HLA-A, B, Haplotypes in Normal Southern Chinese and a Lack of Associations in Renal Failure*

Soh Ha Chan, M.D., Ph.D.
G.B. Wee, M. Biol.
N. Srinivasan

SUMMARY **HLA locus A and B gene and haplotype frequencies from 578 haplotypes of normal southern Chinese in Singapore and Malaysia were documented. No difference was observed in comparing the HLA-A, B gene and haplotype frequency of patients with renal failure and that of normal subjects.**

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Over the last few years, there have been increasingly frequent reports on HLA and disease associations. In some of these reports, it has been suggested that disease associations may be stronger with haplotypes than with individual antigens. However, considerable statistical difficulties had been encountered when attempts were made to distingish haplotype associations from phenotype data. This is particularly so when an individual antigen association with a particular disease already exists.[1] Therefore, to study haplotype effects in disease associations, family studies must first be done in order to identify the haplotypes. In this paper we document the HLA locus A and B haplotype frequencies in the normal southern Chinese population of Singapore and Malaysia and report a lack of haplotype association in patients with renal failure. As far as we are aware, this is the first time that normal locus A and B haplotype frequencies in a Chinese population are being documented.

MATERIALS AND METHODS

All normal subjects and patients were ethnically southern Chinese (Cantonese, Hokkien and Teochew dialect groups) from Singapore and Malaysia; they were studied in family groups in order to assign haplotypes. A total of 289 normal subjects and 96 patients with renal failure on haemodialysis were studied and this generated 578 and 192 haplotypes respectively. The normal subjects, who had not been reported upon before, were spouses of patients in a kidney transplantation programme of Singapore and Malaysia and spouses of other diseased patients from Singapore who were involved in our family studies. The Singaporean and Malaysian Chinese originated from Kwantung and Fukien provinces in China. When studied separately, there were no differences in HLA gene and haplotype frequencies between normal Singaporean (n = 219) and Malaysian (n = 70) Chinese subjects and therefore we have combined the two groups in this report.

HLA typing

Lymphocyte separation and HLA typing were performed on freshly drawn venous blood as discussed previously.[2] The panel of antisera used to define the 26 locus A and B antigens consisted of over 200 antisera of Chinese, Malay, Japanese, Filipino and Caucasian origin. HLA gene frequencies were determined by gene counting from haplotypes. Delta values were calculated according to the formula $D = HF(Ob's) - g(a) \times g(b)$ where HF (Ob) is the observed haplotype frequency and g (a) and g (b) are the gene frequencies of alleles a and b.

RESULTS

Table 1 shows the gene frequencies of HLA locus A and B antigen of normal southern Chinese by gene counting of 578 haplotypes. Also shown in Table 1 are previously reported results of HLA locus A and B gene frequencies of a similar population calculated from phenotype data.[3] The gene frequencies of all locus A and B antigens, including blanks, by these two methods were remarkably similar.

Table 2 shows the observed and expected haplotype numbers in

*From WHO Immunology Research and Training Centre/Department of Microbiology, Faculty of Medicine, National University of Singapore, Singapore 0316.

Table 1 Comparison of HLA locus A and B gene frequencies in normal Chinese using gene counting of 578 haplotypes and estimation from phenotype data.

	Gene counting n = 578	Estimated from phenotype data* n = 238
A 1	.005	0.000
A 2	.322	.314
A 3	.005	.002
A 9	.175	.147
A 10	.017	.026
A 11	.336	.372
A 28	.002	.002
A 29	0.000	.006
AW 19	.109	.109
BL	.029	.022
B 5	.061	.065
B 7	.007	.008
B 8	.002	.002
B 12	.007	.017
B 13	.126	.107
B 14	0.000	0.000
B 15	.114	.118
B 17	.066	.074
B 18	.002	.008
B 27	.022	.036
B 37	0.000	.002
B 40	.253	.233
BW 16	.061	.056
BW 21	0.000	0.000
BW 22	.090	.063
BW 35	.040	.023
BW 46	.138	.121
BL	.017	.067

*Chan, *et al.* Tissue Antigens 1979; 13:361.

normal subjects together with the Chi-squared values (if $p<.01$). Table 3 lists the haplotypes that showed significant linkage disequilibrium together with haplotype frequencies and delta values. Again the haplotype frequencies were similar to the estimated haplotype frequencies reported previously.[3] For example, the frequency of A2 BW46 was .0830 and the previously estimated frequency from phenotype data was .0800. For AW19 B17 it was .0381 and .0311 respectively. There were no differences in the HLA, A and B gene frequency nor in haplotype frequency between normal subjects and patients with renal failure. This lack of association was observed even when Singaporean and Malaysian patients and controls were compared separately.

DISCUSSION

In this report, we have documented the haplotype frequencies of

Table 2 Observed and expected haplotypes in normal Chinese

	B5	B7	B8	B12	B13	B15	B17	B18	B27	B40	BW16	BW22	BW35	BW46	BL	Total
	*0	0	0	0	0	0	0	0	0	0	0	0	0	0	3	+3
A1	0.2	0.02	0.01	0.02	0.4	0.3	0.2	0.01	0.06	0.7	0.2	0.3	0.1	0.4	0.05	0.005
	–	–	–	–	–	–	–	–	–	–	–	–	–	–	117.5	
	8	1	0	0	21	11	2	0	1	45	25	16	4	48	4	186
A2	11.4	1.3	0.4	1.3	23.5	21.2	12.3	0.4	4.1	47.1	11.4	16.8	7.4	25.7	3.2	0.322
	–	–	–	–	–	8.2	12.2	–	–	–	26.3	–	–	32.9	–	
	0	0	0	2	0	1	0	0	0	0	0	0	0	0	0	3
A3	0.2	0.02	0.01	0.02	0.4	0.3	0.2	0.01	0.06	0.7	0.2	0.3	0.1	0.4	0.05	0.005
	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	
	7	2	0	0	8	9	0	0	2	34	3	20	2	13	1	101
A9	6.2	0.7	0.2	0.7	12.7	11.5	6.7	0.2	2.2	25.6	6.2	9.1	4.0	14.0	1.7	0.175
	–	–	–	–	–	–	7.4	–	–	–	–	17.4	–	–	–	
	2	0	0	0	1	1	0	0	0	4	0	0	0	1	1	10
A10	0.6	0.1	0.02	0.1	1.2	1.1	0.6	0.02	0.2	2.5	0.6	0.9	0.4	1.4	0.2	0.017
	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	
	11	1	0	0	23	41	0	1	9	59	7	14	10	17	1	194
A11	11.8	1.4	0.4	1.4	24.5	22.1	12.8	0.4	4.3	49.1	11.8	17.5	7.8	26.8	3.3	0.336
	–	–	–	–	–	27.2	19.0	–	–	–	–	–	–	–	–	
	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	1
A28	0.1	0.01	0.002	0.01	0.1	0.1	0.01	0.002	0.02	0.3	0.07	0.1	0.05	0.2	0.02	0.002
	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	
	6	0	1	2	20	3	22	0	1	4	0	1	3	0	0	63
AW19	3.8	0.4	0.1	0.4	7.9	7.2	4.2	0.1	1.4	15.9	3.8	5.6	2.5	8.7	1.1	0.109
	–	–	–	–	23.4	–	92.3	–	–	12.3	–	–	–	10.1	–	
	1	0	0	0	0	0	14	0	0	0	0	0	1	1	0	17
BL	1.0	0.1	0.03	0.1	2.1	1.9	1.1	0.03	0.4	4.2	1.0	1.5	0.7	2.3	0.3	0.029
	–	–	–	–	–	–	163.5	–	–	–	–	–	–	–	–	
	+35	4	1	4	73	66	38	1	13	146	35	52	20	80	10	
Total	0.061	0.007	0.002	0.007	0.126	0.114	0.066	0.002	0.022	0.253	0.061	0.090	0.040	0.138	0.017	n = 578

* – Observed number
– Expected number
– X^2 if >6.63
+ – Number
– Gene frequency

Table 3 HLA A and B haplotypes that showed significant linkage disequilibrium in normal Chinese

Haplotype		Freq. x10^4	Delta	Diff. from expected p
A1	BL	52	.005	< .0001
A2	BW16	433	.024	< .0001
A2	BW46	830	.039	< .0001
A9	BW22	346	.019	< .0001
A11	B15	709	.033	< .0001
AW19	B13	346	.021	< .0001
AW19	B17	381	.031	< .0001
BL	B17	242	.022	< .0001
A2	B15	190	-.018	< .0050
A2	B17	35	-.018	< .0005
A9	B17	0	-.012	< .0100
A11	B17	0	-.022	< .0001
AW19	B40	69	-.021	< .0005
AW19	BW46	0	-.015	< .0025

HLA locus A and B antigens and delta values in those that showed significant positive or negative linkage disequilibrium in normal southern Chinese. There was a good correlation between observed and estimated gene and haplotype frequencies in our normal populations. However, this may not be the case in diseased populations and hence the importance of actually counting the haplotypes. The association of A2 BW46 with the occurrence among Chinese of nasopharyngeal carcinoma was much stronger when calculated by the observed haplotype method than by the estimated joint occurrence method.[4]

In this study no difference in HLA-A B gene nor haplotype frequencies was observed between patients with renal failure and normal subjects. This suggests that perhaps environmental factors are more important in the aetiology of renal failure. However, patients with renal failure comprise a heterogeneous group and this could be a factor in the lack of HLA associations. In this study we have not attempted to subdivide the patients because of the difficulties encountered in establishing an exact aetiology.

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REFERENCES

1. Thomson G, Bodmer W. HLA haplotype associations with disease. Tissue Antigens 1979; 13:91-102.
2. Simons MJ, Wee GB, Goh EH, et al. Immunogenetic aspects of nasopharyngeal carcinoma. IV. Increased risk in Chinese for nasopharyngeal carcinoma associated with a Chinese-related HLA profile (A2, Singapore 2). J Nat Cancer Inst 1976; 57:977-80.
3. Chan SH, Wee GB, Srinivasan N, et al. HLA antigens in three common populations in South East Asia – Chinese, Malay and Filipino. Tissue Antigens 1979; 13: 361-8.
4. Chan SH, Day NE, Kunaratnam N, Chia KB, Simons MJ. HLA and nasopharyngeal carcinoma in Chinese – a further study. Int J Cancer 1983; 32:171-6.