# Original Article

## Clinical and Laboratory Findings Including the Diagnostic Value of the Widal Test in Pediatric Cases of Typhoid Fever in Tehran

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### ABSTRACT

***Background and Objective:*** **The diagnosis of typhoid fever in children is a challenge due to the non-specific clinical picture. The current role of the Widal test for diagnosis in developing countries has not been clear.**

***Materials and Methods:*** **Charts were reviewed on all children ≤ 15 years of age discharged from 5 pediatric teaching hospitals in Tehran from 1991 to 2004 with a diagnosis of typhoid fever. The Widal test was performed on 58 children with confirmed and 40 children with probable typhoid fever and as control groups, 40 febrile children admitted with infections other than typhoid fever, and 40 afebrile children admitted for elective surgery.**

***Results:*** **For the confirmed cases of typhoid fever, 33 (57%) were male and the mean age was 7.5 ± 3.5 years. Fever was present in 56 cases (97%) and the mean duration of fever before admission was 14 ± 8 days. Other symptoms included tachycardia (60%), anorexia (60%), vomiting (60%), diarrhea (57%), abdominal pain (48%), and headache (34%). Hepatomegaly was present in 55% of cases, splenomegaly in 44%, and Rose spots in 14% of them. Positive cultures were from blood (45/58), stool (18/58), and bone marrow (12/17) with 4 cases having only positive bone marrow cultures. Positive titers of at least 1:40 for anti "O" and/or anti "H" agglutinins were found in 78% of confirmed cases, 65% of possible cases, 12.5% of febrile controls, and no afebrile controls.**

***Conclusion:*** **The Widal test remains a useful test for diagnosis of typhoid fever in developing countries where blood cultures may not be available or may be negative because of prior antibiotic therapy. A titer of at least 1:40 for anti "O" and/or anti "H" agglutinin should be considered a positive titer in Iranian children.**

**Key words: Typhoid fever, Widal test, children**

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## Introduction

The diagnosis of typhoid fever in children is difficult as symptoms and signs are non-specific, especially in children less than 4 years of age (1-2). Diagnosis remains a particular challenge in developing countries (3). The sensitivity of blood cultures is low in typhoid fever in developed countries because small concentrations of Salmonella enteritica serotype typhi (S. typhi) are typically present in the blood, and the sensitivity is undoubtedly even lower in developing countries where technology for processing blood cultures is inferior (2, 4). Furthermore, it may not be practical to obtain outpatient blood cultures, which contributes to the common use of outpatient antibiotics in suspected cases prior to a blood cultures being obtained. The concentration of S. typhi is higher in bone marrow, so the sensitivity is higher than for blood cultures (5), but marrow cultures are even more difficult to obtain. A further problem is that the specificity of stool cultures is limited, as positive cultures in endemic areas can be indicative of asymptomatic carriage (2-3). Therefore, a simple, inexpensive, and sensitive screening test would be remarkably useful in developing countries (6).

The Widal agglutination test was first described in 1896 (7) and is widely available, simple, and inexpensive. The test detects agglutinating antibodies against O (surface polysaccharide) and H (flagellar) antigens of S. typhi. However, interpretation is not standardized and sensitivity is variable (8-13), but may be better in children than in adults (14). Agglutination can occur secondary to cross-reactions with other salmonellae and other Enterobacteriaceae (5, 14). It remains controversial if the Widal test should be used only for epidemiologic studies (3, 15, 16) or if it still has a role to play as a diagnostic test in developing countries (5, 14, 17). However, it is clear that diagnostic cut-off points will vary depending on the background rate of infection or colonization with S. typhi and possibly with other organisms (5, 14). The purpose of this study was to describe the clinical features of children with typhoid fever in Iran and to determine the utility of the Widal test and the appropriate cut-off value for a positive test in this population.

## Materials and Methods

### Children with typhoid fever

A chart review was completed from all children ≤15 years of age discharged from one of five pediatric teaching hospitals in Tehran (Iran) with a diagnosis of typhoid fever from January 1, 1991 to December 31, 2004. Confirmed cases had a compatible clinical course with positive blood and/or bone marrow cultures for S. typhi, with or without positive stool cultures. Probable cases had a compatible clinical course without positive cultures. Data collected on confirmed cases included the history of travel within the preceding 4 weeks to rural or hyper-endemic regions, recent consumption of possible contaminated food or water, and use of antibiotics since the onset of fever. The presence of significant weight loss (defined as more than 10% of body weight since the onset of disease), chills, increased perspiration, headaches, epistaxis, pharyngitis, cough, anorexia, abdominal pain, vomiting, diarrhea, constipation, dysuria, myalgias, dizziness, fever (defined as any temperature above 38°C during admission), tachycardia (heart rate over the 90th percentile for age), bradycardia (heart rate less than the 5th percentile for age), hepatomegaly, splenomegaly, or rose spots was noted. The results of the complete blood count, C-reactive protein, liver enzymes, and imaging of the chest or abdomen were recorded. Results of the Widal test (performed by a tube-agglutination method with an in-house kit) were recorded for confirmed and probable cases with the lowest measurable titer being 1:40.

### Control children

Control patients for determining the appropriate cut-off for the Widal test in this population were afebrile children admitted to the Rasoul Akram teaching hospital in Tehran for elective surgery (mainly adenoidectomy) and febrile children at the same hospital with a discharge diagnosis of pneumonia or urinary tract infection. Children with a previous history of salmonella infection or prolonged febrile illness were excluded. Serum was obtained and a Widal test performed after parental consent was obtained.

## Results

### Clinical characteristics of children with typhoid fever

There were 58 confirmed and 40 probable cases of typhoid fever. For the confirmed cases 33 (57%) were male and the mean age was 7.5 ± 3.5 years. A history was obtained of recent travel in 11 of 37 (30%) patients where the information was available, consumption of possibly contaminated food or water in 10/50 (20%) and recent antibiotic use in 31/50 (62%). The symptoms and signs that occurred in confirmed cases are shown in Table 1, with common symptoms being tachycardia (60%), anorexia (60%), vomiting (60%), diarrhea (57%), abdominal pain (48%), and headache (34%).

Only 12% of them had no gastrointestinal symptoms. Fever was present in 56/58 cases (97%) for a mean duration of 14 ± 8 days prior to admission and 5.3 ± 1.5 days after admission. Other common physical signs were hepatomegaly (55%) and splenomegaly (44%) with 38% of patients having neither.

**Table 1. Clinical characteristics of 58 confirmed cases of typhoid fever in children in five pediatric teaching hospitals in Tehran (Iran) during the years 1991-2004**

| Symptom | N (%) | Sign | N (%) |
|---|---|---|---|
| weight loss | 6(10%) | fever | 56(97%) |
| chills | 11(19%) | tachycardia | 35(60%) |
| increased perspiration | 6(10%) | tachycardia | 35(60%) |
| headaches | 20(34%) | hepatomegaly | 32(55%) |
| epistaxis | 6(10%) | splenomegaly | 25(43%) |
| pharyngitis | 4(7%) | rose spots | 8(14%) |
| cough | 8(14%) | | |
| anorexia | 32(55%) | | |
| abdominal pain | 28(48%) | | |
| vomiting | 32(55%) | | |
| diarrhea | 33(57%) | | |
| constipation | 6(10%) | | |
| dysuria | 4(7%) | | |
| myalgias | 6(10%) | | |
| dizziness | 9(16%) | | |

**Laboratory results of children with typhoid fever**

Laboratory results are shown in Table 2. Positive cultures were from blood (45/58), stool (18/58), and bone marrow (12/17) with 4 cases having only positive marrow cultures. The total white blood cell (WBC) count was normal or decreased in all but one case with 78% of children having predominantly polymorphonuclear cells in the WBC count differential test. Anemia was much more common than thrombocytopenia (both defined on the basis of mean normal values for the age) or elevated liver enzymes. Results of radiological imaging are shown in Table 2 with most examinations being normal.

For the 58 confirmed cases, 78% had a Widal titer ≥1:40 for "O" and /or "H" antigens. Agglutinins to "O" antigen were detected in 41%, 35%, and 26% of cases at titers ≥1:40, 1:80, and 1:160 respectively. Agglutinins to "H" antigen were detected in 76%, 74%, 65%, and 60% of cases at titers ≥ 1:40, 1:80, 1:160 and 1:320 respectively. In 21 out of 58 cases, agglutinins to "H" antigen alone were detected and in only one case agglutinin to "O" antigen alone was detected. For the 40 probable cases, 65% had a titer ≥1:40 for "O" and /or "H" antigens. Agglutinins to "O" antigen were detected in 52%, 52%, and 40% of cases at titers ≥1:40, 1:80 and 1:160 respectively. Agglutinins to "H" antigen were detected in 65%, 57%, 55%, and 47% of cases at titers ≥1:40, 1:80, 1:160, and 1:320 respectively.

**Table 2. Laboratory and radiographic findings in 58 confirmed cases of typhoid fever in children in five pediatric teaching hospitals in Tehran (Iran) during the years 1991-2004**

| Laboratory test | Results | Radiographic test | Results |
|---|---|---|---|
| C-reactive protein:<br>• within normal imits<br>• elevated | <br>2/17 (12%)<br>15/17 (88%) | Abdominal sonography:<br>• normal<br>• hepatomegaly, splenomegaly or both | <br>19/25 (76%)<br>6/25 (24%) |
| Total WBC count :<br>• normal<br>• decreased<br>• increased | <br>3/58(57%)<br>24/58(41%)<br>1/58(2%) | Chest radiograph:<br>• normal<br>• increased vascular markings or nonspecific infiltration | <br>53/58(91%)<br>5/58(9%) |
| Anemia | 43/58(75%) | | |
| Thrombocytopenia | 15/58(26%) | | |
| Elevated liver enzymes | 4/21(19%) | | |

**Widal results of control children**

The control group consisted of 40 afebrile children (mean age = 6.5 years; 23 males) and 40 febrile children (mean age = 3.5 years; 27 males). None of the afebrile children and 12.5% of the febrile children had a Widal titer ≥ 1:40 for "O" and /or "H" antigens with agglutinins to "O" antigen occurring in 7.5% and to "H" antigen in 12.5%. The highest titer in the febrile control group was 1:320 for "H" antigen.

## Discussion

It is common for physicians in developing countries to start empiric antibiotics when the diagnosis of typhoid fever is considered (2, 18). The use of clinical, simple, and inexpensive laboratory tests to guide this decision requires further study as overuse of antibiotics will ultimately lead to increased antibiotic resistance in common pathogens. The current study demonstrates that the diagnosis of typhoid fever is often delayed in developing countries, with symptoms persisting for a mean of 14 days prior to hospital admission. As in previous pediatric studies (1, 18-19), the clinical features were non-specific. Apart from fever, no sign or symptom occurred in more than of 60% of cases. As in a previous study from Malaysia (20), diarrhea and abdominal pain were prominent in addition to tachycardia, anorexia, and vomiting. Headache and constipation were much less common than in adult ones (19). The absence of chills and of decreased perspiration might help distinguish typhoid fever from brucellosis where chills and increased perspiration are common (2). Hepatomegaly or splenomegaly occurred in the majority of patients. As in a previous study (21), an elevated CRP was detected in almost all cases but this can occur with any inflammatory process. Also, as in previous studies (1, 18, 20), normal or decreased total white blood cell counts with neutrophil predominance were observed in most patients. The presence of thrombocytopenia in about one-quarter of cases was also similar to previous studies (20). However, about three-quarters of our patients had anemia, versus 30% in previous studies (20). This could be due to the longer interval from onset of symptoms to admission in the current study, or to a higher rate of nutritional anemia in our population.

Almost two-thirds of patients had received antibiotics for the current illness prior to admission, which could explain why almost half of the total cases had negative blood cultures. Blood cultures were obtained in all probable cases of typhoid fever, indicating that the sensitivity of blood cultures for typhoid fever in children in Iran may be as low as 46% if all probable cases had typhoid fever. This study confirmed the dictum that bone marrow cultures have improved sensitivity over blood cultures for S. typhi which may be because the concentration of organisms is higher in the marrow, and because it has been shown that marrow cultures remain positive for S. typhi longer than do blood cultures once antibiotics are started (2).

Using a cut-off titer of ≥ 1:40 for either the "O" or the "H" antigen, the sensitivity of the Widal test was 78% in the confirmed cases of typhoid. The test was positive in about two-thirds of cases of probable typhoid, but the sensitivity of the test cannot be determined in this population as there was no "gold standard" for confirming the diagnosis, and a discharge diagnosis other than typhoid fever might have been selected by the physician had the Widal test been negative. The specificity of a titer of ≥ 1:40 for either the "O" or the "H" antigen should be acceptable in Iranian children as none of the afebrile controls and only 12.5% of the febrile controls would have had a possible false positive test using this cut-off value. Most studies have recommended a higher cut-off titer than ≥ 1:40 (10, 12, 22), but a study conducted in 1987 from Papua New Guinea showed a cut-off titer of ≥ 1:40 to be appropriate (17). However, a follow-up study in the same population in 1992 suggested a higher titer was is not appropriate, possibly as the disease had become endemic in the intervening years.

Newer diagnostic techniques such as polymerase chain reaction using specific primers for S. typhi flagella appear promising (23). The inactive hemmaglutination test for antigen detection is specific but the sensitivity is low (22). Finally, various rapid serologic tests have developed to detect IgM and IgG with a sensitivity of approximately 70%-90% (3, 17) and a specificity of 50-90% (17). In one direct comparison, three of these tests had improved sensitivity and comparable specificity to the Widal test (3). However, these tests are not available in most developing countries.

In conclusion, the Widal test appears to still have a place as a diagnostic test in developing countries such as Iran. Future studies should track serial results of the Widal test in children with confirmed typhoid fever, clinical typhoid fever, and fever due to other gram negative organisms to better establish the sensitivity and the specificity of the test.

## Authors' contributions

AN wrote the protocol and the manuscript and performed the statistical analysis. JR reviewed the manuscript. SR, MK, and SS helped to carry out the protocol and reviewed the manuscript.

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