RESEARCH ARTICLE

ASSOCIATION OF IL1- α 889CT WITH CLINICAL OUTCOMES OF RHEUMATIC HEART DISEASE

Amit Gupta, Anil K Singh
Department of Cardiology, Max Super Speciality Hospital, Mohali, Punjab, India

Correspondence to: Amit Gupta (docamitgupta@yahoo.co.in)

DOI: 10.5455/ijmsph.2014.030720142 Received Date: 26.05.2014 Accepted Date: 03.07.2014

ABSTRACT
Background: Rheumatic fever (RF) is an inflammatory disease of the heart after a pharyngitis by Group-A beta haemolytic streptococci. The pathogenetic mechanisms highlight a complex interplay of immunological, genetic and environmental factors. Immunity gene polymorphisms, in relation to susceptibility to RF, have been studied by many investigators, and IL-1 α has been the focus of attention.
Aims & Objectives: To investigate the association of ILI- α 889C/T, gene polymorphism with clinical outcomes of rheumatic heart disease.
Materials and Methods: A cohort of 157 patients of established rheumatic heart disease and 200 controls (HS) were enrolled. Genotyping was done for all cases and controls regarding IL-1 α gene.
Results: 58.6% of RHD patients had ILI- α 889T allele, as compared to 49.5% for HC and was not statistically significant (P=0.087;OR=1.4[0.9-2.3]). Frequency of ILI- α 889T allele (64.1%) was higher in cases with history of rheumatic fever compared to HC (49.5%), with statistical significance (P=0.028; OR=1.8 [1.03-3.24]). ILI- α 889C/T gene polymorphism did not show statistically significant relationship with either mitral valve lesion (MiVL) (P=0.252; OR=1.3 [0.80-2.15]), mitral valve lesion along with other valve lesion (MiVL^a) (P=0.99;OR=1.4[0.91-2.25]), Aortic valve lesion (AoVL) [Fisher exact, P=0.72] or Multiple valve lesion (MVL) (P=0.086; OR=1.8 [0.86-4.17], or AF (P=0.329; OR=0.73 [0.37-1.44]).
Conclusion: ILI- α 889C/T polymorphism of the ILI- α gene is not significantly associated with RHD, development of valve lesions or AF, but is significantly associated with history of RF.
Key Words: Valvular Heart Disease; Rheumatic Heart Disease; IL-1 Alpha Gene Polymorphism; Rheumatic Fever

Introduction

Rheumatic fever (RF)/ Rheumatic heart disease (RHD) is a connective tissue disease, and evidence supports the concept of environmental and genetic factors contributing to its pathology. Following exposure of individuals to group-A streptococci, only susceptible people developed RHD - this suggests the involvement of genetic factor. It is characterized by an inflammatory process involving heart, joints, central nervous system and subcutaneous tissues. Precise pathogenic mechanisms of Rheumatic Heart Disease (RHD) still remain elusive but indirect evidence supports the concept of an abnormal, autoimmune host response following exposure of susceptible individuals to group-A streptococcal antigens. Immunological activation involves both cellular and humoral responses.[1] Cross reactivity between streptococcal proteins and cardiac tissues leads to acute carditis, evolving finally into chronic rheumatic heart disease (RHD) and permanent disability.[2] This appears to be the most plausible explanation till date, but there are several unanswered questions. Why only 3% individuals with streptococcal sore throat develop rheumatic fever (RF), and why only 50% patients of acute rheumatic fever (ARF) develop carditis - are a few of them. Why only 1/3rd of affected children progress to development of RHD with such a highly variable pattern of cardiac involvement further needs to be explained. All these facts are pointers to a strong genetic component in the susceptibility to disease process. The initial attack of group-A streptococcal sore throat probably identifies those 3% individuals who have inherent susceptibility to develop rheumatic process.

The absence of ARF in young children suggests that repeated exposures of the host to group-A Streptococcus is essential for precipitating the illness.[3] There is also a higher concordance amongst monozygotic twins for the development of ARF. Certain HLA types, viz. HLA-DR 1, 3 and 4 haplotypes, have been implicated in certain ethnic groups.[4]

Familial aggregation was highlighted first in 1889 by Cheadle, whose own wife and child had the disease.[5] In 1927, a survey was conducted by British Medical council and 721 rheumatic families were studied. Interestingly, 23 of 53 descendants of a man who had RF, also had RF.[6] Subsequently, it was proposed that, RF is 4-8 times more common in relatives of patients with RF, compared to general population.[7] Several additional studies

supported the hypothesis that genetic profile of a given individual decides the response to the streptococcal antigen, and that the response was different in person developing rheumatic fever (RF), compared to person not developing it.[8]

First exploration of genetic susceptibility focus started with ABO blood groups, though studies failed to substantiate it. Second to be studied were the MHCs or HLAs. The lack of consistent association between class 1 HLA and RF prompted studies to investigate role of HLA II (includes HLA DR, DQ and DP) in RF. In many subsequent studies, a significant association of RF was seen with HLA DR3, DR4, and DR7 alleles.[9-11] Furthermore, genetic susceptibility to RF/RHD is shown to be associated with genes of MHC, particularly HLA-DRB1.[12] However, the diversity of association between HLA II antigens and RF suggests that non-HLA genes may also influence the development of the disease.

B cell allo-antigens determine host immune response, and may be a determinant of susceptibility to RF. A B-cell allo-antigen - 883 was identified in 71-74% patients of RF compared to only 17% in controls.[13] A close relationship has been observed between allo-antigen D8/17 and RF in multiple studies, though its role in pathogenesis of disease and clinical outcomes is still uncertain.[14-17]

Materials and Methods

Study Subjects: 157 patients of RHD and 200 controls were recruited from LPS institute of Cardiology, Kanpur, India. Controls were unrelated patients of CAD. All patients were proven cases of chronic RHD on echocardiography. In the study group, cases and controls were unmatched regarding age and sex (Patients in RHD group were of younger age with near equal sex distribution; whereas the control group of CAD patients comprised of subjects of higher age and predominated by males).

Genotyping: Blood samples of patients were obtained and sent to the Biotechnology Department of Indian Institute of Technology, Kanpur for DNA sampling. The polymorphisms at ILI- α 889C/T was typed by an established PCR/RFLP procedure (Migot-Nabias et al, 2000). DNA extraction and data for these 157 patients was compared to the 200 controls.

Analysis of Data: Statistical analysis was performed using Microsoft excel 2007, Epi info 2002 and Graphpad Prism 4.0 software. Genotype and allele frequencies were calculated by direct counting. Comparison between cases and controls was done by using χ^2– test along with odds ratio (OR) and 95% confidence interval [CI]. Hardy-Weinberg expectation (HWE) was determined by comparing the observed number of different genotypes with those expected under HWE for the estimated allele frequency. Statistical comparison was also carried out by Fisher exact test whenever a value in the contingency table was below five.

Results

Table-1: Basic characteristics of the patients

Characteristics	Male		Female	
	N	Mean age ± SD (years)	N	Mean age ± SD (years)
RHD Patients (n=157)	71	27.9 ± 12.3	87	31.1 ± 10.7
MiVL (n=112)	47	27.2 ± 12.9	65	30.9 ± 9.9
MiVLa (n=149)	62	27.3 ± 12.3	87	31.1 ± 10.7
AoVL (n=8)	8	32.4 ± 12.0	0	0
MVL (n=37)	15	27.6 ± 10.4	22	31.8 ± 13.1
RF (n=78)	37	25.2 ± 11.4	41	30.3 ± 10.6
AF (n=48)	24	31.6 ± 12.6	24	31.9 ± 10.0

N: Sample size; SD: Standard deviation; RHD: Rheumatic heart disease; MiVL: mitral valve lesion; MiVLa: mitral valve lesion along with other valve lesion; AoVL: Aortic valve lesion; MVL: Multiple valve

Table-2: Genotype and allele frequencies of ILI- α 889C/T in RF patients and healthy controls (HC)

	Geno-types	RHD (n=157)	HC (n=200)	χ^2	P	OD (95% CI)
IL1- α 889C/T	TT	11 (7.0)	14 (7.0)	2.93[b]	0.087	1.4 (0.9-2.3)
	CT	81 (51.6)	85 (42.5)			
	CC	65 (41.4)	101 (50.5			
Alleles	T	103 (32.8)	113 (28.3)	1.73	0.189	1.2 (0.9-1.7)
	C	211 (67.2)	287 (71.7)			

Figures in parenthesis show percentages; OR: Odds ratio: CI: Confidence interval; [a] AA/GA vs. GG; [b] TT/CT vs. CC

Table-3: IL 1- α 889C/T genotype and alleles in patients with RHD according to clinical phenotypes

IL 1- α 889C/T	TT	CT	CC	χ^2	P	OD [95% CI]
RHD	11 (7.0)	81 (51.6)	65 (41.4)	2.93	0.087	1.4 [0.9-2.3]
MiVL	7 (6.3)	56 (50.0)	49 (43.7)	1.31	0.252	1.3 [0.80-2.15]
MiVLa	10 (6.7)	77 (51.7)	62 (41.6)	2.71	0.099	1.4 [0.91-2.25]
AoVL	1 (2.5)	4 (50.0)	3 (37.5)	Fisher exact	0.72	
MVL	3 (8.1)	21 (56.8)	13 (35.1)	2.95	0.086	1.8 [0.86-4.17]
RF	9 (11.5)	41 (52.6)	28 (35.9)	4.81	0.028	1.82 [1.03-3.24]
AF	2 (4.2)	18 (37.5)	28 (58.3)	0.95	0.329	0.73 [0.37-1.44]
HC	14 (7.0)	85 (42.5)	101 (50.5)			

RHD: Rheumatic heart disease; MiVL: mitral valve lesion; MiVLa: mitral valve lesion along with other valve leasion; AoVL: Aortic valve lesion; MVL: Multiple valve lesion; RF: Rheumatic fever; AF: Atrial fibrillation; HC: Healthy controls; Figures in parenthesis show percentages; χ^2: TT/CT vs. CC

The basic characteristics of patients are shown in table 1. Out of 157 RHD patients, 112 had the mitral valve lesion (MiVL), 149 had mitral valve lesion along with other valve lesion (MiVL[a]), 8 had Aortic valve lesion (AoVL)

and 37 had multiple valve lesions (MVL). Only 78 patients gave the history of Rheumatic fever (RF) and 48 patients had history of atrial fibrillation (AF).

The frequency of – 889 single nucleotide polymorphism (SNP) of ILI- α gene was examined in all 157 RHD patients and 200 controls. Table 2 shows Genotype and allele frequencies of ILI- α 889C/T. No Statistical significance was found when RHD patients were compared to HC in relation to ILI- α 889T genotype (TT and CT) versus CC genotype (P= 0.087; OR=1.4 [0.9-2.3]). The – 889 allele appears to conform the non-susceptibility to RHD (P=0.189; OR=1.2 [0.9-1.7]).

Table 3 shows the stratification of patients according to clinical phenotype and comparison of genotype and allele frequency with HC. 58.6% of RHD patients had ILI- α 889T allele as compared to 49.5% for HC and is not statistically significant (P=0.087; OR=1.4 [0.9-2.3]). Subdivision analysis of RHD patients with history of RF showed high frequency of ILI- α 889T allele (64.1%) compared to HC (49.5%) with statistically significant results (P=0.028; OR=1.8 [1.03-3.24]) but does not show statistically significant relationship with either mitral valve lesion (MiVL) (P=0.252; OR=1.3 [0.80-2.15]), mitral valve lesion along with other valve lesion (MiVL[a]) (P=0.99; OR=1.4 [0.91-2.25]), Aortic valve lesion (AoVL) [Fisher exact, P=0.72], Multiple valve lesion (MVL) (P=0.086; OR=1.8 [0.86-4.17], or AF (P=0.329; OR=0.73 [0.37-1.44]).

Discussion

The pathogenesis of RF /RHD seems to result from an overt immune response involving either humoral or cellular reaction or both, triggered by group-A streptococci infection. The concept of an involvement of autoimmune reactions in the pathogenesis of RF was introduced only in the 1960s by Kaplan who demonstrated that antibodies against GAS reacted with human heart preparations.[18, 19] During the acute phase of the throat infection, inflammatory acute phase proteins, such as Mannose binding lectin (MBL), and the cytokines IL-1, IL-6 and TNF- α should be produced to eliminate the bacteria. There are many genes with inflammatory function, and one of these is the interleukin (IL)-1 gene cluster that is located on chromosome 2. It includes the genes expressing the pro-inflammatory cytokines IL-1a and IL-1b and their inhibitor IL-1 receptor antagonist (IL-1RA). The findings of a study suggested that variations in IL-1b and IL-1RA gene polymorphism are not suitable gene markers for RHD in Taiwan Chinese.[20] Further studies are needed before excluding IL-1 as a susceptible or protective factor. Our results show that IL-1 alpha 889C/T polymorphism of the ILI- α gene is not significantly associated with RHD population, hence suggesting that ILI- α is a not susceptibility locus for RHD.

It has been reported that the production of IL-1, IL-2, and TNF- α in the valvular lesions of RF patients is correlated with Aschoff nodule progression.[24] Our results show that IL-1 alpha 889C/T polymorphism of the ILI- α gene is significantly associated with history of RF. As the basic rheumatic process is inflammation and destruction of connective tissue, the effects of interleukin were involved in the pathogenesis of RHD. The extent of original inflammation and recurrence of RF are not the only predisposing factors for the progression of valvular lesions. Ultimately, the deformed valve is subject to nonspecific fibrosis and calcification. The anatomic changes in severe mitral stenosis or aortic stenosis may result from the combined effects of a persistent rheumatic process and a constant trauma to the mitral valve or aortic valve by the turbulent flow.[21,22]

The findings of a Chinese study suggested that the up regulation of IL-1beta gene expression may contribute to AF through influencing collagen metabolism.[23] Our results show that there is no statistically significant relationship between IL-1 alpha 889C/T polymorphism of the ILI- α gene and AF.

Conclusion

In conclusion, our study shows that ILI- α 889C/T polymorphism of the ILI- α gene is not significantly associated with RHD, development of valve lesions or AF, but is significantly associated with history of RF.

Abbreviations

RF: Rheumatic Fever; RHD: Rheumatic Heart Disease; MiVLa: Mitral Valve lesion with other valve lesion; MiVL: Mitral valve lesion; MVL: Multiple valve lesion; AoVL: Aortic valve lesion

References

1. Bland EF, Jones TD. Rheumatic fever and rheumatic heart disease - A twenty year report on 1000 patients followed since childhood. Circulation 1951;4:836-43.
2. Padmawati S. Rheumatic fever and rheumatic disease in developing countries. Bulletin of the World Health Organization 1978;56:543-

50.
3. Stollerman GH. Rheumatic fever and other rheumatic diseases of the heart. In: Braunwald E, ed. Heart disease: a textbook of cardiovascular medicine, 4th ed. Philadelphia: WB Saunders, 1992. p. 1721–41.
4. Gordi L, Lilienfeld A, Rodrigues R. Studies in epidemiology and preventability of rheumatic fever. I. Demographic factors and the incidence of acute attacks. J Chron Dis 1969;21;645-54.
5. Cheadle WR. Harveian lectures on the various manifestations of the rheumatic state as exemplified in childhood and early life. Lancet 1889;371:821-7.
6. Paul JR. The epidemiology of rheumatic fever. Am J Public Health Nations Health 1941;31:611–8.
7. Pickles WN. A rheumatic family. Lancet 1943;2:241.
8. Zinsser H, Yu H. The bacteriology of rheumatic fever and the allergic hypothesis. Arch Intern Med 1928;42:301-9.
9. Guilherme L, Weidebach W, Kiss MH, Snitcowsky R, Kalil J. Association of human leukocyte class II antigen with rheumatic fever or rheumatic heart disease in Brazilian population. Circulation 1991;83:1995-8.
10. Ozkan M, Carin M, Sonmez G, Senocak M, Ozdemir M, Yakut C. HLA antigens in Turkish race with rheumatic heart disease. Circulation 1993;87:1974-8.
11. Anastasiou-Nana MI, Anderson JL, Carlquist JF, Nanas JN. HLA-DR typing and lymphocyte subset evaluation in rheumatic heart disease: a search for immune response factors. Am Heart J 1986;112:992-7.
12. Guilherme L, Faé K, Oshiro SE, Kalil J. Molecular pathogenesis of rheumatic fever and rheumatic heart disease. Expert Rev Mol Med 2005;7:1-15.
13. Patarroyo ME, Winchester RJ, Vejerano A, Gibofsky A, Chalem F, John B, et al. Association of a B-cell alloantigen with susceptibility to rheumatic fever. Nature 278:173-4.
14. Khanna AK, Buskirk DR, Williams RC jr., Gibofsky A, Crow MK, Menon A, et al. Presence of a Non-HLA B Cell Antigen in Rheumatic Fever Patients and Their Families as Defined by a Monoclonal Antibody. J Clin Invest 1989;83:1710-6.
15. Harel L, Zeharia A, Kodman Y, Straussberg R, Zabriskie JB, Amir J. Presence of the d8/17 B-cell marker in children with rheumatic fever in Israel. Clin Genet 2002;61:293-8.
16. Harrington Z, Visvanathan K, Skinner NA, Curtis N, Currie BJ, Carapetis JR. B-cell antigen D8/17 is a marker of rheumatic fever susceptibility in Aboriginal Australians and can be tested in remote settings. Med J Aust 2006;184:507-10.
17. Regelmann WE, Talbot R, Cairns L, Martin D, Miller LC, Zabriskie JB, et al. Distribution of cells bearing "rheumatic" antigens in peripheral blood of patients with rheumatic fever/rheumatic heart disease. J Rheumatol 1989;16:931-5.
18. Kaplan MH, Suchy ML. Immunological relation of streptococcal and tissue antigens II. Cross-reactions antisera to mammalian heart tissue with cell wall constituent of certain strains of group A streptococci. J Exp Med 1964;119:643–50.
19. Kaplan MH, Svec KH. Immunologic relation of streptococcal antibody cross-reactive with heart tissue: association with streptococcal infection, rheumatic fever and glomerulonephritis. J Exp Med 1964;19:51–66.
20. Chou HT, Tsai CH, Chen WC, Tsai FJ. Lack of association of genetic polymorphisms in the interleukin-1beta, interleukin-1 receptor antagonist, interleukin-4, and interleukin-10 genes with risk of rheumatic heart disease in Taiwan Chinese. Int Heart J 2005;46:397–406.
21. Schoen FJ, St. John Sutton M. Contemporary pathologic considerations in valvular disease. In: Virmani B, Atkinson JB, Feuoglio JJ, edi. Cardiovascular Pathology. Philadelphia: Saunders; 1991. p. 334-53.
22. Kawanishi DT, Rahimtoola SH. Mitral stenosis. In: Rahimtoola SH, edi. Valvular Heart Disease. II. St. Louis: Mosby; 1996. p. 8.1-8.24.
23. Wu W, Ke D, Xu CX, Deng YL, Chen L, Zhang JC, et al. Collagen type I and interleukin-1 beta gene expression in human atria during atrial fibrillation. Zhonghua Nei Ke Za Zhi 2006;45:807-10.
24. Fraser WJ, Haffejee Z, Jankelow D, Wadee A, Cooper K. Rheumatic Aschoff nodules revisited. II. Cytokine expression corroborates recently proposed sequential stages. Histopathology 1997;31:460–4.

Cite this article as: Gupta A, Singh AK. Association of IL1- α 889CT with clinical outcomes of rheumatic heart disease. Int J Med Sci Public Health 2014;3:1192-1195.
Source of Support: Nil
Conflict of interest: None declared