of almost 100% whereas sensitivity ranges between 40 to 80%[8]. About 5 to 10 ml of venous blood was collected using aseptic technique and was inoculated on MacConkey, chocolate and blood agar plates and incubated at 37^{0}C for 7 days. Positive blood cultures were identified using specific biochemical tests. Drug susceptibility testing was done using Kirby-Bauer disc diffusion method.

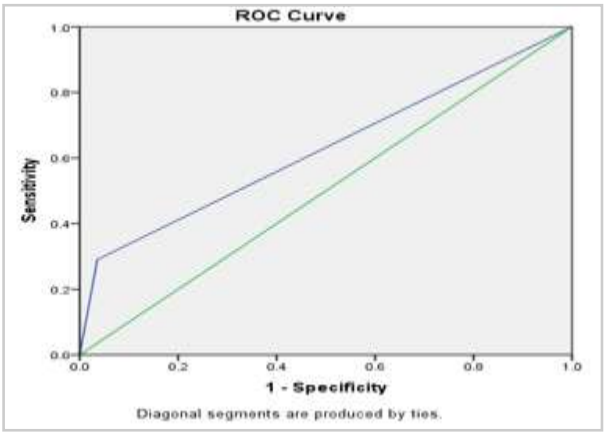

Figure-1: The ROC curve for sensitivity and specificity.

Data Analysis

The data was collected on predesigned performa and analyzed systematically using SPSS v 20. Patients were divided into two groups; first group comprised patients with typhoid fever and second group included patients with nontyphoidal illnesses. Mean ± SD for age was calculated whereas frequency and percentage were calculated for categorical variables. Sensitivity, specificity, positive predictive value and

negative predictive values were calculated using 2x2 contingency tables on SPSS v 20. The association between Typhidot and blood cultures was established using Chi Square test. A *p*-value less than 0.05 was considered significant.

RESULTS

The sample initially consisted of around 290 patients out of whom 79 patients were excluded due to inconclusive diagnosis, non-infectious causes of fever, or incomplete medical records. Majority of the patients 164 (77.7%) were males and 47 (22.3%) were females. Mean age was 27.87 years (SD ± 11.30). Out of 211 patients included in the statistical analysis, 155 (73.5%) tested positive for Typhidot IgM and 56 (2.65%) tested negative. Blood culture results for salmonella typhi were positive for 49 (23.2%) patients whereas 162 (76.8%) patients had negative results. On the basis of these results, the sensitivity and specificity of Typhidot for typhoid fever was found to be 96% and 33.3% respectively. The positive predictive value of Typhidot for the diagnosis of typhoid fever was 30.3% and negative predictive value was 96.4%. The diagnostic accuracy was found to be 47.86%. In the nontyphoidal group, false positive rate of Typhidot was as high as 66.7%. The *p*-value using Chi square test was less than 0.001 showing significant association between Typhidot and blood culture results. The ROC curve for sensitivity and specificity is shown in fig-1. The results are summarized in table. The frequency of various diagnosis among the patients is show in fig-2.

Table: Cross tabulation between the disease (typhoid and nontyphoidal fevers) vs Typhidot (positive or negative).

		Diagnosis		Total
		Typhoid (Culture Positive)	Nontyphoidal (Culture Negative)	
Typhidot	Positive	47 (TP)	108 (FP)	155
	Negative	2 (FN)	54 (TN)	56
		49	162	211

TP: True positive, FP: False positive, FN: False negative, TN: True negative

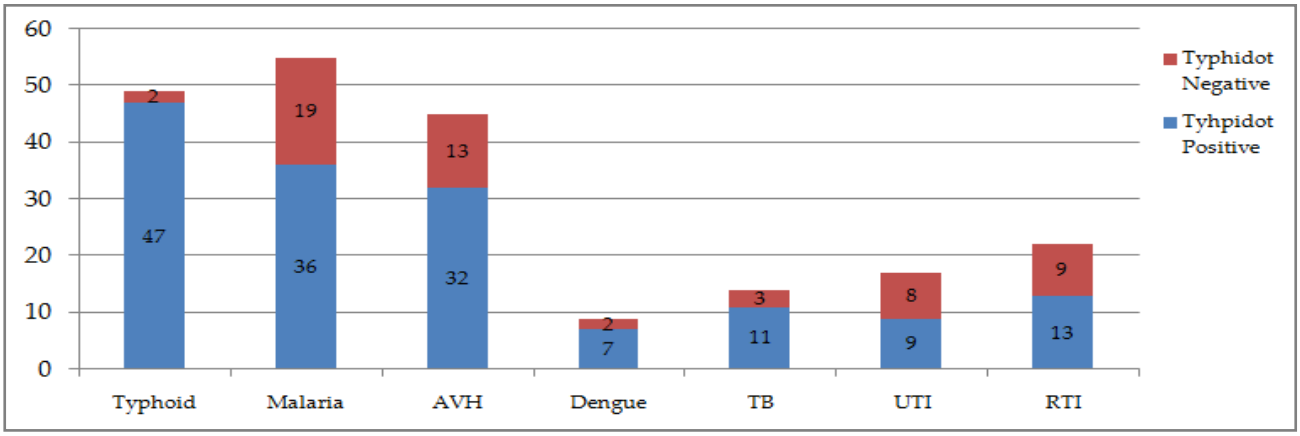

Figure-2: Distribution of different diseases with Typhidot results.

DISCUSSION

The definitive diagnosis of typhoid fever has remained an elusive aim for the clinicians due to widespread empirical use of antibiotics and non-availability of laboratory facilities[8]. The strength of our study is that it included culture positive cases of typhoid fever with laboratory proven cases of common non typhoidal febrile illness like malaria and AVH etc. It was found out that the sensitivity of Typhidot IgM was high (96%) but specificity was quite low (33.3%). In one of the earliest studies evaluating Typhidot in Pakistan,

Bhutta *et al*. concluded that the sensitivity and specificity of Typhidot for culture proven cases of typhoid fever (n=46) was 85 and 89% respectively[9]. In a latest national study it was found in 147 patients, the sensitivity of Typhidot was 27% and specificity was 61%. In this study the only 15 (10%) patients had a positive blood culture for Salmonella typhi and the false positive rate of Typhidot was as high as 35%[10]. In an Indian study using blood culture as standard, the sensitivity and specificity of Typhidot was found to be 100% and 63% respectively[11]. In an African study comparing various rapid diagnostic tests for Typhoid fever, the sensitivity, specificity, PPV and NPV were 75%, 61%, 57% and 78% respectively[12]. In a study from Bangladesh, the sensitivity of Typhidot was found to be 60% and specificity was 80%[13]. In another Bangladeshi study patients were divided into 6 groups including blood culture positive, Widal positive, healthy adults and patients with illnesses other than typhoid[13]. They showed that the sensitivity and specificity of Typhidot varied among different groups depending on whether the culture proven cases were compared with different patient groups i.e. whether the comparison group comprised culture negative cases only, or it comprised healthy adults or patients with nontyphoidal illnesses. The sensitivity varied from 64% to 54% and specificity varied from 80% to 72% in different groups of the same study. An important aspect of our study is the high false positive rate (66.7%) of Typhidot test in patients with laboratory proven nontyphoidal illnesses like malaria or AVH. This can be due to previous subclinical exposure to Salmonella antigens or due to cross reactivity with nontyphoidal salmonella antigens in the sera of patients[14]. This fact was also highlighted in an Indian study which evaluated patients with concurrent typhoid and malaria infection and found out that although serological test were positive in a significant number of patients (8.5%), the actual rate of coinfection as defined by positive blood culture results was only 1.6%[15].

CONCLUSION

Our study reveals that Typhidot IgM has high sensitivity for typhoid fever but specificity and diagnostic accuracy are very low. Nevertheless, a high negative predictive value means it can help rule out the disease in suspected cases.

CONFLICT OF INTEREST

The study has no conflict of interest to be declared by any author.

REFERENCES

1. Darton TC, Blohmke CJ, Pollard AJ. Typhoid epidemiology, diagnostics and the human challenge model. Curr Opin Gastroenterol 2014; 30: 7-17.
2. Crump JA, Luby SP, Mintz ED. The global burden of typhoid fever. Bull World Health Organ 2004; 82: 346–53.
3. Andrews JR, Ryan ET. Diagnostics for invasive Salmonella infections: Current challenges and future directions. Vaccine 2015; 33, Suppl 3: C8-15.
4. Bhutta ZA. Current concepts in the diagnosis and treatment of typhoid fever. Br Med J 2006; 333: 78–82.
5. Fadeel MA, House BL, Wasfy MM, Klena JD, Habashy EE, Said MM, et al. Evaluation of a newly developed ELISA against Widal, TUBEX-TF and Typhidot for typhoid fever surveillance. J Infect Dev Ctries 2011; 5: 169-75.
6. Hussain M, Hakeem A, Sharif F, Abdulhaq N. Comparative efficacy of commonly used serologic tests used in the diagnosis of enteric fever. J Sheikh Zayed Med Coll 2013; 4(3): 475-78.
7. Olsen SJ, Pruckler J, Bibb W, Thanh NT, Trinh TM, Minh NT et al. Evaluation of Rapid Diagnostic Tests for Typhoid Fever . J Clin Microbiol 2004; 42(5): 1885-89.
8. Upadhyay R, Nadkar M, Muruganathan A, Tiwaskar M, Amarapurkar D. API Recommendations for the Management of Typhoid Fever. J Assoc Physicians India 2015; 63: 77-96
9. Bhutta ZA, Ali NM. Rapid serological diagnosis of pediatric typoid fever in an endemic area: A prospective comparative evaluation of two dot-enzyme immunoassays and the Widal test. Am J Trop Med Hyg 1999; 61(4): 654-57.
10. Mehmood K, Sundus A, Naqvi IH, Ibrahim MF, Siddique O. Typhidot. A blessing or a menace. Pak J Med Sci 2015; 31(2): 439-43.
11. Appalaraju A, Anila A. Mathews, Priya P. The clinical utility of Typhidot in the diagnosis of typhoid fever. J Pharm Biomed Sci 2013; 29(29): 831-35.
12. Keddy KH, Sooka A, Letsoalo ME, Hoyland G, Chaignat CL, Morrisseye AB, et al. Sensitivity and specificity of typhoid fever rapid antibody tests for laboratory diagnosis at two sub-Saharan African sites. Bull World Health Organ 2011; 89: 640-47.
13. Islam K, Sayeed M.A, Hossen E, Khanam F, Charles RC, Andrews J, et al. Comparison of the Performance of the TPTest, Tubex, Typhidot and Widal Immunodiagnostic Assays and Blood Cultures in Detecting Patients with Typhoid Fever in Bangladesh. PLoS Negl Trop Dis 2016; 10(4): e0004558.
14. Khanna A, Khanna M, Gill KS. Comparative Evaluation of Tubex Tf(Inhibition Magnetic Binding Immunoassay) for Typhoid Fever in Endemic Area. J Clin Diagn Res 2015; 9(11): DC14-DC17.
15. Verma D, Kishore S, Siddique M.E. Comparative Evaluation of Various Tests for Diagnosis of Concurrent Malaria and Typhoid Fever in a Tertiary Care Hospital of Northern India. J Clin Diagn Res 2014; 8(5): DC41-DC44.