Human Immunology 77 (2016) 555–558

Contents lists available at ScienceDirect

journal homepage: www.elsevier.com/locate/humimm

HLA-DRB1 in Henoch-Schönlein purpura: A susceptibility study from North India

Ritu Aggarwal [a,*], Anju Gupta [b], Jasmine Naru [a], Noureddine Berka [c], Neha Nanda [a], Deepti Suri [b], Surjit Singh [b]

[a] Department of Immunopathology, Post Graduate Institute of Medical Education and Research, Chandigarh, India
[b] Department of Pediatrics, Post Graduate Institute of Medical Education and Research, Chandigarh, India
[c] Department of Pathology and Laboratory Medicine, University of Calgary, Calgary, Canada

ARTICLE INFO

Article history:
Received 2 January 2015
Revised 3 July 2015
Accepted 12 May 2016
Available online 13 May 2016

Keywords:
Human leucocyte antigen
IgA vasculitis
Nephritis
Pathogenesis
Polymerase chain reaction

ABSTRACT

Etiology of Henoch-Schönlein purpura (HSP) a small vessel vasculitis remains elusive. Susceptibility may be conferred by major histocompatibility complex. There are limited reports on the association of human leucocyte antigens (HLA) and HSP. The aim was to investigate the association of HLA-DRB1 (HLA class II antigen) with HSP. Forty three patients, <14 years, who fulfilled the diagnostic criteria of HSP, laid by 'European League Against Rheumatism' were enrolled. Fifty four age-matched, healthy controls were included. PCR with 24 sequence specific primers for HLA-DRB1 allotypes was performed. Commercially available HLA-DR tissue typing kit (Inno-train, Kronberg im Taunus, Hesse, Germany) was utilized. The mean age of patients and controls was 8.5 ± 3.2 and 7.6 ± 3.6 years, respectively ($p = 0.18$). Gastrointestinal and renal involvement was observed in 25 (58.1%) and 14 (32.6%) patients, respectively. A greater frequency of HLA-DRB1*11 was observed in patients (11.6%) as compared to controls (1.9%), however, the results were not significant following Bonferroni correction. No significantly protective HLA genotype was observed. None of the HLA-DRB1 antigen was noted to increase the susceptibility to gastrointestinal or renal involvement. In conclusion, in the first study from India, none of the HLA-DRB*1 genotypes were observed to increase the susceptibility of North Indian children to HSP.
© 2016 American Society for Histocompatibility and Immunogenetics. Published by Elsevier Inc. All rights reserved.

1. Introduction

Henoch-Schönlein purpura (HSP) is a small vessel vasculitis typically observed in children 3–10 years old. Recently renamed immunoglobulin A vasculitis, it is the most common form of systemic vasculitis in children [1]. A multisystem disease, it can affect the skin, joints, gastrointestinal tract and kidneys. It is characterized by non-thrombocytopenic palpable purpura, abdominal pain, arthritis and glomerulonephritis [2]. It tends to be self-limiting in majority, except for serious gastrointestinal or renal involvement. In a study from the UK, HSP was reported to be more common in Asians [3]. The incidence was 24.0, 17.8 and 6.2 per 100,000 children per year in Asians, Whites and Blacks, respectively [3].

Although the pathogenesis of HSP remains elusive, it is accepted to be an immune mediated disease characterized by deposition of IgA containing immune complexes [1]. It is hypothesized that HSP is triggered in genetically susceptible individuals by exposure to several environmental factors, including infections [4]. A seasonal predilection has been demonstrated in a series of 209 children with HSP from our center [5]. An increased familial occurrence suggests genetic predisposition. The genetic basis of susceptibility to HSP may be conferred by a number of gene loci, including the major histocompatibility complex [6]. There is limited literature on the association of susceptible human leucocyte antigens (HLA) and HSP. An association with HLA-DRB1*01 phenotype has been suggested in Caucasian individuals with HSP [7,8]. In a recent large study of 342 patients with HSP from Spain, a significant increase of HLA-DRB1*01 when compared with controls was found (43% vs 7%, respectively; $p < 0.001$) [9]. The association has not been explored in Asian-Indians. The study was conducted to investigate the HLA-DRB1 locus in Indian children with HSP.

2. Materials and methods

Patients who had been diagnosed with HSP, as per the European League against Rheumatism criteria [10], and on follow up in the Pediatric Rheumatology clinic of the Institution were enrolled

* Corresponding author.
E-mail address: ritu_immunopath@yahoo.co.in (R. Aggarwal).

http://dx.doi.org/10.1016/j.humimm.2016.05.009
0198-8859/© 2016 American Society for Histocompatibility and Immunogenetics. Published by Elsevier Inc. All rights reserved.

randomly. The patients were included in the study at a variable duration from diagnosis. Age (but not sex) matched, healthy controls, were enrolled as well. The controls were not enrolled from the pediatric rheumatology clinic and were not related to the patients. The study was approved by the institute's ethics committee. An informed consent was obtained from parents/guardians.

2.1. DNA extraction

One ml of blood in EDTA was collected from the patients as well as controls. DNA was extracted with commercially available DNA isolation kit (Qiagen, Valencia, CA, USA). Quantity of the extracted DNA was checked by spectrophotometry. Polymerase chain reaction (PCR) for β-actin gene was performed in each sample to check the quality of DNA. Tissue typing by PCR was performed on DNA that was satisfactory in quantity and quality.

2.2. Determination of HLA-DRB1 allotypes

HLA-DRB1 typing by PCR amplification with sequence-specific primers was performed. Commercially procured HLA-DRB1 tissue typing kits (Inno-Train Diagnostik GmbH, Kronberg im Taunus, Germany) were utilized. The PCR was performed according to the manufacturer's instructions. The kit contained a set of 24 sequence specific primers (SSP). The PCR master mix was prepared with the following constituents: Ready PCR mix 84 µl, Taq polymerase (5 U/µl) 2.2 units, distilled water 168 µl and DNA 26 µl. The thermocycling conditions followed were: 96 °C for 2 min, followed by 96 °C for 15 s and 65 °C for 60 s for 10 cycles, followed by 96 °C for 15 s, 61 °C for 50 s and 72 °C for 30 s for 20 cycles. The PCR products were resolved on 2% agarose gel electrophoresis and visualized under ultraviolet light.

2.3. Statistical analysis

The strength of association of HLA-DRB1 with HSP was evaluated using odds ratio and corresponding 95% confidence intervals. The p values were determined using chi-square or Fisher exact test. The Fischer exact test was utilized if the expected value was less than 5. A $p \leqslant 0.05$ was considered significant. The results were adjusted for Bonferroni correction. Analyses were performed with GraphPad Software (GraphPad Software, Inc., CA, USA) and SPSS software (version 20.0, 2011; SPSS, Inc., Chicago, Illinois, USA).

3. Results

Forty three patients and 54 healthy controls were enrolled. The mean age of the patients and controls was 8.5 ± 3.2 years (range: 3–14) and 7.6 ± 3.6 years (range: 1–14), respectively (p = 0.18). The male: female ratio of the patients and controls was 3.8:1 and 1.6:1, respectively (p = 0.08). Twenty five (58.1%) patients had gastrointestinal symptoms, including nine patients with gastrointestinal bleeding. Renal involvement was observed in 14 (32.6%) patients; none had renal insufficiency. Thirty five (81%) had received steroids.

The frequency of the HLA-DRB1 allotypes is illustrated in Table 1. The allotypes with the highest frequency in patients included, HLA-DRB1*15 (28%), HLA-DRB1*11 (11.6%) and HLA-DRB1*04 (11.6%). A greater frequency of HLA-DRB1*11 was observed in patients (11.6%) as compared to controls (1.9%) (OR = 6.97 [95%CI: 1.48–32.74], p = 0.014). However, the results were not significant following Bonferroni correction. None of the allotypes were observed to be significantly protective for HSP. No HLA-DRB1 allotypes was noted to increase the susceptibility to gastrointestinal or renal involvement (Tables 2 and 3).

4. Discussion

The pathogenesis of HSP remains elusive; Immunologic, genetic and environmental factors appear to play a role [7]. The disease is characterized by deposition of IgA dominant immune complex, infiltration of small blood vessels with polymorphonuclear leucocytes, increased IgA and pro-inflammatory cytokines that suggest HSP to be an immune mediated disease. The genetic susceptibility to HSP may be deliberated by numerous gene loci, including the major histocompatibility complex [6]. HLAs that play an important role in antigen presentation and immune responses may contribute to genetic susceptibility. Recent reports propose that HSP may be precipitated in genetically predisposed individuals by several environmental stimuli, including infections [4]. Researchers in the past have attempted to explore several HLA antigens belonging to HLA class I (HLA-A and HLA-B antigens) and class II (HLA-DR and HLA-DQ antigens) as plausible susceptibility or protective factors in the pathogenesis of HSP. Data on the implication of HLA antigens with HSP has been reported from limited countries, including Italy, Spain and Turkey [7–9,11]. Effect of HLA class I antigens on susceptibility and severity of HSP have been inconsistent. Salient results of the reports on the association of HLA-DRB1 in patients with HSP are illustrated in Table 4.

We did not observe any of the HLA-DRB1 allotypes to be significantly associated with childhood HSP as compared to controls. The odds ratio (6.97) of DRB1*11 in our study is greater than earlier studies, however, the 95% confidence interval (1.48–32.74) is wide, in all probability, due to a limited sample size.

Amroso et al. in 1997, reported an increased frequency of HLA-DRB1*11 along with HLA-DRB1*01 in an Italian cohort of 152

Table 1
HLA-DRB1 frequency in patients and controls.

Allotypes	Patients (n = 43) Genes = 86 (%)	Controls (n = 54) Genes = 108 (%)	OR (95% CI)	Adjusted OR (95% CI) for sex	P value*	P value adjusted for sex*	Odds ratio**
DRB1*01	3 (3.5)	2 (1.9)	1.92 (0.31–11.73)	1.620 (0.247–10.601)	0.66	0.615	0.565
DRB1*03	8 (9.3)	13 (12)	0.75 (0.30–1.90)	0.381 (0.107–1.362)	0.65	0.138	0.082
DRB1*04	10 (11.6)	4 (3.7)	3.42 (1.03–11.32)	3.322 (0.922–11.964)	0.06	0.066	0.093
DRB1*07	6 (7)	14 (13)	0.50 (0.19–1.37)	0.795 (0.242–2.609)	0.24	0.705	0.705
DRB1*08	1 (1.2)	1 (0.9)	1.26 (0.78–20.42)	–	1.00	–	–
DRB1*09	1 (1.2)	6 (5.6)	0.2 (0.02–1.69)	0.133 (0.015–1.186)	0.14	0.071	0.095
DRB1*10	6 (7)	12 (11.1)	0.6 (0.22–1.67)	0.393 (0.109–1.410)	0.46	0.152	0.128
DRB1*11	10 (11.6)	2 (1.9)	6.97 (1.48–32.74)	5.972 (1.175–30.342)	0.014	0.031	0.025
DRB1*12	1 (1.2)	Nil	–	–	0.45	–	–
DRB1*13	8 (9.3)	19 (17.6)	0.48 (0.20–1.16)	0.928 (0.310–2.784)	0.21	0.894	0.972
DRB1*14	7 (8.2)	12 (11.1)	0.71 (0.27–1.89)	0.574 (0.194–1.694)	0.63	0.315	0.291
DRB1*15	24 (27.9)	20 (18.5)	1.70 (0.87–3.35)	2.280 (0.897–5.792)	0.24	0.083	0.079
DRB1*16	1 (1.2)	3 (2.8)	0.41 (0.04–4.03)	0.413 (0.039–4.339)	0.63	0.461	0.375

* Significant corrected p value is 0.0038 (Bonferroni correction).
** Mantel-Haenszel test.

Download English Version:

https://daneshyari.com/en/article/3349494

Download Persian Version:

https://daneshyari.com/article/3349494

Daneshyari.com