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CLINICAL & BASIC RESEARCH

The Association between Human Leukocyte Antigens and Hypertensive End-Stage Renal Failure among Yemeni Patients

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العلاقة بين انواع مُسْتَضِدَّاتُ الكُرَيَّاتِ البيضِ البَشَرِيَّة في مرض المرحلة الاخيرة من الفشل الكلوي المصاحب لارتفاع ضغط الدم بين المرضى اليمنيين

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ABSTRACT: ***Objectives:*** Many studies have attempted to locate a connection between various genetic factors and the pathogenesis of certain diseases. A number of these have found human leukocyte antigens (HLAs) to be the most significant genetic factors affecting the susceptibility of an individual to a certain disease. The present case-control study aimed to determine the connection between class I and class II HLAs and cases of hypertensive end-stage renal failure (HESRF), as contrasted with healthy controls, in Yemen. ***Methods:*** The study was carried out between March 2013 and March 2014 and included 50 HESRF patients attending the Urology & Nephrology Center at Al-Thawra University Hospital in Sana'a, Yemen, and 50 healthy controls visiting the same centre for kidney donation. Among both patients and controls, HLA class I (A, B and C) and class II (*DRB1*) genotypes were determined by polymerase chain reactions. ***Results:*** There was an association (odds ratio: 4.0) with *HLA-A9(24)* and HESRF, although this was not statistically significant. A significant protective function was found for the *HLA-CW3* and *DRB1-8* genes against the development of HESRF. Although *HLA-B14* was present in some patients (0.06) and not in the controls, this difference was not statistically significant enough to conclude that *HLA-B14* plays a role in the genetic predisposition for end-stage renal disease development. There was a high frequency of *HLA-A2, B5, CW6, DRB1-3, DRB1-4* and *DRB1-13* in both patients and controls. ***Conclusion:*** Although no HLAs were found to play a highly significant role in genetic predisposition to HESRF, certain HLA genes could be considered as protective genes against HESRF development.

Keywords: Hypertension; Renal Failure, End-Stage; HLA Antigens; Case-Control Study; Yemen.

الملخص: الهدف: حاولتْ العديد منْ الدراسات تَحديد العلاقة بين العوامل الوراثية المُخْتَلفة وحدوث بَعْض الأمراض. وَجد عدد منْ هذه الدراسات علاقة بين انواع مُسْتَضِدَّاتُ الكُرَيَّاتِ البيضِ البَشَرِيَّة وتعرضَ بعض الافراد بَصورة أكبر للاصابة ببعَض الامراض. هدّفتْ الدراسةُ لتَقْرير العلاقة بين الصنفِ الاول والصنفِ الثّاني من مُسْتَضِدَّاتُ الكُرَيَّاتِ البيضِ البَشَرِيَّة وحدوث الفشل الكلوي المصاحب ارتفاع ضغط الدم بين المرضَى و مقارنةَ ذلك بانتشار هَذه المُسْتَضِدَّاتُ بَين الاصحاءَ كعينةَ للمقارنَة. **الطريقة:** نُفّذتْ الدراسة بين مارس2013 وآذار 2014 وتَضمّنتْ 50 من مرضى الفشل الكلوي المصاحبَ لارتفاع ضغط الدم الذين وصلوا الى مركزَ طبّ الكلى وطبّ المجاري البوليةَ في مَستشفى الثورة الجامعي في صنعاء، اليمن، و 50 من الاصحاء الذين راجعو نفس المركز للتبرّع بالكلى. تم تصنيف جينات الصنف الاول والصنف الثّاني من مُسْتَضِدَّاتُ الكُرَيَّات البيضِ البَشَرِيَّة بين كلا من المرضى والاصحاء بطَريقة تفَاعلات بُوليميراز المتسلسلةَ. **النتائج:** كان هَناك علاقة (لَيسَتَ ذو دلالة إحصائية) بنسبة إحتمالات: 4.0 بين HLA-A9(24) و HESRF، كما وجدتِ وَ ظيفة و قائيَة هامّة لجينات HLA-CW3 و DRB1-8. وبالرغم من أن HLA-B14 جين كانَ موجود في بَعْض المرضى (0.06) ولَم يكن موجدَ في الاصحاء الا ان هذا الإختلاف لما يكنَ هامَّ بشكل إحصائي بما فيه الكفاية لإسْتنْتاج الميل الوراثي لتطوير المرض الكلوي النهائي بسبب وجود هذا الجين. كان هناك تكرار إتش إل أي أي-2 (HLA-A2)، إتش إل أي بي (HLA-B5)، إتش إل أي سَي دبليَو (HLA-CW6) وإتش إل أي دي آر بي (HLA-DRB1-3, HLA-DRB1-4 و HLA-DRB1-13) في كلا من المرضى و عينة المقارنة من الاصحاء. **الخاتمة:** بالرغم من أن الدراسة لم تثبت وجود علاقة ذات اهمية بين نشوء الفشل الكلوي وبعض انواع مُسْتَضِدَّاتُ الكُرَيَّات البيض البَشَرِيَّة الا اننا يُمْكِنُ أنْ تُعتَبرَ أنَ بعض الجينات من اللمكن ان تكون لها وظيفة وقائية ضدّ نشوء الفشل الكلوي المَصاحب لارتفَاع ضغَط الدم.

مفتاح الكلمات: إرتفاع ضغط الدم؛ الفشل الكلوي، مرحلة نهائية؛ مُسْتَضِدَّاتُ الكُرَيَّاتِ البيضِ البَشَرِيَّة؛ دراسة مقارنة؛ اليمن.

Advances in Knowledge

- *This study gives information about human leukocyte antigens (HLA) in individuals from Yemen and investigates the relationship between HLAs and hypertensive end-stage renal failure (HESRF).*
- *This study provides important information as it is the first time that the frequency of HLA genes in the general Yemeni population and in HESRF Yemeni patients has been investigated.*

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- *The findings revealed an association between HLA-A9(24) and HESRF and a highly significant protective function for the HLA-CW3 and DRB1-8 genes against HESRF development.*

Application to Patient Care

- *Research in the HLA field is still in its early stages in Yemen. It is therefore necessary to study the relationship between HLAs and various diseases in Yemen in order to improve treatment options for affected patients.*
- *The incidence and social burden of end-stage renal disease is growing. Slowing or preventing the progression of this disease to renal failure is an important public health priority.*

The progression of chronic renal failure (CRF) usually leads to end-stage renal disease (ESRD), at which point renal replacement therapy is the only option. Since the mid-1980s, there has been a noticeable rise in the incidence and prevalence of ESRD around the world.[1–4] Gender, genetic profile, ethnicity, lipids, hypertension and smoking have been identified as factors which can influence the development of ESRD.[4,5] However, there is a need for further research to be carried out on the role of the immune system in renal diseases, as this could be the origin or cause of the disease and its progression.[6] This theory is the outcome of investigations into positive relations between human leukocyte antigens (HLAs) and a broad variety of renal diseases.[6] Studies have illustrated some significant associations between HLA class I and II alleles with renal diseases. Methods for associating HLAs with renal disease have been developed since the late 1980s, mostly as a result of more detailed knowledge of class I and II molecules and their structure and function. HLA phenotypes are interrelated, with an increased or reduced risk of alloantibody sensitisation in ESRD candidates for first or repeat kidney transplantation.[6]

The incidence and prevalence of ESRD and its relationship with class I and II HLA alleles in patients from Yemen and other Middle Eastern and Arab countries has only recently been studied due to deficiencies in national epidemiological research. Prior to this study, no information was available on the frequency of class I and II HLAs, either in the Yemeni population in general or in hypertensive end-stage renal failure (HESRF) patients. As such, this case-control study aimed first to determine the relationship between HLA class I (A, B and C) and class II (DRB1) with HESRF. Second, it aimed to compare the HLAs found to have a relationship with HESRF with those from other races or national groups outside of Yemen in a literature review. Third, this study was designed to determine the potential genes that might give protection from HESRF development.

Methods

This case-control study was carried out over a 12-month period between March 2013 and March 2014. A total of 100 individuals were enrolled in the study; the patient group comprised 50 adults with HESRF while the control group consisted of 50 healthy adult individuals. All patients and controls originated from Sana'a, Yemen. The patient group consisted of 40 males and 10 females (aged from 18 to 57 years) who had attended the Urology & Nephrology Center at Al-Thawra University Hospital in Sana'a for a kidney transplant. Hypertension was the cause of renal failure in all of these patients. All of the patients showed proteinuria and underwent a kidney biopsy to exclude an extensive focal and global glomerulosclerosis association with the *APOL1* gene. All HESRF patients in the study were on haemodialysis before they underwent kidney transplantation.

The control group consisted of 32 males and 18 females (aged from 18 to 57 years) who had visited the Urology & Nephrology Center at Al-Thawra University Hospital for a kidney donation. They were confirmed to be healthy following a clinical examination by specialist physicians. As age and gender does not influence an individual's HLA frequency profile, the control group was not age- and gender-matched with the patient group. Both the patients and controls were required to undergo certain tests, including blood pressure, abdominal ultrasound, complete blood count, blood sugar, kidney and liver function tests. In addition, a general urine examination and a 24-hour urine examination for protein detection and creatinine clearance were carried out. Results for all of the aforementioned tests were normal in the control group.

HLA genotyping was completed for each individual in the patient and control groups. Blood specimens were collected and treated with ethylenediaminetetraacetic acid. As the time of blood collection has no effect on genotyping results, blood specimens were collected whenever patients arrived at the hospital. Blood specimens from both groups were used for HLA class I (A, B and C) and class II (DRB1) genotyping.

The typing of all of the subjects' *HLA-A*, *-B*, *-C* and *-DRB1* alleles were identified using low-resolution BAGene Sequence Specific Primers polymerase chain reaction kits (BAG Health Care GmbH, Lich, Germany). The tests were carried out according to the manufacturer's instructions. The genomic DNA of each

Table 1: Age and gender of Yemeni hypertensive-end stage renal failure cases and controls

Characteristic	Patient group		Control group		Total	
	n	%	n	%	n	%
Age in years						
18–27	35	70	31	62	66	66
28–37	8	16	12	24	20	20
38–47	5	10	6	12	11	11
48–57	2	4	1	2	3	3
Gender						
Male	40	80	32	64	72	72
Female	10	20	18	36	28	28
Total	**50**	**100**	**50**	**100**	**100**	**100**

sample was purified using the spin columns method of the QIAGEN DNA purification kit (QIAGEN, Hilden, Germany). Allele frequencies in HESRF patients and controls were calculated by direct counting.[7]

The association of HESRF with *HLA-I* alleles was analysed by comparing the frequency of *HLA-A* and *-B* alleles in the HESRF patients with those in the 50 healthy controls. The maximum likelihood method was used to evaluate the haplotype frequencies for the two *loci* of HLA. The Chi-squared test for two-way tables after Yates correction for continuity was used to define the differences between allele frequencies in patients and controls, using the Epi Info™ programme, Version 6 (Centers for Disease Control and Prevention, Atlanta, USA). The odds ratios (OR) with 95% confidence intervals (CI) were calculated. A *P* value of <0.05 was considered significant.

The study was approved by the Department of Medical Microbiology & Clinical Immunology in the Faculty of Medicine & Health Sciences at Sana'a University, Yemen. Written consent was obtained from all of the participants in the study.

Results

The age and gender of the subjects in the patient and control groups are shown in Table 1. The HESRF patients' mean age was 38.03 ± 10.9 years and that of the controls was 35.15 ± 8.9 years (range: 18–57 years for both groups). A total of 40 patients (80%) were male and the remaining 10 patients (20%) were female. There were 32 male (64%) and 18 female (36%) controls [Table 1].

The results showed that *HLA-A2* and *A9* had the highest frequency among *HLA-A* alleles in the patients (0.48 and 0.18); *HLA-A2* and *A3* were most frequent among the controls (0.48 and 0.16). *HLA-A28* and *A1* were the next most frequent alleles in the patient group, whereas *HLA-A28* and *A9* were the next most frequent alleles in the controls. A comparison of the frequency of *HLA-A* alleles in the patients and controls showed higher frequencies of *HLA-A9(24)* (OR = 4.0, 95% CI = 0.42–103.9; *P* = 0.16) and *A9* (OR = 1.98; *P* = 0.24) and lower frequencies of *HLA-A19(30)* (OR = 0.0; *P* = 0.04) and *A19(33)* (OR = 0.23; *P* = 0.16) in the HESRF patients [Table 2].

The most numerous *HLA-B* allele in the patients was *HLA-B5*. This allele was articulated in the HESRF patients at a higher frequency in comparison to the controls (0.26 versus 0.18; OR = 1.65; *P* = 0.33). On the other hand, the *HLA-B16(38)*, *21(49)*, *70* and *73* alleles were among the least expressed *HLA-B* alleles in the patients. A total of 10 alleles were not found at all in the control group (*B14*, *21[44]*, *21[45]*, *27*, *40*, *40[60]*, *50*, *51*, *53*, *62* and *78*). The most frequent alleles expressed in the control subjects was *HLA-B5* (OR = 1.65; *P* = 0.33) and *B35* (OR = 0.46; *P* = 0.21). *HLA-B21* and *21(50)* were also observed at a lower frequency in the patients compared to the controls (OR = 0.47; *P* = 0.29 each) [Table 3].

The results showed that *HLA-CW6* and *CW7* had the highest frequency among *HLA-CW* alleles in the patients (0.44 and 0.38, respectively) and the controls (0.52 and 0.38, respectively). *HLA-CW4* and *CW2* were the next most frequent alleles in the patients (0.28 and 0.12, respectively); *HLA-CW4* and *CW3* were next most frequent in the controls (0.26 and 0.14, respectively). In comparison, the *HLA-CW3* allele was not found among the patients (*P* = 0.006), indicating a probable protective function [Table 4].

The results showed that *DRB1-4* and *13* had the highest frequency among *DRB1* alleles in the patients (0.32 and 0.34, respectively) and controls (0.42 and 0.30, respectively). *DRB1-3* was the next most frequent allele in the patients and controls (0.26 each). A comparison of the frequency of *DRB1* alleles among those in the patient and control groups showed higher frequencies of *DRB1-8* in the controls (0.12), while in the patients it was 0.02 (OR = 0.15; *P* = 0.05), indicating a probable protective function. In addition, a comparison of the frequency of *DRB1* alleles between both groups showed higher frequencies of *DRB1-10* (OR = 2.3, 95% CI = 0.71–7.8; *P* = 0.11) and *15* (OR = 2.3, 95% CI = 0.63–8.4; *P* = 0.16) and lower frequencies of *8* (OR = 0.15; *P* = 0.05) in the HESRF patients [Table 5].

Discussion

Renal failure is mainly caused by a patient suffering from chronic high blood pressure over a period of

Table 2: Human leukocyte antigen-A (HLA-A) and gene frequency in Yemeni hypertensive-end stage renal disease patients and healthy controls (N = 100)

HLA-A	Patients			Controls			Odds ratio (95% confidence interval)	*P* value*
	n	Antigen frequency	Gene frequency	n	Antigen frequency	Gene frequency		
1	7	0.14	0.072	4	0.08	0.040	1.9 (0.5–8.3	0.33
2	24	0.48	0.279	24	0.48	0.279	1 (0.4–2.4)	0.84
3	3	0.06	0.030	8	0.16	0.083	0.34 (0.1–1.5)	0.11
9	9	0.18	0.094	5	0.10	0.051	1.98 (0.54–7.5)	0.24
9(23)	5	0.10	0.051	3	0.06	0.030	1.74 (0.3–9.8)	0.48
9(24)	4	0.08	0.040	1	0.02	0.010	4.0 (0.42–103.9)	0.16
10	1	0.02	0.010	4	0.08	0.040	0.23 (0.01–2.3)	0.18
10(25)	1	0.02	0.010	0	0.00	0.000	Undefined	0.3
10(26)	1	0.02	0.010	0	0.00	0.000	Undefined	0.3
11	2	0.04	0.020	3	0.06	0.030	0.65 (0.1–5.1)	0.6
19	3	0.06	0.015	5	0.10	0.051	0.6 (0.1–2.99)	0.46
19(24)	1	0.02	0.010	0	0.00	0.000	Undefined	0.31
19(29)	1	0.02	0.010	1	0.02	0.010	1 (0.0–37.8)	0.84
19(30)	0	0.00	0.000	4	0.08	0.040	0.0 (0.0–1.5)	0.04
19(31)	0	0.00	0.000	2	0.04	0.020	0.0 (0.0–4.1)	0.15
19(32)	3	0.06	0.030	2	0.04	0.020	1.5 (0.2–13.8)	0.64
19(33)	1	0.02	0.010	4	0.08	0.040	0.23 (0.01–2.37)	0.16
23	0	0.00	0.000	2	0.04	0.020	0.0 (0.0–4.1)	0.15
24	0	0.00	0.000	2	0.04	0.020	0.0 (0.0–4.1)	0.15
28	8	0.16	0.083	6	0.12	0.062	1.4 (0.4–5.03)	0.56
29	2	0.04	0.020	0	0.00	0.000	Undefined	0.15
30	2	0.04	0.020	4	0.08	0.04	0.48 (0.06–3.2)	0.4
31	1	0.02	0.010	0	0.00	0.000	Undefined	0.31
32	2	0.04	0.020	1	0.02	0.010	2.04 (0.14–58.9)	0.55
33	3	0.06	0.030	5	0.10	0.051	0.6 (0.1–2.99)	0.46
36	0	0.00	0.000	1	0.02	0.010	0.0 (0.0–17.6)	0.31
80	0	0.00	0.000	1	0.02	0.010	0.0 (0.0–17.8)	0.31

**P <0.05 was considered statistically significant.*

many years. Hypertension is also the second main cause, after diabetes, of ESRD and is accountable for 25–30% of all reported cases.[8] Hypertension is an important risk factor for the progression of ESRD in women as well as men.[9] Certain ethnicities, such as those of black African origin, have a high prevalence of the more severe form of hypertension; in addition, these patients have a higher prevalence of hypertension occurring at an earlier age than patients of white Caucasian origin.[10] Essentially, hypertension suggests that at least one of the genes to blame for the genetic susceptibility to ESRD is to be found in or close to the HLA complex.[11,12]

Many studies have been performed worldwide on the HLA complex and disease, including in various Arab countries, but this type of research is still in its infancy in Yemen. Indeed, to the best of the authors' knowledge, the current study was the first in Yemen on HLA typing. The annual incidence of ESRD in Yemen is 120 per million which is comparable to the incidence reported in other countries of the same region.[13] This study aimed mainly to investigate the association

Table 3: Human leukocyte antigen-B (HLA-B) and gene frequency in Yemeni hypertensive end-stage renal disease patients and healthy controls (N = 100)

HLA-B	Patients			Controls			Odds ration (95% confidence interval)	*P* value*
	n	Antigen frequency	Gene frequency	n	Antigen frequency	Gene frequency		
5	13	0.26	0.139	9	0.18	0.094	1.65 (0.6–4.6)	0.33
5(51)	4	0.08	0.040	4	0.08	0.040	1.0 (0.2–5.1)	0.84
5(52)	2	0.04	0.020	1	0.02	0.010	2 (0.14–5.8)	0.55
7	3	0.06	0.030	5	0.10	0.051	0.6 (0.1–2.99)	0.46
8	4	0.08	0.040	3	0.06	0.030	1.4 (0.24–8.2)	0.7
12	1	0.02	0.010	2	0.04	0.020	0.49 (0.02–7.23)	0.55
12(44)	7	0.14	0.073	5	0.10	0.051	1.47 (0.38–5.8)	0.53
13	2	0.04	0.020	1	0.02	0.010	2.04 (0.14–5.8)	0.55
14	3	0.06	0.030	0	0.00	0.000	Undefined	0.07
16	1	0.02	0.010	3	0.06	0.030	0.32 (0.01–3.6)	0.3
16(38)	0	0.00	0.000	1	0.02	0.010	0 (0.0–17.5)	0.31
16(39)	1	0.02	0.010	2	0.04	0.020	0.49 (0.02–7.2)	0.55
17	6	0.12	0.062	6	0.12	0.062	1 (0.26–3.1)	0.84
17(57)	2	0.04	0.020	5	0.10	0.051	0.38 (0.05–2.3)	0.23
17(58)	1	0.02	0.010	1	0.02	0.012	1 (0.0–37.8)	0.84
18	5	0.10	0.051	3	0.06	0.030	1.7 (0.3–9.8)	0.46
21	3	0.06	0.030	6	0.10	0.060	0.47 (0.1–2.3)	0.29
21(44)	1	0.02	0.010	0	0.00	0.000	Undefined	0.31
21(45)	1	0.02	0.010	0	0.00	0.000	Undefined	0.31
21(49)	0	00.00	0.000	2	0.04	0.020	0.0 (0.0–4.1)	0.15
21(50)	3	0.06	0.030	6	0.12	0.062	0.47 (0.09–2.3)	0.29
22	1	0.02	0.010	1	0.02	0.010	1 (0.0–37)	0.84
27	1	0.02	0.010	0	0.00	0.000	Undefined	0.31
35	4	0.08	0.041	8	0.16	0.083	0.46 (0.11–1.8)	0.21
37	2	0.04	0.020	2	0.04	0.020	1 (0.1–10.5)	0.84
40	1	0.02	0.010	0	0.00	0.000	Undefined	0.31
40(60)	1	0.02	0.010	0	0.00	0.000	Undefined	0.31
41	4	0.08	0.041	4	0.08	0.041	1 (0.19–5.14)	0.84
50	1	0.02	0.010	0	0.00	0.000	Undefined	0.31
51	2	0.04	0.020	0	0.00	0.000	Undefined	0.15
52	1	0.02	0.010	0	0.00	0.000	Undefined	0.31
53	6	0.12	0.062	4	0.08	0.041	1.6 (0.4–7.2)	0.5
62	1	0.02	0.010	0	0.00	0.000	Undefined	0.31
70	0	0.00	0.000	1	0.02	0.010	0.0 (0.0–17.5)	0.31
72	1	0.02	0.010	2	0.04	0.020	0.49 (0.02–7.2)	0.55
73	0	0.00	0.000	1	0.02	0.010	0.0 (0.0–17.5)	0.31
78	2	0.04	0.020	0	0.00	0.000	Undefined	0.15

**P <0.05 was considered statistically significant.*

Table 4: Human leukocyte antigen-CW (HLA-CW) and gene frequency in Yemeni hypertensive end-stage renal disease patients and healthy controls (N = 100)

HLA-CW	Patients			Controls			Odds ratio (95% confidence interval)	*P* value*
	n	Antigen frequency	Gene frequency	n	Antigen frequency	Gene frequency		
1	1	0.02	0.010	1	0.02	0.010	1 (0.0–3.7)	0.84
2	6	0.12	0.062	6	0.12	0.062	1 (0.26–3.9)	0.84
3	0	0.00	0.000	7	0.14	0.072	0 (0.0–0.72)	0.006
4	14	0.28	0.151	13	0.26	0.139	1.1 (0.42–2.9)	0.82
5	1	0.02	0.010	3	0.06	0.030	0.3 (0.01–3.6)	0.30
6	22	0.44	0.252	26	0.52	0.307	0.73 (0.3–1.7)	0.42
7	19	0.38	0.213	19	0.38	0.213	1 (0.41–2.4)	0.84

**P <0.05 was considered statistically significant.*

between HLAs (A, B, CW and DRB1) and HESRF in comparison with healthy individuals.

In this study, the male-to-female patient ratio was 4:1. The high prevalence of renal disease among males might be due to physiological differences between the genders as well as the higher susceptibility of males to hypertension, which can lead to renal failure. The higher prevalence of hypertension among males in this study was in agreement with the findings of another study, which reported a male-to-female ratio of 2:1.[14] The present study also concurred with that of Hsu *et al*., who reported that the male gender was a risk factor for ESRD.[15] Another possible reason could be the statistically small sample size of females in the current study, partly due to the fact that Yemeni females have less access than males to medical services due to social and cultural reasons.[16]

The findings of this study revealed the phenotypic and gene frequencies of the *HLA-A*, *HLA-B*, *HLA-CW* and *HLA-DRB1* genes in 100 Yemenis (50 patients and 50 controls). *HLA-B14* was found in three patients, but was not identified in the controls. The patients, as compared to the controls, had lower frequencies of certain HLA genes such as *HLA-A19(30)*, *CW3* and *DRB1-8*. These genes were found in the controls but not in the patients, apart from *DRB1-8* which was found in one patient.

On the other hand, there was no statistically significant difference between the renal failure

Table 5: Human leukocyte antigen-DRB1 (HLA-DRB1) and gene frequency in Yemeni hypertensive end-stage renal disease patients and healthy controls (N = 100)

HLA-DRB1	Patients			Controls			Odds ratio (95% confidence interval)	*P* value*
	n	Antigen frequency	Gene frequency	n	Antigen frequency	Gene frequency		
1	7	0.14	0.072	9	0.18	0.094	0.74 (0.22–2.4)	0.58
3	13	0.26	0.139	13	0.26	0.139	1 (0.37–2.7)	0.84
4	16	0.32	0.175	21	0.42	0.238	0.65 (0.26–1.6)	0.3
7	7	0.14	0.073	9	0.18	0.094	0.74 (0.22–2.4)	0.58
8	1	0.02	0.010	6	0.12	0.060	0.15 (0.01–1.3)	0.05
10	12	0.24	0.128	6	0.12	0.060	2.3 (0.71–7.8)	0.11
11	5	0.10	0.051	3	0.06	0.030	1.74 (0.33–9.9)	0.46
12	0	0.00	0.000	1	0.02	0.010	0.0 (0.0–176)	0.31
13	17	0.34	0.188	15	0.30	0.163	1.2 (0.48–3.0)	0.66
14	3	0.06	0.030	1	0.02	0.010	3.13 (0.3–80.9)	0.30
15	10	0.20	0.106	5	0.10	0.051	2.3 (0.63–8.4)	0.16
16	4	0.08	0.040	5	0.10	0.051	0.78 (0.16–3.7)	0.72

**P <0.05 was considered statistically significant.*

patients and the controls; this suggests that there are no HLA genes predisposing to ESRD. This result was in agreement with those of Agrawal *et al*. and Prasanavar *et al*. who determined that HLA and haplotype frequencies were not significantly different in renal transplant patients and healthy donors.[17,18] This contrasts sharply with another study that compared race-matched controls of white Caucasian and black African ethnicity and showed how genetic differences associated with the HLA system accounted for racial differences in hypertensive renal failure.[19] A comparison between patients of white Caucasian and black African ethnicity with hypertensive renal failure demonstrated that the patients of black African origin had an increased frequency of *HLA-DR3* alleles, which was greater than that normally known to exist among healthy individuals.[19]

These outcomes differ from the results of the current study which found no statistically significant differences between the patients and controls. However, this might be due to the fact that patients and healthy controls in this study were genetically similar; in fact, some of the patients and controls were related by family. However, the authors of the current study were of the opinion that it can be helpful to compare relatives; if there is a higher frequency of similar genes it is easier to indicate susceptibility to the disease rather than to race. It may also be the case that patients and controls were subjected to similar racial, genetic and environmental factors that may contribute to the development of ESRD.

Zachary *et al*. also found that frequencies of HLA alleles and their distribution among donors and renal patients in the United Network for Organ Sharing (UNOS) registry varied between races.[20] However, no difference was found in disease susceptibility; this was in agreement with the current study. Similarly, several studies have indicated racial differences in HLAs and their relationships with many diseases.[4] The results of the current study were similar to those of Crispim *et al*. who described associations of class I and II HLAs with ESRD, independent of other factors, among CRF patients with a variety of diagnoses.[21] Crispim *et al*. found that the antigens positively associated with ESRD were HLA-A78 and HLA-DR11.[21]

In a previous study among Mapuche and non-Mapuche people in Chile, *HLA-B8* alleles were found significantly more frequently in Mapuche recipients than in non-Mapuche recipients and Mapuche donors.[22] This result differs to that of the current study, in which *HLA-B8* alleles were roughly similar in both the patients and the controls. On the other hand, there was no positive association of *HLA-A2* alleles with ESRD in the current study; this is similar to the findings reported among the Zulian population in Venezuela, in which *HLA-A2* alleles were also not associated with ESRD.[6]

The most numerous *HLA-B* allele in patients was *HLA-B5*. This allele was articulated in the HESRF patients at a higher frequency in comparison to the controls (0.26 versus 0.18). This result is different from that reported among the Zulian population in Venezuela, in which *HLA-B5* was less frequent in both ESRF patients and controls (3.9% and 2.4%, respectively).[6]

In the current study, a comparison of the frequency of *HLA-CW* alleles showed considerably higher frequencies of *HLA-CW3* among the controls than the patients ($P = 0.006$), indicating the protective function of *HLA-CW3*. This result is different from that reported in thyrotoxicosis patients, in whom *HLA-CW3* was found to be significantly increased in patients with endocrine ophthalmopathy (EOP) compared to thyrotoxic patients without EOP.[23] However, as yet, there have been no reported findings regarding the protective function of *HLA-CW3* alleles in renal diseases.

A comparison of the frequency of *DRB1* alleles in the patients and controls showed higher frequencies of *DRB1-8* in the control group than the patient group ($P = 0.05$), indicating the protective function of *DRB1-8*. This result is different to that reported among Hispanic renal recipient patients, in which *DRB1-8* was found to be significantly increased in patients with renal failure compared with healthy donors (15% versus 0%).[17]

The results of this study demonstrate that *HLA-A2*, *A9*, *B5*, *CW6*, *CW7*, *DRB1-4* and *DRB1-13* have very high frequencies among the HLA alleles in both patients and controls in the studied population. This result matches those of studies performed on the frequency of HLAs in Iraqi, Emirati and Lebanese populations.[24–26] In addition, comparing the antigen frequencies in the current study to those from kidney recipients and donors of other ethnic groups (i.e. African-Americans, Caucasians, Asians and Hispanics) included in the UNOS renal registry data on HLA-A, B and DR *loci* showed that, while the population in the current study differed from the other groups in certain respects, there was a similarity with Hispanics and Caucasians in the highest frequencies of HLA alleles.[17]

The diversity of the findings of different researchers may be due to the considerable irregularity in the frequency of HLA alleles present in different populations or ethnic groups. A further cause may be the unequal relationship between these HLA alleles and other nearby genes involved in accommodating the immune response. An example of this is reported by Ranganath *et al*.; they stated that polymorphisms

in genes encoding certain cytokines, including interleukin (IL)-6, IL-4 and tumour necrosis factor, may be affected in the progression to ESRD.[27]

This study had the limitation of being underpowered as only 50 patients were studied. However, it has laid the groundwork for future case-control studies in Yemen with larger sample sizes of cases and controls. These future studies should aim to confirm the connection between class I and II HLAs and cases of HESRF as compared with healthy controls.

Conclusion

This study found a high association between *HLA-A9(24)* and HESRF and a highly significant protective function for the *HLA-CW3* and *DRB1-8* genes against HESRF development. At the same time, the findings showed no significant role for other HLA molecules in the predisposition to developing ESRD among Yemeni patients. Although the *HLA-B14* gene was found only in the patients and not in the control group, this difference was not statistically significant enough to conclude that the *B14* gene is involved in a genetic predisposition to ESRD development. Certain HLA genes, such as *HLA-A2*, *B5*, *CW6* and *DRB1-3*, *DRB1-4* and *DRB1-13*, were found to have a high frequency in both the patients and the controls.

CONFLICT OF INTEREST

The authors declare no conflicts of interest.

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