To Study the Seroprevalence of Anti-Streptolysin O (ASO) Titers in Children Aged between 5 to 18 Years in Rural and Urban Region in Solan District, Himachal Pradesh: A Hospital based Study

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ABSTRACT

Introduction: Presence of Antistreptolysin O antibodies in a patient's sera may be an isolated evidence of recent infection by group A or less commonly, group C or G Streptococcus, especially in patients suspected of having a non-suppurative sequel to this infection. Current research aimed to study the seroprevalence of Anti-streptolysin O (ASO) titer in children aged between 5 to 18 years in rural and urban region in Solan district, Himachal Pradesh

Material and methods: The study was carried out for 12 months (April 2018- April 2019). For ASO testing, the ASO titre was measured by using rapid latex agglutination intended for semi quantitative determination of ASO titre in serum using (ARKRAY) ASO test kit.

Results: A total of 664 samples were received in the Laboratory during the study period. Of these, 237 (35.69%) were found to be positive for the presence of ASO titre of >200 IU/mL

Conclusions: The prevalence of ASO was found to be highest in the age group (10-14yrs). The presence of elevated streptococcal antibody titres in such a population reflects a high background prevalence of streptococcal infections. Thus, determination of ASO antibodies should be taken into consideration when evaluating group A streptococcus non-purulent complications.

Keywords: Antistreptolysin, ASO, Streptococcus, Antibody

INTRODUCTION

Acute rheumatic fever (ARF) is an auto immune consequence of infection of the throat (pharyngitis) with Group A hemolytic Streptococci (GAS) which causes an acute generalized inflammatory response and an illness that selectively affects the heart, joints, brain and skin. However, damage to the heart valves, particularly the mitral and aortic valves, may persist 2-3weeks even after an acute episode has resolved. This involvement of the cardiac valves is known as Rheumatic Heart Disease (RHD), the most significant sequela of ARF.[1]

Certain M protein serotypes, such as M types 1, 3, 5, 6, 14, 18, 19, and 24 of GAS, are found associated with throat infections and rheumatic fever (Stollerman, 1997, Mandor et al., 2013).[2] Streptococcal pharyngitis typically precedes the onset of acute rheumatic fever by 1 to 5 weeks.[1] Acute rheumatic fever(ARF) and its sequela, rheumatic heart disease (RHD), remain important public health problems in low- and middle-income countries, and persist in certain (predominantly Indigenous-minority) groups in high-income countries.[3]

Acute rheumatic fever (ARF) can occur at any age, although most cases occur in children 5 to 15 years of age. The mean incidence of acute rheumatic fever (ARF) is 19 per 100,000 school-aged children worldwide.[4] Surveys done in healthy school going children in the age range of 6 to 10 years found anti-streptolysin-O titers of more than 200 Todd units in 15 to 70% of the children.[10]

In infections caused by beta- hemolytic streptococci, streptolysin O is one of the two hemolytic exotoxins liberated from the bacteria. This stimulates the production of anti-streptolysin O (ASO) antibodies in the human serum.[5] Lack of primary prevention (treatment of group A streptococcal infections), and lack of screening programs to detect early RHD, results in late disease presentation, with most patients only seeking medical care due to symptoms related to complications of the disease.[6]

The antibody produced by the human host against this toxin, ASO, is the most widely used and the most standardized of the group A streptococcal antibody tests available.[7] Streptococcal antibody tests are used for the diagnosis of infections caused by group A Streptococcus and are particularly useful in the diagnosis of acute rheumatic fever and poststreptococcal glomerulonephritis.

Ideally, it is recommended that the titer be determined in the acute phase and then determined in the convalescent

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phase 2-4 weeks later, with a positive result defined as a rise in titer of two fold or more.[8] Often it is not feasible to get a second titer, then a single titer greater than the upper limit of normal can be considered evidence to presume streptococcal infection. A single ASO titer retains its role as a useful diagnostic tool and inversely correlates with disease progression.[8] Hence a single specimen when available requires to be compared with a predetermined baseline value or upper limit of normal (ULN) in a particular geographical area.[9] Present study has been conducted to determine the seroprevalence of Antistreptolysin O antibodies in a tertiary care health facility of Solan, Himachal Pradesh.

MATERIAL AND METHODS

An analytical study with cross-sectional design was carried out for 12 months (April 2018- April 2019).

Blood samples (2-3 mL) were collected by venipuncture from 664 children (5-18 years) visiting various departments of Maharishi Markandeshwar Hospital. Sera was separated and stored at 4ºC in a refrigerator till further use. For ASO testing, the ASO titre was measured by rapid latex agglutination intended for semi quantitative determination of ASOT in serum using (ARKRAY) ASO test kit.

This test method is based on an immunological reaction between exo-enzymes bound to biologically inert latex particles and streptococcal antibodies in the test sample. Positive and negative controls, provided along with the kit, were put up with every run of the test process.

A positive test is indicated by the presence of agglutination in the sera sample, within 2 minutes of adding the latex reagent. The sensitivity of the latex reagent has been adjusted to yield agglutination when the level of ASO is greater than 200 IU/mL. This cut off has been determined by epidemiological and clinical studies.

Data was collected using pre-designed and semi structured Performa through interview technique by the investigator herself. Written and informed consent was taken from all the subjects before initiating the enrollment. The confidentiality of the information was assured. Ethical approval was taken from Institutional Ethics Committee.

In our study we have excluded children less than 5yrs and more than 18yrs age and those who have active throat infection or had infection in last 2 month. Children who were already on antibiotic therapy and had already developed complications such us osteomyelitis, carditis, rheumatic heart disease, glomerulonephritis etc. have also been excluded.

RESULTS

A total of 664 samples were received in the Microbiology laboratory of MMMC&H Solan during the study period. Of these 237 (35.69%) were found to be positive for the presence of ASO having titre of >200 IU/mL. All the participants were divided into 3 groups according to the age criteria. Group 1 includes participants of 5 -9 yrs age, group 2 (10-14 yrs), group 3 (15-18 yrs). Out of these age group, maximum seropositivity, with ASO titre >200 IU/ml were seen in group 2 (10-14yrs). Out of the total participants, gender wise distribution showed that 386 were males and 278 were females. The seropositivity among the male participants (54.85%) was slightly towards higher side as compared to the female participants (48.14%). The participants were also divided on the basis of the location of the living into urban and rural population. In our study majority of the participants (72.5%) were located in the rural area of the Himachal where there are no medical facilities easily available due to difficulty in transportation and 27.4% belonged to the urban hilly area where they can have easy medical care accessibility.

Age group (years)	No. of patients (n= 664) N (%)	Positive ASO Titre (>200 IU/ml) (n=237) N (%)
Group 1(5-9 yr)	292 (43.97%)	90 (37.97%)
Group 2 (10-14 yr)	232 (34.93%)	130 (54.85%)
Group 3 (15-18 yr)	140 (21%)	17 (7.17%)

Table-1: Comparison table (according to age group) showing positive ASO titre out of 664 participants.

Gender	No. of patients (n= 664) N (%)	Positive ASO Titre(>200IU/ml) (n=237) N (%)
Male	386 (58.13%)	130 (54.85%)
Female	278 (41.86%)	107 (45.14%)

Table-2: Comparison table (according to gender) showing positive ASO titre out of 664 participants

Location of living	No. of patients (n= 664) N (%)	Positive ASO Titre (>200 IU/ml) (n=237) N (%)
Urban	182 (27.4%)	90 (37.97 %)
Rural	482 (72.5%)	147 (62%)

Table-3: Comparison table (according to location of living) showing positive ASO titre out of 664 participants.

High number of positive ASO titres were noted in age group 2 (10-14 yrs), a total of 54.85% cases. This correlates well with the fact that, this age group is more likely to get infected because of highest level of outdoor activity (table-1).

Gender wise distribution was well co-relating with most of the studies. ASO titres were higher in males in our study(n=130), as males are more exposed to outdoors in our rural population. Although in our study, there was no major difference in both the genders (M=130, F=107), possibly owing to higher incidence of streptococcal sore throat infections in general population in a hilly areas (table-2).

In congruence with other studies, our study also showed a lower incidence in urban population, owing to possible early, prompt and proper treatment of upper respiratory tract infections. Lack of proper facilities, lack of awareness and dependency on local practices rather than proper medical treatment could be attributable factors in a higher prevalence in a rural population (table-3).

DISCUSSION

Acute rheumatic fever is an autoimmune disease that follows

infection with group A streptococcus. However, isolation of group A streptococcus microorganism is uncommon, therefore confirmation of the diagnosis often relies on streptococcal antibody tests.

Most frequently performed test is Antistreptolysin O (ASO) titre. Ideally, it is recommended that the titre should be determined in the acute phase and determined in convalescent phase 14 to 28 days later with a positive result defined as a rise in a titre of twofold or more. However, it is not practicable to obtain a second sample for titre determination especially in a hilly area where frequent travelling from far off area is a big challenge. Therefore, it is generally accepted that if only single specimen is available and titre is greater than the upper normal limit at initial testing is considered presumptive evidence of a preceding streptococcal infection. The ASO titres are elevated in the acute phase and show a subsequent lower titre levels as the disease progresses. Presence of ASO in serum of a patient or an increase in ASO titre is usually suggestive of recent streptococcal infection. ASO antibodies is a simple, cost –effective way for detecting streptococcal infection.

Although ASO titer has provided a useful guideline to physicians, but this has been shown to vary with age, geographical location and site of infection. Therefore, we should interpret results judiciously keeping in mind the upper normal limits of ASO titre for the given geographical area since upper normal limits varies geographically.

In this present study, the seropositivity rate of ASO antibody is 35.69%. It has been found to be highest in age group 10-14yrs. The presence of elevated antibody titre in such a population reflects a high background of streptococcal infection.

ASO titre may vary for different population by age, socioeconomic status, geographic area and other factors related to the frequency of streptococcal infections.

The children who have higher ASO titre belong to rural area of Solan. These children belong to lower socioeconomic status having higher incidence as compared to high socioeconomic status children residing in urban area in solan. The high incidence in a low socioeconomic group is attributed to overall low standards of living associated with poorly ventilated, overcrowded houses, poor nutritional status and lack of awareness.

As we know from various studies that females are more prone to group A streptococcal infection as compared to male, but in our study, it has been found that males are more seropositive than females i.e. (9.7%).

ASO antibodies persist for 4-6 months, it is likely that healthy individuals in an endemic area may have persistently high titre due to repeated exposure. It is thus necessary to collect data in our own population so that we can have a baseline status of titre in local population.

CONCLUSION

The present study was done to estimate the seroprevalence of ASO antibodies. The prevalence of ASO was found to be highest in the age group 10-14 yrs. The presence of elevated streptococcal antibody titres was more in rural area than urban area. The presence of elevated titre in such a population reflects a high background prevalence of streptococcal infections. Thus, determination of ASO antibodies should be taken into consideration when evaluating the role of group A streptococcus in non-purulent complications of infections. However, we didn't focus on clinical outcome of the disease and didn't follow up the patients which is a drawback of the study.

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