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HLA Class II (DRB1, DQA1 and DQB1) Allele and Haplotype Frequencies among HIV-Infection Discordant Thai Couples

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SUMMARY We investigated the association of HLA-DRB1, -DQA1 and -DQB1 alleles and haplotypes in 33 Thai HIV discordant couples. A significantly lower frequencies of DRB1*14 (3.0% *vs* 11.3%, $p = 0.048$) and DQA1*0103 (0.0% *vs* 5.63%, $p = 0.042$) alleles were found in the seropositive individuals when compared with HIV-negative controls. In contrast, there was no significant difference in HLA-DQB1* allele frequencies. The haplotype analysis revealed that DRB1*1501-DQA1*0102-DQB1*0601 (7.6% *vs* 0.0%, $p = 0.002$), DRB1*0405-DQA1*0302-DQB1*0401 (7.6% *vs* 1.3%, $p = 0.024$) and DRB1*1401-DQA1*0104-DQB1*05031 (6.1% *vs* 0.0%, $p = 0.007$) were found to be significantly higher frequencies when compared between HIV seronegative partners and HIV negative controls, but DRB1*1501-DQA1*0102-DQB1*0502 (0.0% *vs* 8.1%, $p = 0.01$) was significantly lower. The DRB1*1602-DQA1*0101-DQB1*0502 (4.6% *vs* 0.0%, $p = 0.024$) haplotype was found to be significantly higher frequencies in HIV seropositive individuals when compared to HIV negative controls but the DRB1*1502-DQA1*0101-DQB1*0501 (1.5% *vs* 8.1%, $p = 0.049$) haplotype was lower.

Infection with human immunodeficiency virus type 1 (HIV-1) and progression to its end-stage manifestation, acquired immune deficiency syndrome (AIDS), are controlled by numerous factors. Human genetic is believed to be one of these factors because not all people exposed to HIV become infected with the virus.[1] Both, the class I and class II HLA genes are highly polymorphic. The numbers of alleles that can be identified at the product level for the class I genes are 209 for the HLA-A locus, 414 for the B locus and 101 for the C locus.[2] Within class II loci, the numbers of alleles of HLA DRB1, DQA1 and DQB1 are 273, 21 and 48, respectively.[2] Examining these alleles at the molecular level reveals a much higher degree of polymorphism than can be appreciated at the product level.[3] Variability in the susceptibility to infections and diseases caused by infectious agents is a characteristic of all populations. Among susceptible individuals exposed to an infection, not all develop disease. It seems logical to hypothesize that the variability of HLA which confers susceptibility to certain infections and diseases may be applicable to HIV.

Certain HLA antigens have been found more frequent than the

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other among patients with AIDS. In addition, different HLA antigens seem to influence the variability of disease progression in patients from different risk groups with different ethnic backgrounds. An increased prevalence of HLA-B35 and HLA-DR5 was initially observed in HIV associated Kaposi's sarcoma cases.[4-5] The increase in HLA-DR5 occurred primarily among AIDS patients of Italian or Ashkenazi Jewish descent but not among Caucasian patients of Northern European background.[6] HLA-DR5 and HLA-DR2 were found to be associated with susceptibility to Kaposi's sarcoma in patients of different Caucasian subpopulations. It was also noted that AIDS patients with opportunistic infections had a normal frequency of HLA-DR2 and HLA-DR5 antigens and a significantly increased frequency of HLA-DR3 and that the ultimate clinical expression of AIDS in patients with unexplained lymphadenopathy may depend upon genetic factors associated with these particular HLA-DR types.[7] Other studies also observed an increased prevalence of HLA-DR1 in AIDS patients, which was most evident in patients with Kaposi's sarcoma.[8-10] Two extended HLA haplotypes, HLA-A1, -Cw7, -B8, -DR3, -DQ2 and HLA-A11, -Cw4, -B35, -DR1 and -DQ1 were found to be associated with a faster progression to AIDS. The exact role of HLA haplotypes in AIDS progression remains elusive,[11] but there is sufficient evidence for a possible association of HLA or HLA-linked factors and host susceptibility or resistance to AIDS.

Data from several studies have suggested that certain HLA antigens confer susceptibility or resistance to HIV-1 infection. HLA markers associated with increased susceptibility to HIV infection in African Americans were defined as A31, B55, Cw6, Cw7, DR6, DR11, DR12, DQ6 and DQ7 while Cw4 and DR6 were associated with possible resistance to infection.[12] The same study found A28, A66, B48, B65, B70, Cw7, Cw8, DR10, DR12, DQ6, and DQ7 to be associated with susceptibility to HIV-1 infection in the Caucasian Mississippi population.

In this pilot study, we analysed the distribution of HLA antigens and haplotypes in a group of Thai HIV-1 infected individuals and their heterosexual seropositive or seronegative partners. We also tried to determine whether HLA alleles and/or haplotypes were associated with susceptibility or resistance to infectivity with HIV-1.

MATERIALS AND METHODS

Study Subjects

Twenty eight Thai HIV-infected, discordant couples who attended the Antenatal Care Unit, Department of Obstetrics and Gynaecology, Faculty of Medicine Siriraj Hospital, Bangkok, Thailand and five HIV-infected discordant couples from Chaophya Abhaibhuberhj Hospital, Prachin Buri province, Thailand were recruited for this pilot study. Questionnaires for information on HIV risk factors such as sexual behaviour, condom use, drug abuse and history of other sexually transmitted diseases were obtained. EDTA blood (5 ml) was collected from all subjects with informed consents. DNA extraction was performed using phenol-chloroform and stored at -20°C until HLA class II DNA typing was done.

HIV seropositivity was defined by the presence of anti-HIV antibody using three HIV-1/2 screening assays (Axsym HIV ½ MEIA, Abbott, USA; Uniform I HIV ½ EIA, Organon Teknika, Belgium and Serodia, Fujirebio, Japan) according to World Health Organization recommendations.[13] HIV seronegativity was defined by the absence of anti-HIV antibody in at least 2 screening assays and absence of p24 antigen in serum by ELISA (Organon Teknika, Belgium). HIV discordant couples were operationally defined as those whose partners had different serological HIV test results at the time of recruitment. Eighty HIV seronegative blood donors were included in this pilot study as normal controls.

HLA-DRB1, -DQA1 and -DQB1 typing

HLA-DRB1, -DQA1, and -DQB1 alleles were genotyped on genomic DNA using polymerase chain reaction and sequence-specific oligonucleotide probes (PCR-SSOP), with digoxigenin (DIG)-labelled probes and chemiluminescent detection.[14] Primers and probes (DNAgency, Aston, PA, USA) used in this study were validated by the method recommended by the 12th International Histocompatibility Workshop.[15]

Statistical analysis

The allele frequencies of the HLA Class II (-DRB1, -DQA1 and -DQB1) alleles were compared among HIV infected-individuals, their seronegative heterosexual partners and HIV negative controls using chi-square test or Fisher's exact probability tests. Corrected *p*-values were obtained by multiplying *p*-

values by the total number of comparisons for the loci examined. A p-value < 0.05 was considered statistically significant. DRB1, DQA1, DQB1 haplotypes were elucidated using a computer program for estimating allele and haplotype frequencies following the analysis at the Eleventh International Histocompatibility Workshop.[16]

RESULTS

Characteristics of discordant couples

The descriptive statistics for discordant couples are presented in Table 1. The background information of those discordant couples where only the wife was HIV-1 infected (n = 22) was compared to those where only the husband was HIV-1 infected (n = 11). In general, the two groups appeared not to be different. About one-third of the infected wife couples (36.36%) and about half of the infected husband couples (54.55%) lived with the current partners for more than two years. More than half of the infected wife couples (68.18%) and one-third of the infected husband couples (36.36%) had children. About 13.64% of the infected wives were currently pregnant. Interestingly, half of the couples reported having had extra-marital sex with others. About 40.0% of the couples in both groups reported never having had a sexually transmitted disease before. Most couples (77.28% of infected wife couples and 91.91% of infected husband couples) perceived that their partners got HIV infection from sexual contacts. About 54.55% of the couples with infected wives reported never having used a condom before, but after knowing about the infection 50.0% used condoms consistently. About 9.09% of the couples with infected husbands reported constant use of condoms before the infection, but after knowing the infection results, this percentage increased to 27.27%. In contrast, the percentage of couples with infected husbands, who reported never using condoms before and after knowing about the infection were 36.36% and 18.18%, respectively. Moreover, about 50.0% of infected wives never received any HIV-1 treatment before participating in the study, whereas only about 36.36% of infected husbands never received treatment before enrolment in this study.

HLA-DRB1, -DQA1 and -DQB1 genotypes

The HLA-DRB1 allele frequencies among the discordant couples and the HIV negative controls are shown in Table 2. HLA-DRB1*14 was significantly lower in the seropositive discordant individuals compared to the HIV negative controls (3.0% *vs* 11.3%, p = 0.048). The HLA-DQA1*0103 allele was absent in the seropositive discordant group, which was found to be statistically significant when compared to the HIV negative controls (0.0% *vs* 5.6%, p = 0.042). In contrast, there was no significant difference in HLA-DQB1 allele frequencies (Table 3).

HLA-DRB1-DQA1- DQB1 haplotype analysis

Table 4 demonstrates the elucidated frequency of HLA-DRB1-DQA1-DQB1 haplotypes among the discordant couples and HIV negative controls. In the comparison of all HIV seronegative partners and all HIV negative controls, four haplotypes were found to be statistically significant. Three haplotypes were found in significantly higher frequencies: DRB1*1501-DQA1*0102-DQB1*0601 (7.6% *vs* 0.0%, p = 0.002), DRB1*0405-DQA1*0302-DQB1*0401 (7.6% *vs* 1.3%, p = 0.024) and DRB1*1401-DQA1*0104-DQB1*05031 (6.1% *vs* 0.0%, p = 0.007). In contrast, DRB1*1501-DQA1*0102-DQB1*0502 (0.0% *vs* 8.1%, p = 0.01) was significantly found in lower frequency.

Similarly, the DRB1*1602-DQA1*0101-DQB1*0502 (4.6% *vs* 0.0%, p = 0.024) haplotype was significantly found in higher frequency in the HIV-1 seropositive discordant individuals when compared to the HIV negative controls, and DRB1*1502-DQA1*0101-DQB1*0501 (1.5% *vs* 8.1%, p = 0.049) was significantly found in lower frequency.

DISCUSSION

Exposure to human immunodeficiency virus does not necessarily result in infection with the virus. Pathogenesis and progression of this viral infection to its end-stage manifestation, acquired immune deficiency syndrome, are also variable phenomena. Exactly why some exposed persons do not get infected and why some infected persons progress quickly and others slowly is not totally understood.

In some studies, major histocompatibility complex (MCH)-restricted, HIV-specific cellular immune responses have been detected in the absence of any evidence of HIV infection, suggesting that an exposure to the virus has induced an immune response that may

Table 1 Characteristics of discordant couples*

Characteristics	Discordant couples (N = 33)			
	Wife (HIV+) - husband (HIV -) (N = 22)		Wife (HIV-) - husband (HIV+) (N = 11)	
	(N)	%	(N)	%
Living with current partner				
Less than 1 year	4	18.18	1	9.09
1 - 2 years	9	40.91	1	9.09
More than 2 years	8	36.36	6	54.55
History of pregnancy				
No children	2	9.09	2	18.18
Have children	15	68.18	4	36.36
Ever had stillbirth	2	9.09	0	0.00
Currently pregnant	3	13.64	3	27.27
History of having extra marital sex:				
Yes	11	50.00	5	45.45
No	5	22.73	3	27.27
History of other infections				
Never	9	40.91	4	36.36
Gonorrhea	1	4.55	1	9.09
Syphilis	3	13.64	1	9.09
Hepatitis-B	1	4.56	0	0.00
Other	4	18.18	1	9.09
Don't know	0	0.00	1	9.09
Perceived risk factors of HIV infection:				
Sexual	14	63.64	8	72.73
Drug	1	4.55	0	0.00
Other risk	3	13.64	1	9.09
Sexual with other risks	3	13.64	2	18.18
No risk	1	4.55	0	0.00
Condom use before infection:				
Constantly	2	9.09	1	9.09
Sometimes	4	18.18	5	45.45
Never	12	54.55	4	36.36
Condom use after infection:				
Constantly	11	50.00	3	27.27
Sometimes	5	22.73	4	36.36
Never	2	9.09	2	18.18
History of HIV treatment:				
Never received treatment before	11	50.00	4	36.36
Currently received treatment	10	45.45	5	45.45

*Numbers varied because of missing data from the questionnaires.

Table 2 Comparisons of HLA-DRB1 alleles in discordant couples and HIV-1 negative controls

DRB1* alleles	Discordant couples				Control		*p- value	Pc	Odds ratio	Relative risk
	[I] Case (HIV +)		[II] Case (HIV -)		[III] Control (HIV -)					
	N = 33	%	N = 33	%	N = 80	%				
0101	0	0	0	0	1	0.63	NS			
0301	6	9.09	4	6.06	8	5.00	NS			
04	11	16.67	8	12.12	16	10.00	NS			
0401	0	0	0	0	1	0.63	NS			
0403	3	4.55	2	3.03	3	1.88	NS			
0404	1	1.52	0	0	0	0	NS			
0405	5	7.58	6	9.09	6	3.75	NS			
0406	2	3.03	0	0	6	3.75	NS			
0701	6	9.09	7	10.61	13	8.13	NS			
08	2	3.03	2	3.03	5	3.13	NS			
0802	1	1.52	0	0	1	0.63	NS			
0803	1	1.52	1	1.52	4	2.50	NS			
0809	0	0	1	1.52	0	0	NS			
0901	5	7.58	6	9.09	16	10.00	NS			
1001	0	0	1	1.52	1	0.63	NS			
11	3	4.55	4	6.06	11	6.88	NS			
1101	3	4.55	4	6.06	9	5.63	NS			
1106	0	0	0	0	2	1.25	NS			
12	10	15.15	8	12.12	23	14.38	NS			
1201	0	0	1	1.52	1	0.63	NS			
1202	10	15.15	7	10.61	22	13.75	NS			
13	3	4.55	1	1.52	3	1.88	NS			
1301	0	0	0	0	1	0.63	NS			
1302	2	3.03	1	1.52	2	1.25	NS			
1312	1	1.52	0	0	0	0	NS			
14	2	3.03	5	7.58	18	11.25	0.048 [I] *vs* [III]	NS	0.25 (0.04-1.15)	0.27 (0.06-1.13)
1401	1	1.52	3	4.55	9	5.63	NS			
1402	0	0	0	0	1	0.63	NS			
1404	1	1.52	2	3.03	5	3.13	NS			
1410	0	0	0	0	3	1.88	NS			
15	11	16.67	14	21.21	36	22.50	NS			
1501	3	4.55	6	9.09	13	8.13	NS			
1502	8	12.12	8	12.12	22	13.75	NS			
1504	0	0	0	0	1	0.63	NS			
1602	7	10.61	6	9.09	9	5.63	NS			

*presented only when *p*-value < 0.05

Table 3 Comparisons of HLA-DQA1 and -DQB1 alleles in discordant couples and HIV-1 negative controls

Alleles	Discordant Couples				Control		*p-value	Pc	Odds ratio	Relative risk
	[I] Case (HIV +)		[II] Case (HIV -)		[III] Control (HIV -)					
	N = 33	%	N = 33	%	N = 80	%				
DQA1*										
01	22	33.33	29	43.94	75	46.88	NS			
0101	5	7.58	6	9.09	24	15.00	NS			
0102	15	22.73	15	22.73	33	20.63	NS			
0103	0	0.00	1	1.52	9	5.63	0.042 [I] vs [III]	NS	0.00 (0.00-1.40)	
0104	2	3.03	7	10.61	9	5.63	NS			
0201	6	9.09	8	12.12	13	8.13	NS			
03	19	28.79	14	21.21	33	20.63	NS			
0301	6	9.09	3	4.55	12	7.50	NS			
0302	13	19.70	11	16.67	21	13.13	NS			
0401	1	1.52	1	1.52	1	0.63	NS			
0501	9	13.64	8	12.12	19	11.88	NS			
0601	9	13.64	6	9.09	19	11.88	NS			
DQB1*										
02	9	13.64	9	13.64	18	11.25	NS			
03	29	43.94	21	31.82	60	37.50	NS			
0301	12	18.18	10	15.15	30	18.75	NS			
0302	8	12.12	3	4.55	10	6.25	NS			
0303	9	13.64	8	12.12	20	12.50	NS			
04	4	6.06	6	9.09	7	4.38	NS			
0401	2	3.03	5	7.58	5	3.13	NS			
0402	2	3.03	1	1.52	2	1.25	NS			
05	19	28.79	21	31.82	50	31.25	NS			
0501	2	3.03	7	10.61	15	9.38	NS			
0502	16	24.24	9	13.64	28	17.50	NS			
05031	1	1.52	5	7.58	7	4.38	NS			
06	5	7.58	9	13.64	25	15.63	NS			
0601	3	4.55	7	10.61	21	13.13	NS			
0602	0	0	1	1.52	1	0.63	NS			
0603	0	0	0	0	2	1.25	NS			
0604	1	1.52	0	0	0	0	NS			
0605	1	1.52	1	1.52	1	0.63	NS			

*presented only when *p*-value < 0.05

Table 4 Significant differences among HLA haplotypes in discordant couples and HIV negative controls

Haplotype DRB1-DQA1-DQB1	Discordant Couples [I] Case (HIV +)		[II] Case (HIV -)		Control [III] Control (HIV -)		*P value	Pc	Odds ratio	Relative risk
	N = 33	%	N = 33	%	N = 80	%				
0405-0302-0401	2	3.03	5	7.58	2	1.25	0.024 [II] *vs.* [III]	NS	6.48 (1.08-49.64)	6.06 (1.21-30.46)
1401-0104-05031	0	0.00	4	6.06	0	0.00	0.007 [II] *vs.* [III]	NS	Undefined	-
1501-0102-0502	1	1.52	0	0.00	13	8.13	0.010 [II] *vs.* [III]	NS	0.00 (0.00-0.91)	-
1501-0102-0601	1	1.52	5	7.58	0	0.00	0.002 [II] *vs.*[III]	NS	Undefined	-
1502-0101-0501	1	1.52	6	9.09	13	8.13	0.049 [I] *vs.* [III]	NS	0.17 (0.01-1.32)	0.19 (0.02-1.40)
1602-0101-0502	3	4.55	0	0.00	0	0.00	0.024 [I] *vs.* [III]	NS	Undefined	-

* presented only when *p*-value < 0.05

be protective.[17] Nevertheless, what determines which individuals are at lower risk of infection remains unclear.[18] Both, host genetic factors and viral factors have some responsibility for these discrepancies. One host factor that may contribute to disease progression is the individual's HLA antigens, which have been shown to be associated with susceptibility and progression.[19] Factors such as older age,[20] and ongoing high-risk sexual activity[21] have also been shown to contribute to disease progression.

An earlier analysis of data from a European study of heterosexual HIV transmission as well as some other studies suggested that the relationship between the number of accrued sexual contacts of discordant couples and the probability of transmission was not simple.[22-23] A model assuming a constant risk of infection per contact underestimated the risk for smaller numbers of sexual contacts between discordant partners and overestimated it for larger numbers. One interpretation of this observation is that a degree of protection against HIV infection can be acquired following repeated exposure. A further suggestion that exposure is itself protective has come from the more recent observation that among long-term exposed uninfected prostitutes in Nairobi, protection could be lost as a result of a decrease in exposure level.[24] One possible component of the protective response is alloreactivity, an immune response to a mismatched HLA antigen.[25] It has been shown in a macaque monkey model that vaccination with purified Class I molecules can be protective against virus grown in cells expressing the same Class I protein, by virtue of the acquisition of the viral envelop during budding.[26] As HLA DR has been found to be protective in the macaque,[27] it is possible that Class I molecules are particularly significant in HIV protection.

In this pilot study, we investigated whether there is a possible association between HLA Class II (-DR, -DQ) antigens and susceptibility or resistance to HIV infection in a subset of a cohort of HIV infected individuals and their heterosexual HIV seronegative partners (discordant couples of Thai ethnic background). It was found that there was no significant difference in allele frequencies for HLA Class II (DRB1, DQA1, and DQB1) alleles between exposed uninfected individuals and healthy blood donors (Tables 2-4). On the other hand, DRB1*14 and DQA1*0103 alleles were found to be associated with protection from HIV infection. Our results suggest that there may be other HLA haplotypes involved in susceptibility or resistance to HIV infection. It may be worth to note, however, that because of the small sample size for this type of study, we are unable to control for any possible confounding variables, thus the data should be cautiously interpreted.

We also found that the HLA DRB1*1602-DQA1*0101-DQB1*-0502 haplotypes were observed more frequently in the seropositive discordant individuals than in HIV negative controls ($p = 0.024$) which may indicate a positive association with susceptibility to HIV infection. On the other hand the HLA DRB1*-1501-DQA1*0102-DQB1*0601, DRB1*0405-DQA1*0302-DQB1*-0401, and DRB1*1401-DQA1*0104-DQB1*05031 haplotypes were found more frequently in the seronegative discordant individuals than in the normal healthy controls ($p = 0.002$; $p = 0.024$; $p = 0.007$, respectively) which may indicate a positive association with resistance to HIV infection. But when using corrected p-values, the results were not statistically significant. The lack of significant HLA-HIV associations may be due to the small sample size. Haplotype analysis yielded a relationship as strong as or stronger than for any individual component alleles.

There are additional limitations to this study, such as being a pilot study and that some variables such as the duration of the relationship with the current partner as well as condom use before infection (Table 1) may not have been accurately measured, as they were captured posteriori thus making data interpretation rather complicated. Because of the sensitive nature of this infection, we were unfortunately unable to verify data with our volunteers. Future studies that examine HLA association with AIDS need to use larger sample sizes and should include a more thorough analysis of genes closely linked to HLA in order to more precisely locate the disease causing locus.

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REFERENCES

1. Roe DL, Lewis RE, Cruse JM. Association of HLA-DQ and -DR alleles with protection from or infection with HIV-1. Exp Mol Pathol 2000; 68: 21-8.
2. Marsh SG, Bodmer JG, Albert ED, *et al.* Nomenclature for factors of the HLA system, 2000. Tissue Antigens 2001; 57: 236-83.
3. Schreuder GM, Hurley CK, Marsh SG, *et al.* The HLA dictionary 2001: a summary of HLA-A, -B, -C, -DRB1/3/4/5, -DQB1 alleles and their association with serologically defined HLA-A, -B, -C, -DR, and -DQ antigens. Hum Immunol 2001; 62: 826-49.
4. Friedman-Kien AE. Disseminated Kaposi's sarcoma syndrome in young homosexual men. J Am Acad Dermatol 1981; 5: 468-71.
5. Pollack MS, Safai B, Myskowski PL, Gold JW, Pandey J, Dupont B. Frequencies of HLA and Gm immunogenetic markers in Kaposi's sarcoma. Tissue Antigens 1983; 21: 1-8.
6. Prince HE, Schroff RW, Ayoub G, Han S, Gottlieb MS, Fahey JL. HLA studies in acquired immune deficiency syndrome patients with Kaposi's sarcoma. J Clin Immunol 1984; 4: 242-5.
7. Pollack MS, Gold J, Metroka CE, Safai B, Dupont B. HLA-A,B,C and DR antigen frequencies in acquired immunodeficiency syndrome (AIDS) patients with opportunistic infections. Hum Immunol 1984; 11: 99-103.
8. Contu L, Cerimele D, Pintus A, Cottoni F, La Nasa G. HLA and Kaposi's sarcoma in Sardinia. Tissue Antigens 1984; 23: 240-5.
9. Klein MR, Keet IP, D'Amaro J, *et al.* Associations between HLA frequencies and pathogenic features of human immunodeficiency virus type 1 infection in seroconverters from the Amsterdam cohort of homosexual men. J Infect Dis 1994; 169: 1244-9.
10. Mann DL, Murray C, Yarchoan R, Blattner WA, Goedert JJ. HLA antigen frequencies in HIV-1 seropositive disease-free individuals and patients with AIDS. J Acquir Immune Defic Syndr 1988; 1: 13-7.
11. Roger M. Influence of host genes on HIV-1 disease progression. FASEB J 1998; 12: 625-32.
12. Cruse JM, Brackin MN, Lewis RE, Meeks W, Nolan R, Brackin B. HLA disease association and protection in HIV infection among African Americans and Caucasians. Pathobiology 1991; 59: 324-8.
13. Global Programme on AIDS: Recommendations for the selection and use of HIV antibody tests. Wkly Epidemiol Rec 1992; 20: 145-9.
14. Bignon JD, Fernandez-Vina MA. Protocols of the 12th International Histocompatibility Workshop for typing of HLA class II alleles by DNA amplification by the polymerase chain reaction (PCR) and hybridization with sequence specific oligonucleotide probes (SSOP). In: Charron D, ed. Genetic Diversity of HLA. Functional and Medical Implication I. Paris, France, EDK, 1997; pp. 584-95.
15. Fernandez-Vina MA, Bignon JD. Primers and oligonucleotide probes (SSOP) used for DNA typing of HLA class II alleles. In: Charron D, ed. Genetic Diversity of HLA. Functional and Medical Implication I. Paris, France, EDK, 1997; pp. 596-632.

16. Imanishi T, Akaza T, Kimura A, *et al.* Estimation of allele and haplotype frequencies for HLA and complement loci. In: Tsuji K, Aizawa M, Sasazuki T (eds.). HLA 1991; Vol. 1, Oxford University Press, Oxford, 1992; pp. 76-9.
17. Rowland-Jones SL, Dong T, Dorrell L, *et al.* Broadly cross-reactive HIV-specific cytotoxic T-lymphocytes in highly-exposed persistently seronegative donors. Immunol Lett 1999; 66: 9-14.
18. Plummer FA, Ball TB, Kimani J, Fowke KR. Resistance to HIV-1 infection among highly exposed sex workers in Nairobi: what mediates protection and why does it develop? Immunol Lett 1999; 66: 27-34.
19. Winchester R, Charron D, Louie L, *et al.* The role of HLA in influencing the time of development of a particular outcome of HIV-1 infection. In: Charron D, ed. Genetic Diversity of HLA: Functional and Medical Implication. Vol. II. Paris, France, EDK, 1997; pp. 423-8.
20. Chaisson RE, Keruly JC, Moore RD. Race, sex, drug use, and progression of human immunodeficiency virus disease. N Engl J Med 1995; 333: 751-6.
21. Phair J, Jacobson L, Detels R, *et al.* Acquired immune deficiency syndrome occurring within 5 years of infection with human immunodeficiency virus type-1: the Multicenter AIDS Cohort Study. J Acquir Immune Defic Syndr 1992; 5: 490-6.
22. Downs AM, de Vincenzi I. Probability of heterosexual transmission of HIV: relationship to the number of unprotected sexual contacts. European Study Group in Heterosexual Transmission of HIV. J Acquir Immune Defic Syndr Hum Retrovirol 1996; 11: 388-95.
23. Mastro TD, Kitayaporn D. HIV type 1 transmission probabilities: estimates from epidemiological studies. AIDS Res Hum Retroviruses 1998; 14 (Suppl 3): S223-7.
24. Kaul R, Rowland-Jones S L, Kimani J, *et al.* Late seroconversion in HIV-resistant Nairobi prostitutes despite pre-existing HIV-specific CD8+ responses. J Clin Invest 2001; 107: 341-9.
25. Lehner T, Shearer GM, Hackett CJ, Schultz A, Sharma OK. Alloimmunization as a strategy for vaccine design against HIV/AIDS. AIDS Res Hum Retroviruses 2000; 16: 309.
26. Chan WL, Rodgers A, Grief C, *et al.* Immunization with class I human histocompatibility leukocyte antigen can protect macaques against challenge infection with SIVmac-32H. AIDS 1995; 9: 223-8.
27. Arthur LO, Bess JW Jr, Urban RG, *et al.* Macaques immunized with HLA-DR are protected from challenge with simian immunodeficiency virus. J Virol 1995; 69: 3117-24.