SHORT PAPER

Human Leukocyte Antigen B27 in 453 Asian Indian Patients with Seronegative Spondyloarthropathy

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ABSTRACT

Background: Spondyloarthropathies are a group of closely related inflammatory arthritis which involve the axial skeleton and are negative for rheumatoid factor. **Objective:** This case-control study was conducted to examine HLA- B27 positivity in patients with seronegative spondyloarthritis (SSA) as per ESSG criteria and compare the frequency with healthy controls because a lower positivity is reported in Indians. **Method:** The study included 453 patients and 200 controls. HLA -B27 typing was done by microlymphocytoxicity and/or by sequence specific primers (SSP) using commercial kits. Patients were categorised as Ankylosing Spondylitis (AS), Undifferentiated Spondyloarthropathy, SSA with inflammatory bowel disease, reactive arthritis, psoriatic arthritis and juvenile spondyloarthropathy. **Results:** HLA-B27 antigen was present in 56% of patient and 3.5% controls with highest frequency in juvenile spondyloarthropathy (80%), followed by AS (76%). The P value < 0.001 for all categories of SSA and overall Odds ratio was 34.9. **Conclusion:** This study showed HLA-B27 frequency slightly lower than reported in Caucasian SSA patients and in our opinion HLA- B27 testing is extremely useful in young patients with suspected SSA.

Key words: HLA- B27, Spondylarthropathies, Asian Continental Ancestry Group

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INTRODUCTION

Seronegative spondyloarthritis (SSA) refers to a spectrum of joint disorders characterized by involvement of axial skeleton and a negative rheumatoid factor (1). Most of these conditions are insidious in onset and have a chronic course. HLA- B27 is the strongest predisposing factor for disease pathogenesis (2). This association with SSA is documented across the globe. To date 75 different subtypes of HLA- B27 (B*27:01-B*27:62) have been described. The aim of this study was to determine the prevalence of HLA- B27 in patients of Asian Indian origin diagnosed as SSA by ESSG criteria referred to the Pathology Department for HLA- B27 typing and to compare it with a control population (4). Various studies in India have examined the frequency of the antigen in diverse Indian populations and also its use for the diagnosis of SSA (5-7).

MATERIALS AND METHODS

This was a prospective study performed during the period of August 01, 2007 – October 15, 2010 and included 453 patients and 200 controls. All patients were tested for complete blood count (CBC) by a semi- automatic haematology counter, erythrocyte sedimentation rate (ESR) by Westergreen method, C reactive protein estimation by ELISA or semi-quantitative method by latex agglutination in addition to HLA- B27. X-Ray/ CT scan or MRI of some of the clinically affected joints was done where indicated. The subjects which included 376 males (83%) and 77 females (17%) were from all over India. A diagnosis of SSA was made as per ESSG criteria for all subjects (4). The data of control population (n=200) was derived from reports of HLA typing done for kidney donors, BMT donors and healthy controls. Typing for HLA- B27 was done by NIH micro- lymphocytotoxicity assay (Innotrain, Germany), for 75 patients and 60 controls, by SSP for 344 patients and 144 controls and by both methods for 34 patients. Statistical analysis of the accrued data was done by using Epi Info version 3.5.1 Aug 2008. As it was a case control study for only Odds Ratio (OR) and P values were calculated.

RESULTS

An abnormal ESR was seen in 60%, CRP was increased in 42%,and CBC was abnormal in 22% of the patients.The most common abnormality was normocytic normochromic anaemia. Six patients had a positive family history of AS. There was a male preponderance among patients (> 81%) for all subcategories. The duration of symptoms in positive patients ranged from one month to 20 years. The youngest patient was five years old and the oldest was 74 years with a maximum age range of 20-30 years. Table 1 shows the numbers of patients in each age group. Most of the patients (73%) and controls (85%) were Hindus. Overall, 255 samples (56%) were positive for B27 and 198 (44%) were negative. The diagnosis for various categories of SSA is shown in Table 2 with the percentages of B27 positive and negative cases in each category.

Table 1: Age distribution of SSA patients

Age in years	Number
< 20	35
20-30	175
31-40	155
41-50	62
>50	25

Table 2: Laboratory, imaging, HLA- B27 results and P values for various categories of SSA

SSA category	Males %	Lab. abnormality	Imaging. abnormality	B 27 +	B 27 -	Odds ratio	P value
Ankylosing Spondylitis	85	14	34	97	25	107	<0.001
Psoriatic arthritis	91	16	10	5	7	19.7	<0.001
Undifferentiated Spondyloarthropathy	82	17	21	114	136	22.7	<0.001
SSA with IBD	82	23	23	20	18	30.6	<0.001
Reiter's/Reactive arthritis	85	13	10	15	11	37.6	<0.001
Juvenile SPA	100	25	0	4	1	110	<0.001
Total		108	98	255	198		

Odds ratio and P values of each category of SSA is tabulated. A highly significant association was found between HLA -B27 positivity and SSA with P-value <0.001. All the serologically B27 positive samples also were positive for Bw4. There was 100% concordance in the results of all samples for which B27 typing was performed by both methods. Eight patients had ocular manifestations in addition to SPA; five of them were positive for B27 antigen. No cardiac or pulmonary extra articular manifestations were observed in any patient.

DISCUSSION

The human MHC class I molecule, HLA-B27, is strongly associated with seronegative spondyloarthropathies, the most common of which is AS. The association of AS and HLA-B27 was recognized in 1973 and is among the strongest association with HLA for any disease (8). HLA-B27 was among the first MHC molecules whose structures were elucidated because of the pronounced disease association (9). About 95% of the Caucasian AS patients express HLA-B27, although the frequency of this antigen is below 10% in healthy Caucasian adults (10). In contrast, the frequency of HLA- B27 in AS patients was 76% in our study. This is in agreement with another recent study

which showed the prevalence of B27 in 71.4% of juvenile SSA (11). Overall positivity was 56% which is higher than that reported by Shankarkumar and Sonkar *et al.* and who had total positivity of B27 in SSA was 8.4 and 43.6% of the samples, respectively (6, 12). As is characteristic of AS and other SSA, maximum number of patients were in the age groups of 20-30 years with a male predominance and a minimum in after more than 50 years old where degenerative conditions are more likely to predominate.

Frequency in control population of our study was 3.5% which is similar to the findings of Shankarkumar *et al.* who reported HLA- B27 frequencies of 2.37% in 5000 controls in various population groups in India (6). Lower frequency of HLA B 27 has been reported in seronegative Venezuelan and Iranian patients (20.96 and 17.26%) respectively (13, 14). The Venezuelan study included 620 SSA cases and 220 controls and showed an HLA- B27 positivity of 33.3% in AS, 30% in Reiter's arthritis and 4.3% in the controls (13). The other study had a low frequency of HLA- B27 positivity because it included 'suspected' and not confirmed SSA (14). Sonkar *et al.*, in a study on 110 patients with SSA, had an overall B27 positivity of 43.6% with a higher positivity in the children (68.75%) and in males (81.81%). In this study also there was a male predominance and higher B27 frequency in children.

The higher incidence of HLA- B27 in SSA pationts of this study as compared to that of other non- caucasian populations can be attributed to the stringent selection of patients as per ESSG criteria for this study (4). The B27 frequency range in controls [range 1.4-8%] and the patients [18-94%] was within the range for Indian population who were from all over the country and not from a particular community (6).

CONCLUSION

HLA- B27 positivity rate in Asian Indian population with SSA in our study was higher than that reported by others from this country and was similar to Caucasian population implying that HLA- B27 testing is important for all suspected cases of SSA in young patients. It may provide a clue to diagnosis before the onset of radiological changes and also it may help in identifying at risk family members. In our study we found 100% concordance between serological and SSP methods, so either method may be adopted depending on their availability.

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