FALSE-POSITIVE WIDAL IN MELIOIDOSIS

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ABSTRACT

Enteric fever is endemic in this part of the world, and Widal test is one of the time-honored laboratory tests that are being used for years to diagnose the disease. On the other hand, melioidosis is a newly emerging disease from this region, which is most often misdiagnosed or underdiagnosed by clinicians. It is well accepted that false-positive Widal reactions following certain non-typhoid Salmonella infections may occur commonly. Three cases of high titers of Widal test are described, where melioidosis was the actual diagnosis in every occasion and was never suspected until diagnosed microbiologically. All the patients had shown a partial response to ceftriaxone. Blood and pus cultures grew Burkholderia pseudomallei, whereas Salmonella typhi was not isolated from blood in any patient. With appropriate antibiotics, the patients showed clinical and microbiological improvement with lowering of Widal titers. These 3 cases show that high Widal titer in any patient may mislead the diagnosis of melioidosis, and further laboratory workup should always be done to rule out melioidosis, especially in cases with nonresponsiveness to treatment.

Key words: Burkholderia pseudomallei, melioidosis, Widal

DOI: 10.4103/0019-5359.57647 **PMID:** 19901486

INTRODUCTION

Widal reaction is a diagnostic test for typhoid fever that measures agglutinating antibodies to the O and H antigens of *Salmonella typhi.* In developing countries, it remains the only practicable test because of its relatively lower cost and non-availability of other facilities for isolation in every hospital. However, this test is nonspecific and is often confusing as its results are difficult to interpret due to cross reactivity with nontyphoidal agglutinins.[1-3] Melioidosis is an emerging infection in India, which may present with a fatal outcome in many cases. There is no report of high-titer cross reactivity to *Burkholderia pseudomallei,* which is the causative agent of melioidosis. We describe 3 patients of melioidosis who were admitted to our tertiary care hospital situated in the coastal region of Karnataka, India, with high Widal titers; and these titers returned to normal with blood culture becoming sterile after treatment for melioidosis.

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CASE REPORTS

Case 1

A 50-year-old male clerk, a diabetic on oral hypoglycemic drugs, presented with acute onset of febrile illness and pain in the right shoulder. Clinical examination revealed mild pallor, icterus, bilateral swelling of the feet, hepatosplenomegaly and cardiac failure. Laboratory investigations revealed anemia, neutrophilic leukocytosis, elevated erythrocyte sedimentation rate and alkaline phosphatase with mildly elevated SGOT and SGPT(serum gamma glutamyl transferase and serum alanine amino transferase). Widal test (Span Diagnostics Ltd., Surat) showed high titer of O and H antigens (1:640, and 1:1280, respectively) as compared to the cut off titer of 1:160 (O and H). Parenteral ceftriaxone was started, to which the patient showed initial clinical response for 72 hours but continued to be febrile thereafter. Technetium 99 bone scan showed osteomyelitis of the right humerus. Blood cultures grew nonfermenting gram-negative bipolar stained *B. pseudomallei* (mini API ID 32 GN), which were sensitive to meropenam, cotrimoxazole, doxycycline, amoxicillin-clavulanic acid and ceftazidime, and resistant to gentamicin and polymyxin. The patient was treated with meropenam for 2 weeks and was later discharged with oral cotrimoxazole and doxycycline for another 12 weeks. He showed remarkable clinical improvement at the time of discharge. His repeat Widal titer done after 2 weeks was low (1:40 for O and 1:80 for H antigens) and the blood cultures were sterile. During follow-up, bone scan showed resolution of osteomyelitis.

Case 2

A 65-year-old male contractor, a diabetic, presented with fever of 15 days' duration and sudden onset of right-sided weakness and altered sensorium of 1-day duration. General examination was unremarkable. Central nervous system examination revealed confusion (GCS- 13/15), upper motor neuron palsy of the right facial nerve, right hemi paresis, facial nerve palsy and Broca's aphasia, with signs of meningitis. Laboratory investigations showed elevated erythrocyte sedimentation rate and alkaline phosphatase, and hyponatremia. CSF showed RBC- 20 cells, WBC- 20 cells with N- 62%,L- 38%, protein- 45 mg/dL and glucose- 190 mg/dL. Other parameters were ADA- 4 U/L and blood sugar (random) - 388 mg/dL. Computed tomography of the brain was normal, MRI brain showed small subdural collection bilaterally, although aspiration could not be done as it was very small. Widal titers for O and H antigens were 1:320, respectively, which was higher as compared to the cut off titer. Parenteral ceftriaxone was started, to which the patient showed initial clinical response for 72 hours but continued to be febrile thereafter. Meanwhile his sensorium improved after correction of hyponatremia. Ultrasound of the abdomen showed multifocal hypo-echoic lesions in the liver, the largest measuring 3.6 × 2.9 cm. Aspiration of the pus culture grew *B. pseudomallei* sensitive to meropenam, amoxicillin-clavulanic acid, amikacin, ceftazidime, ciprofloxacin and cotrimoxazole. The patient was treated with meropenam and amoxicillin-clavulanic acid for 2 weeks. He became afebrile and sensorium improved. The liver abscess reduced in size, subdural collection disappeared and the patient could be weaned off the ventilator. During the hospital stay, the patient developed

aspiration pneumonia, leading to respiratory arrest, and was continued on mechanical ventilation. The bronchoalveolar lavage grew *Pseudomonas aeruginosa,* which was sensitive to meropenam. As he was already continued on meropenam and amoxicillin-clavulanic acid, the treatment was not altered. The patient gradually showed improvement with therapy. Repeat Widal titers done after 6 weeks of therapy showed a declining trend; they came down to 1:40 for O and H antigens, respectively; however, the patient succumbed to fungal sepsis.

Case 3

A 38-year-old woman presented with febrile illness of 1-month duration. She was treated for enteric fever at a local hospital in view of significantly high Widal titers. Clinical examination revealed hepatosplenomegaly. Laboratory investigations showed leucopenia, elevated ESR and liver enzymes and high Widal titers for O and H antigens, viz., 1:320 and 1:640, respectively. The patient showed partial clinical response to ceftriaxone but continued to be febrile after 72 hours of therapy. Workup for other causes for pyrexia of unknown origin was inconclusive. During the hospital stay, she developed pain in the lateral condyle of the left tibia, and the bone scan confirmed the presence of osteomyelitis. Pus was aspirated from the osteomyelitis site, which grew *B. pseudomallei.* However, the blood culture was sterile. She was treated with imipenam for 2 weeks, when there was a remarkable clinical improvement in the patient and the Widal titers went down to 1:80 for O and H antigens, respectively. Later she was continued with doxycycline and cotrimoxazole for another 12 weeks.

DISCUSSION

The signs and symptoms of uncomplicated typhoid fever are nonspecific, and an accurate diagnosis on clinical grounds alone is difficult.[4] Although a definitive diagnosis can be made by isolation of *S. typhi* from blood or bone marrow,[5] in areas of endemicity, such as India, bacterial culture facilities are often unavailable and the Widal test is the only specific diagnostic investigation tool available. The Widal test has been in use for more than a century as an aid in the diagnosis of typhoid fever.[6,7]

False-positive Widal test results have been reported for patients with nonenteric salmonellae infections, malaria, typhus, *Cryptococcus neoformans* meningitis, immunological disorders and chronic liver disease.[8-10] 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 *S. 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 than 40 cross-reacting antigens between *S. typhi* and other *Enterobacteriaceae.*[11] False-positive Widal reaction due to nontyphoidal infection is a possibility in areas where typhoid fever is a common occurrence. In all the 3 above-described cases of melioidosis, the Widal titers were significantly high. Serological cross reactions could be due to sharing of common antigens between *Salmonella* and *Burkholderia.* This in turn is supported by the low Widal titer when the blood culture became sterile. Interestingly, all the 3 patients showed

a partial response to ceftriaxone drug, which is used for enteric fever. Ceftriaxone has been shown to have *in vitro* activity against *B. pseudomallei.*[12]

These cases re-emphasize the fact that a diagnosis of typhoid should not be made from a single positive Widal report and should be always backed up by blood culture of *S. typhi.* As melioidosis is an emerging infection in India[13,14] and has high mortality rates, awareness is needed among clinicians and microbiologists to look for *B. pseudomallei* in any suggestive case of sepsis, multiple abscess, chronic osteomyelitis or multi-organ failures. The outcome of this catastrophic infection could be better if early detection is done and appropriate treatment administered.

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Source of Support: Nil. **Conflict of Interest:** None declared.