

Evaluate the role of Widal test in diagnosis of typhoid fever

Dina A. Abdullah AL-Roubaey*

BSc, PhD

Summary:

Background: Widal test and blood culture together must be depended to diagnosis of typhoid fever.

Patients and Methods: 328 patients admitted to AL-Yarmouk teaching hospital suspected with typhoid fever, their diagnosis was made by using widal test and isolation of Salmonella species by using culture for blood.

Results: The results of patients with suspected typhoid fever to 258 positive widal test showed 27 patients with blood culture positive (19 patients for Salmonella typhi, 2 patients for paratyphi A, and 6 patients for Salmonella paratyphi B). Most infections between age groups (11-20) years and (21-30) years.

Conclusions: The signs and symptoms of typhoid fever are non-specific, and an accurate diagnosis on clinical grounds alone is difficult although a definitive diagnosis can be made by isolation of Salmonella typhi or Salmonella paratyphi A, B from blood. The resulting widal test lack sensitivity and specificity, particularly in a community with endemic typhoid fever.

Keywords: Widal test, typhoid fever, blood culture.

Fac Med Baghdad
2011; Vol. 53, No.1
Received Sept.2010
Accepted Oct. 2010

Introduction:

Typhoid is a common problem in developing countries. Infection acquired by the ingestion of pathogens, could remain localized in the gastrointestinal tract or could disseminate to other organs and body systems [1,2]. Salmonella typhi and Salmonella paratyphi A,B that cause enteric fever, these species of Salmonella are restricted to humans and do not have a reservoir in animals. Therefore, spread of the infection is from person to person, usually through contaminated food or water. After infection, the Salmonella that survive the antibacterial defenses of the stomach and small intestine penetrate the gut mucosa through the peyer's patches then reach the the intestinal lymph nodes, where they survive and multiply within macrophages then transported to mesenteric lymph nodes and to thoracic duct and then eventually discharged in to blood stream[3,4]. Disseminated to many organs like spleen, bone marrow, liver and peyer's patches (reticuloendothelial system) from this system the bacteria reinvade the blood to reach other organs like kidney and gall bladder and then enters the intestine for a second time in much larger numbers causes a strong inflammatory response [5].

Patients and methods:

Three hundred fifty eight febrile patients admitted to AL-Yarmouk teaching hospital with symptoms suspected with typhoid fever [6], and one hundred healthy persons as control group.

Serological methods: For detection of antibody titer in patients with unknown illness. Serum agglutinins rise sharply during the second and third week of Salmonella infection. At least 2 serum specimens should be obtained at intervals of 7-10 days to prove rise in titer [7].

The tube dilution agglutination test (widal test): Serial

(2-fold) dilutions of unknown serum are tested against antigens from representative Salmonella. The results are interpreted as follows [8, 9, and 10]:

A-High titer of «O» (1:160 or more) suggests that active infection is present.

B-High titer of «H» (1:160 or more) suggests past vaccination or past infection.

C-High or rising titer of «V» occurs in some carriers.

Bacteriological methods for isolation of Salmonella species:
Specimens: blood, serum.

Media:

- Enrichment cultures: use tetrathionate broth after incubation for (1-2) days, plated on.

- Selective media: like salmonella-shigella (ss) agar or on deoxycholate-citrate agar which favors growth of Salmonella and Shigella over coliform organisms.

- Differential media: like MacConkey's agar or deoxycholate media permits rapid detection of lactose non-fermenters.

Gram-positive organisms are some what inhibited.

-Bismuth sulfite media: permits rapid detection of Salmonella typhi which forms black colonies because of H₂S production.

Final identification: Suspect colonies from solid media are identified by biochemical tests like TSI (Triple Sugar Iron agar test), Urea's test, Motility test [11].

Statistical Methods: The results values were given as a percentage frequency, and a significant difference between these frequencies was assessed by Fisher's exact probability.

Results:

The results of patients with suspected typhoid fever to 328 patients showed significantly higher difference between widal test positive and blood culture positive as shows in table (1).

*Dept. of microbiology, College of Dentistry, University of Baghdad.

Table (1): the numbers and percentage of cases with suspected typhoid fever.

Results	Patients no.	Percentage %	
Widal test positive	258	78.66	P=1×10 ⁻⁶
Blood culture & widal test positive	27	8.23	
Total patients	328	100	

From blood culture positive results for the bacterial isolates shows in table (2), in this table there is no significant difference among percentage of Salmonella typhi and Salmonella paratyphi A or B.

Table (2): type of bacterial isolates from blood culture for 258 patients (widal test positive).

Type of bacteria	Number	Percentage %		
Salmonella typhi	19	7.36	P=N.S.	P= N.S.
Salmonella paratyphi B	6	2.32		
Salmonella paratyphi A	2	0.77		

N.S.=Not Significant.

Most positive widal test results have titer more 1:320 as shows in table (3), in this table there is no significant difference between O and H antibodies for Salmonella typhi or paratyphi A, B.

Table (3): the numbers and percentage of antibodies titers (1:320).

Type of bacteria	Type of antibody	Number of cases	Percentage%	
Salmonella typhi	O	68	30	P=N.S.
Salmonella typhi	H	40	20	
Salmonella paratyphi A	O	19	11	P=N.S.
Salmonella paratyphi A	H	17	10	
Salmonella paratyphi B	O	35	18	P=N.S.
Salmonella paratyphi B	H	19	11	
Total no. of widal test positive 258 patients			100	

N.S.=Not Significant.

The other titers have higher numbers from the total widal test positive cases as shows in table (4).

Table (4): positive widal test results for patients suspected with typhoid fever (febrile patients) and blood culture negative.

Organisms no.	Salmonella typhi antigen	No. of organisms for which the titer was as follows			
		1/80	1/160	1/320	1/640
Escherichia coli (61)	O	37	18	1	1
	H	43	5		
Klebsiella spp. (20)	O	4	1	1	
	H	5	3	2	
Proteus mirabilis (19)	O	1	2	1	
	H	2	2	1	1
Pseudomonas aeruginosa (20)	O	3	5	1	
	H	2	1	2	1
Salmonella spp. (25)	O	10	8	4	5
	H	13	5	3	3
Staphylococcus aureus (22)	O	12	8		
	H	19	7	3	
Streptococcus spp. (29)	O	11	9		
	H	18	5	1	
Acinetobacter spp. (5)	O	2			
	H	1	2	1	
Control persons (30)	O	2			
	H	1	1		
Total no.		231 patients			

The most age group infected patients between (11-20) years and (21-30) years and less in other age groups as shows in table (5).

Table (5): age groups of patients with suspected typhoid fever (widal test positive).

Age groups	No.of patients	Percentage %
1-10	18	6.98
11-20	58	22.48
21-30	88	34.11
31-40	34	13.18
41-50	27	10.46
51-60	18	6.98
61-70	15	5.81
Total no.	258	100

Discussion:

False-positive widal test results have been reported for patients with non-enteric fever salmonella infections, malaria, typhus, meningitis, immunological disorders and chronic liver disease [12,13,14]. In this study, elevated levels of agglutinins were found in patients with a variety of other bacteremic illnesses, including those caused by other *Salmonella* spp., *E. coli*, *Klebsiella* spp., and *Staphylococcus aureus*. In general, the level of O antibodies in these patients was higher than that of H antibodies. The elevated levels may have been due to cross-reacting antigens or an anamnestic response. There are more 40 cross-reacting antigens between typhi and other Enterobacteriaceae [15]. So this test needs to be evaluated using bacterial isolation as gold standard [16], and a definitive diagnosis can be made by isolation of salmonella typhi and salmonella paratyphi A, B from blood [17]. Thus results of serologic tests for salmonella infection must be interpreted cautiously, because of the possible presence of cross-reactive antibodies limits the use of serology in the diagnosis of salmonella infections.

Finally we must ask why our labs in all hospitals depended on widal test as best serological method for diagnosis of typhoid fever and do not make the right design by culturing of the blood to isolate the bacterial cause for this endemic disease in our country.

References:

- 1- Hoa, N.T. et. al. Community-acquired septicemia in an infectious diseases hospital in Vietnam. The importance of multi-drug resistant *Salmonella typhi*. *Trans. R. Soc. Trop. Med. Hyg.* 1998; 29: 503-508.
- 2- Levine, M. M. et. al. Diagnostic value of the widal test in areas endemic for typhoid fever. *Am. J. Trop. Med. Hyg.* 1978; 27: 795-800.
- 3- Haque, A.; Ahmed, J. and Qureshi, J. Early detection of typhoid by polymerase chain reaction. *Ann. Saudi. Med.* 1999; 19(4): 337-340.
- 4- Boyd, R. F. *General Microbiology*. Times mirror, Mosbay. 1984; pp: 640-641.
- 5- Miller, SI. ; Pegues, DA. *Salmonella species, including Salmonella typhi*. In: Mandell, Douglas, and Bennett's *Principles and Practice of Infectious Diseases*, 5th ed. Mandell GL, Bennett JE, Dolin R. (editors). Churchill Livingstone, 2000; pp: 260-268.
- 6- Parry, C. M. et. al. Value of a single-tube widal test in diagnosis of typhoid fever in Vietnam. *Journal of clinical Microbiolog.* September. 1999; 37(9): 2882-2886.
- 7- Bopp, CA. et. al. *Escherichia, Shigella, and Salmonella*. In: *Manual of Clinical Microbiology*, 8th ed. Murray PR et. al. (editors). ASM Press, 2003; pp: 146-160.
- 8- Anonymous, A. Typhoid and its serology. *Br. Med. J.* 1: 389-390.
- 9- Pang, T.; and Puthuchery, S. D. Significance and value of the non typhoid *Salmonella* infections. *Southeast Asian. J. Trop. Med. Public Health.* 1983; 20: 163-164.
- 10- Schroeder, S. A. Interpretation of serological tests for typhoid fever. *J. Am. Med. Assoc.* 1968; 206: 839-840.
- 11- Brooks, G. F.; Butel, J. S. and Morse, S. A. Jawetz, Melnick and Adelberg's *Medical Microbiology*. Twenty-Third edition. Mc crow Hill. Pp: 257-259.
- 12- Harries, A. D.; Kamoto, O.; Mather, D.; Mukibii, J. and Khoromana, C. Specificity of widal test in healthy blood donors and patients with meningitis. *J. Infect.* 1995; 31: 149-150.
- 13- Senewiratne, B. and Senewiratne, K. Reassessment of the widal test in the diagnosis of typhoid. *Gastroenterology.* 1977; 73: 233-236.
- 14- Sharma, J. R.; Par mar, I. B.; Sharma, S. J. and Kesavan, A. False positive widal reaction in malaria. *Indian Pediatr.* 1993; 30: 1343-1347.
- 15- Esperson, F.; Hoiby, N. and Herts, J. B. Cross reaction between *Salmonella typhi* and 24 other bacterial species by CIEP. *Acta. Pathol. Microbiol. Scand.* 1980; 81: 243-248.
- 16- Khan, M.; Coovadia, Y. M.; Connolly, C. and Sturm, A. W. The early diagnosis of typhoid fever prior to the widal test and bacteriological culture results. 1998; 69: 165-173.
- 17- Hoffman, S. L. et. al. Duodenal string-capsule culture compared with bone-marrow, blood and rectal-swab cultures for diagnosing typhoid and paratyphoid fever. *J. Infect. Dis.* 1984; 149: 157-161.