Determination of baseline Widal titre among apparently healthy population in Dehradun City

Dr.Vijay K Kataria¹, Dr. Niti Bhai², Dr. Bhim S Mahawal¹, Dr. Reshmi C Roy¹

¹ (Department of Microbiology, SGRRIM & HS, Patel Nagar Dehradun. Uttarakhand, India) ²(Regency Hospital, Kanpur, India)

Abstract: Present study was conducted to determine the baseline widal titer of healthy population of Dehradun city. A total of 300 serum samples were collected from healthy individual with no history of fever and who had not received any vaccination for enteric fever. Tube agglutination test was done with commercially available antigens which contained the Salmonella enterica serovar typhi O and H antigens, the Salmonella enterica serovar paratyphi AH antigen and paratyphi BH antigen. In the present study an agglutination titer for TO - 1:20 is 28%, for 1:40 is 24%, followed by 1:80 and 1: 160 which is 10%, 4% respectively. The highest sample with an anti-H titre found with 1:20 (22%) followed by 1:40(17%). Based upon the results of the study it has been recommended that a single Widal can be significant in an endemic region when higher titre (1:160) is obtained.

Keywords: Baseline titre, Dehradun, Salmonella typhi, Typhoid fever, Widal.

I. INTRODUCTION

Enteric fever is endemic in developing country like India and it continues to be one of the major health problem here [1]. 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 mechanism for its persistence in its host that helps the organism to ensure its survival and transmission[2]. Enteric fever afflicts the local community and the travelers to the endemic areas. The incidence tends to be rise during the rainy season due to water logging and contamination of the water with sewage. The social factors that add to the enigma are the pollution of the drinking water supplies due to open air defecation, urination, contaminated food, personal hygiene, habits and poor health practices. The definite diagnosis of enteric fever in the patients with a compatible clinical picture is made on the basis of isolation of the Salmonella from blood, bone marrow, stool or urine and demonstration of the 4 fold rise in the antibody titer to both O and the H antigen of the organism between the acute and the convalescent phase [3].

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 detection of agglutinating antibodies to *S. typhi* and *S. paratyphi* by Widal test. Classically four-fold rise of antibody in paired sera is considered diagnostic of typhoid fever [4]. But paired sera are often difficult to obtain and specific chemotherapy has to be instituted on the basis of a single Widal test [5]. The aim of the present study was to determine the baseline titer of healthy population of Dehradun city.

II. Materials And Methods

This prospective epidemiological cross sectional study was conducted in the Department of Microbiology, Shri Guru Ram Rai Institute of Health and Medical Sciences, Dehradun, India after taking ethical clearance from the Institute's Ethical committee. The non-repetitive samples were collected from 300 healthy adults (>18 years old) who were apparently free from disease and not vaccinated for typhoid and not had fever since last six months . In order to calculate baseline antibody titres against the O and H antigens of *Salmonella enterica* serovar *Typhi/Paratyphi A and B*, tube agglutination method was adopted. The antigens used were from Span Diagnostics kit, GIDC, Sachin 394 230 (Surat) India. Kit literature was strictly followed.

III. Results

A total of 300 healthy adult volunteers of different age, sex and socioeconomic groups were screened for the agglutination against *Salmonella enterica* subspecies enteric serotypes, Typhi, Paratyphi A and Paratyphi B. Majority of individuals screened belonged to 18 to 40 years age group (Table 1). A titre of $\geq 1/20$ for O and H was present in 67% and 50.3% of individuals respectively, while a titre of $\leq 1/20$ for AH and BH was 92.6% and 85% of individuals respectively (Table 2). Table 3 shows tube agglutination result of the same at different dilutions.

Table 1 Age/gender wise distribution of samples					
Age of the healthy	Total	samples			
individual	received				
18-40		282			
>40		18			
Gender					
Male]	176			
Female]	124			

Table 1 Age/gender wise distribution of samples

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Serotype	Antibody type	≥1:20	%	≤1:10	%
Typhi	Anti O antigen	201	67	99	33
Typhi	Anti H antigen	151	50.3	149	49.6
Paratyphi A	Anti H antigen	22	7.3	278	92.6
Paratyphi B	Anti H antigen	45	15	255	85

Table 3 Tube agglutination result at different dilutions							
Antigen titre	0	Η	AH	BH			
	(%)	(%)	(%)	(%)			
≥1:20	84	66	14	20			
	(28%)	(22%)	(4.6%)	(6.6%)			
1:40	74	51	6	14			
	(24.6%)	(17%)	(2%)	(4.6%)			
1:80	31	23	2	10			
	(10.3%)	(7.6%)	(0.6%)	(3.33%)			
≥1:160	12	11	0	1			
	(4%)	(3.6%)	(0%)	(0.3%)			

IV. Discussion

Bacteriological culture remains the gold standard for definitive diagnosis of enteric fever. In an acute febrile illness in endemic typhoid region with ambiguous clinical picture, a rapid, accurate, specific and sensitive test should be used to differentiate typhoidal from non-typhoidal febrile illness [6]. However due to various constraints, in developing country like India we need an alternative test which is readily available, cheaper yet reliable. This makes widal agglutination test as the most common alternative laboratory procedure for the diagnosis of enteric fever. This test detects 'O' and 'H' antibodies against various Salmonella species . The O antigen is the somatic antigen and antibodies against the O antigen are predominantly IgM which rise early in the illness and disappear early. The H antigens are flagellar antigens of Salmonella typhi, paratyphi A and paratyphi B. Antibodies to H antigens are both IgM and IgG which rise late in the illness and persist for a longer time [4, 6]. In endemic areas, baseline anti O and anti H antibodies are present in the population owing to repeated subclinical infections with Salmonella Typhi / Paratyphi [6]. These antibody titers vary with age, socio economic strata, urban or rural areas and prior immunization with the TAB vaccine [7]. The specific purpose of this study was to develop local recommendations for the interpretation of Widal test. In the present study an agglutination titer for TO -1:20 is 28%, for 1:40 is 24%, followed by 1:80 and 1: 160 is 10% and 4% respectively. The highest sample with an anti-H titre found with 1:20 followed by 1:40 [Table 3]. This study showed 94 % of the samples with a titre which was equal to or less than 1 in 80 to O antigen and 92.7% samples had a titre which was equal to or less than 1 in 80 to the H antigen of Salmonella enteric serovar typhi. The distribution of 22 samples with an anti-AH titre of $\geq 1:20$ was seen in 14 samples (4.6%) and rest with an anti-AH titre \geq 1:40. Whereas the distribution of 45 samples with an anti-BH titre of \geq 1:20 was seen in 20 samples (6.6%) and 1:40 and 1:80 is 4.6% and 3.3% respectively. As 90% (20/22) and 75% (34/45) individual had a titre of $\leq 1:40$ for the Salmonella enterica servar paratyphy A and B, anything above this can be taken as diagnostic titre in this area[8].

Various Indian studies showed the baseline titre of 1:80 for O antigen [1, 8, 9] and same with anti-H agglutination [8,9] where as Pal et al., has documented significant titre of 1:40 for O and 1:80 for H antigen [3].

Punia et al., concluded in his studies that titers of 160 for anti-O and 320 for anti-H for enteric fever in Chandigarh [1]. The variation depends on the degree to which the enteric fever is endemic in each area a fact which may change over time. Hence this needs to be updated from time to time. In our study 4% of healthy population has shown titre of 1:160 for O antigen and 3.6% population shown agglutination against H-antigen.

Therefore 1 in 160 or above would be considered as significant and indicative of enteric fever if repeat sample is not available.

V. CONCLUSION

As in endemic areas population is permanently "immunologically sensitized" due to constant exposure, the response to infection is more rapid and reaches higher levels [10]. We may thus conclude that, despite the fact that knowledge of baseline titre of a region may help in the interpretation for diagnosis of typhoid fever, a single Widal can be significant in an endemic region when higher titre (1:160) is obtained.

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