Research letters

Transmission of HIV-1 and *HLA-B* allele-sharing within serodiscordant heterosexual Zambian couples

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Factors that might increase risk of HIV-1 transmission include age, sex, and amount of HIV-1 RNA in plasma, but findings for *HLA* allele-sharing are not in agreement. We tested the hypothesis that allele sharing at *HLA* loci is associated with increased risk of transmission of HIV-1 infection in cohabiting heterosexual Zambian couples. We studied 125 initially serodiscordant partners with sequence-confirmed interpartner HIV-1 transmission and 104 couples who were persistently serodiscordant, and we analysed relations with molecularly typed *HLA-A*, *B*, and *C* alleles by survival techniques. After adjustment for other genetic and non-genetic risk factors seen with heterosexual transmission of HIV-1 in this cohort, sharing of *HLA-B* alleles was independently associated with accelerated intracouple transmission (relative hazard 2·23, 95% CI 1·52–3·26, p<0·0001). Selective pressure by *HLA-B* alleles on transmitted viruses accords with current understanding of the effect of B locus polymorphism in HIV-1 and perhaps other infections.

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HIV-1 is spreading in southern Africa, mainly by heterosexual transmission. In Zambia, about 20% of cohabiting couples are HIV-1-serodiscordant—ie, one partner is seropositive and the other is seronegative.1 Despite aggressive behavioural interventions, cohabiting serodiscordant Zambian couples prospectively counselled and assessed for more than 5 years seroconvert (mainly with HIV-1 subtype C) at a frequency of about eight per 100 person-years.[1,2]

Factors reported to modify infectiousness or susceptibility of individuals include amount of HIV-1 RNA in plasma, age, sex, history of sexually transmitted disease, alcohol use, and possibly HIV-1 subtype.[3] Findings of studies of host immunogenetic factors in mother-to-child and horizontal transmission have suggested that both sharing of *HLA* alleles and possession of specific alleles affect HIV-1 transmission.[4–7] However, findings in prospective investigations of *HLA* relations have been disparate most probably because of ethnic admixture, modest sample size, serological typing, absence of virologically documented transmission between study partners, or statistical considerations. We aimed to test the hypothesis that allelic sharing at *HLA* class I loci is associated with increased risk of HIV-1 transmission.

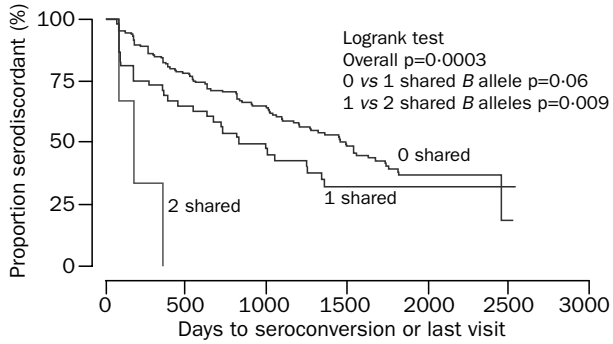
From 1994 to 2000, we followed more than 1300 cohabiting HIV-1 serodiscordant couples four times a year in a voluntary counselling and testing programme (the Zambia UAB HIV Research Project).[1,2,8] Eligibility criteria for the present study included discordant serostatus, 6 months or more of cohabitation in Lusaka, and age 48 years or younger for women and 65 years or younger for men. We provided free condoms and outpatient medical care and gave newly seroconverted partners further appropriate counselling. Study procedures and forms received institutional ethics approval from the University of Alabama at Birmingham, the University Teaching Hospital, Lusaka, and the National Institutes of Health, Bethesda, for consent, initially for HIV testing and counselling and subsequently for analysis of blood samples.

For all couples, we measured amount of HIV-1 RNA in the positive partner at enrolment.[1] In both partners we typed *HLA* class I and class II loci by standard PCR-based methods (including sequencing when necessary for four-digit allele assignment). We compared couples transmitting phylogenetically linked viruses (95% subtype C)[2] with non-transmitting couples who met the following criteria: at fairly high risk of HIV-1 transmission (because of infrequent condom use and advanced age), ample follow-up (>85% with ⩾2 years of observation), and available DNA. We compared these two groups of couples for sharing of *HLA* class I alleles,

	Non-transmitting couples	Transmitting couples	p*
Characteristic			
Number of couples	104 (96)	125 (120)	..
Age of partners (years)			
Index	30·1 (30·1)	30·6 (30·7)	0·6
Non-index	31·7 (31·6)	28·1 (28·2)	0·002
Male/female (index partner)	42/62	80/45	0·0004
Number of years cohabiting	6·3 (6.4)	6·0 (6·0)	0·5
Number of sex acts per 3 months (as reported by female partner)	23·4 (24·3)	28·6 (29·5)	0·05
Follow-up time (median [IQR], days)	1549 (886–2021)	550 (186–1067)	<0·0001
Genital inflammation (%)			
Index	11·5 (12·5)	16·0 (16·7)	0·3
Non-index	2·9 (3·1)	11·2 (10·0)	0·02
Genital ulceration (%)			
Index	4·8 (5·2)	16·0 (15·8)	0·007
Non-index	1·0 (1·0)	16·8 (15·8)	<0·0001
Number of pregnancies	3·4 (3·5)	3·3 (3·3)	0·6
Number of births	2·9 (3·0)	2·8 (2·8)	0·5
HIV-1 RNA level in index partner (log_{10})	4·5 (4·5)	5·0 (5·0)	<0·0001

Data are means unless otherwise indicated. Data in parentheses are corresponding values for the subset of couples with data sufficient for inclusion in multivariable models. *Univariate comparison.

Characteristics of couples



Number of couples at risk

Days	0	300	600	900	1200	1500	1800	2100	2400
2 shared	3	1	0	0	0	0	0	0	0
1 shared	55	40	31	24	17	10	7	4	1
0 shared	171	147	122	96	77	55	36	19	4

Kaplan-Meier plot for *HLA-B* sharing in heterosexual transmission of HIV-1

display of certain individual class I alleles, and homozygosity at class I loci.

For survival analysis of associations with class I allele sharing, by Kaplan-Meier plots and Cox regression models (SAS version 9.00; SAS Institute, Cary, NC, USA), we used enrolment date as the zero timepoint and HIV-1 seroconversion as the event in the initially seronegative partners. Multivariable proportional hazards models of the *HLA-B* sharing relation included several other covariates predicting transmission. The assumption of proportional hazards was investigated for all covariates and we tested effects of any non-proportionality in additional models.

Both partners in 125 of 129 transmitting couples had sufficient DNA for *HLA* typing; 120 had non-genetic data for multivariable analysis. A further 104 couples were non-transmitting, of whom 96 had sufficient data for multivariable analysis. At baseline, transmitting and non-transmitting pairs were similar with respect to age of index partners, length of cohabitation, and number of pregnancies and births. The two groups differed by sex of the index partner (attributable to enrolment and retention patterns rather than a difference in transmission rates)[1] and in certain behavioural and clinical characteristics predictably related to transmission (table). Frequency of intercourse was 20% higher and median follow-up time (representing time to event for transmitting pairs and total time enrolled for non-transmitting pairs) nearly three-fold longer (550 *vs* 1549 days) in non-transmitting couples than in transmitting pairs.

Because couples in the present analysis were selected from the larger Zambia UAB HIV Research Project cohort for being at high risk, the annual HIV-1 transmission rate for the couples analysed here (about 14%) and for every separate sharing category (figure) exceeded the annual rate for the entire cohort of the Zambia UAB HIV Research Project (about 8%).[1] Nevertheless, the HIV-1 transmission rate was significantly higher in partners who shared *HLA-B* alleles in a univariate comparison with those who did not share alleles, whether sharing was assessed at two-digit or four-digit allelic resolution (relative hazard 1·69, 95% CI 1·17–2·44, p=0·005). The association with accelerated transmission in *B*-allele sharing partners seemed somewhat stronger after adjustment for non-genetic variables (2·20, 1·50–3·23, p<0·0001).

B-sharing was independent of the following potential transmission factors: age of seronegative partner (relative hazard 0·97, 95% CI 0·95–1·00, p=0·04); sex of index partner (1·48, 0·92–2·39, p=0·11); frequency of sexual intercourse (1·01, 1·00–1·01, p=0·008); genital inflammation or ulcer in index partner during 3-month interval before transmission or preceding last visit (1·88, 1·21–2·93, p=0·005); genital inflammation or ulcer in susceptible partner during 3-month interval before transmission or preceding last visit (2·46, 1·58–3·84, p<0·0001); HIV-1 RNA amount in index partner (1·72, 1·32–2·23, p<0·0001); and frequency of alcohol consumption by male partner (1·07, 0·95–1·21, p=0·24). Exclusion of a brief interval of non-proportionality for the *B*-sharing hazard and two covariates (male alcohol use and frequency of sexual intercourse) showing non-proportional hazards did not alter the results appreciably (relative hazard for *B*-sharing 2·25, 95% CI 1·56–3·26, p<0·0001 for exclusion of both non-proportionality and the behavioural variables).

In transmitting couples, median time to transmission was 185 days (IQR 91–365) in the three who shared both *B* alleles, 395 days (99–823) in 33 couples who shared one, and 638 days (274–1210) in 89 who shared none (logrank p=0·0003 for the three categories of *B* allele sharing). Certain individual HLA class I alleles were also associated with increased or decreased rates of transmission (data not shown). However, in a separate model adjusted for these class I alleles and the non-genetic variables, the association of *HLA-B* sharing with a higher transmission rate remained strong and independent (relative hazard 2·23, 95% CI 1·52–3·26, p<0·0001). By contrast, association with sharing was absent at *HLA-A*, *HLA-DRB1* and *DQB1* loci but weak at the *C* locus, presumably as a result of *B* locus linkage disequilibrium in couples analysed here. Class I homozygosity in the susceptible partner was not associated with transmission rate. The increased transmission rate in *B* allele-sharing couples could not be easily accounted for by an unusual distribution of *B* alleles in the entire set of partners analysed (frequencies were comparable with those in all typed Zambians)[9] or by a higher frequency of more common alleles or supertypes in susceptible people.[10]

Enhanced HIV-1 transmission due to *HLA* class I allele sharing is biologically plausible. Virus already adapted to or escaped from the specific class I-mediated cytotoxic T lymphocyte response of the index partner would presumably be partly primed for further advantage in a susceptible host who is similar or identical for certain class I alleles. The specificity of the association with the *B* locus might reflect its known distinction as the class I locus with the most extensive polymorphism, the most rapid generation and divergence of alleles, the highest degree of conservation of T-cell receptor and CD8 interaction sites, and the highest average heterozygosity of peptide-interacting sites. In solid-organ transplantation, survival improves more with matching for *B* than for *A* or *C* alleles; and in HIV-1 infection, *B* alleles have been most consistently associated with altered rates of disease progression. Alternatively, mechanisms other than CTL can account for enhanced *B* locus effects. Pathways in the innate immune system (eg, natural-killer cell activity) might be involved, and alloimmunity inhibits in-vitro viral susceptibility of activated CD4+ T cells from HIV-1-exposed but uninfected patients.[11]

Our observations here suggest that transmission of HIV-1 (and perhaps certain other sexually transmitted agents) might be most efficient in family pairings or in genetically homogeneous populations in which *HLA-B*-allele sharing is common. Prospects for prediction or prevention of HIV-1 transmission through routine genetic typing and counselling in exposed uninfected individuals are not yet compelling enough to justify the effort or expense. More immediately, however, documentation of *B*-allele sharing could enhance interpretation of findings of epidemiological

studies of transmission and efficiency in the conduct of prevention trials.

Contributors

M T Dorak participated in data analysis, interpretation of results, and preparation of the manuscript. J Tang and E S Lobashevsky were responsible for HLA typing, laboratory data acquisition and management, and contributed to analysis and preparation of the manuscript. A Penman-Aguilar, A O Westfall, and M M Schaen were responsible for statistical analysis and management of the full dataset and reviewing of the final version of the manuscript. I Zulu and N G Kancheya assisted with protocol development and assembling of the participants, supervised onsite collection of epidemiological data and biological specimens, and reviewed the latest version of the manuscript. S A Allen and R A Kaslow designed the original study and framework for immunogenetics analyses, supervised collection of relevant clinical and epidemiological data and biological specimens, data analysis and interpretation, and reviewed successive drafts of the manuscript.

Conflict of interest statement

None declared.

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Human monoclonal antibody as prophylaxis for SARS coronavirus infection in ferrets

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SARS coronavirus continues to cause sporadic cases of severe acute respiratory syndrome (SARS) in China. No active or passive immunoprophylaxis for disease induced by SARS coronavirus is available. We investigated prophylaxis of SARS coronavirus infection with a neutralising human monoclonal antibody in ferrets, which can be readily infected with the virus. Prophylactic administration of the monoclonal antibody at 10 mg/kg reduced replication of SARS coronavirus in the lungs of infected ferrets by 3·3 logs (95% CI 2·6–4·0 logs; p<0·001), completely prevented the development of SARS coronavirus-induced macroscopic lung pathology (p=0·013), and abolished shedding of virus in pharyngeal secretions. The data generated in this animal model show that administration of a human monoclonal antibody might offer a feasible and effective prophylaxis for the control of human SARS coronavirus infection.

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Severe acute respiratory syndrome (SARS) has emerged as a frequently fatal respiratory-tract infection caused by the newly identified SARS coronavirus. After the worldwide SARS epidemic in 2002–2003, sporadic cases continue to arise in southern China, possibly because of human contact

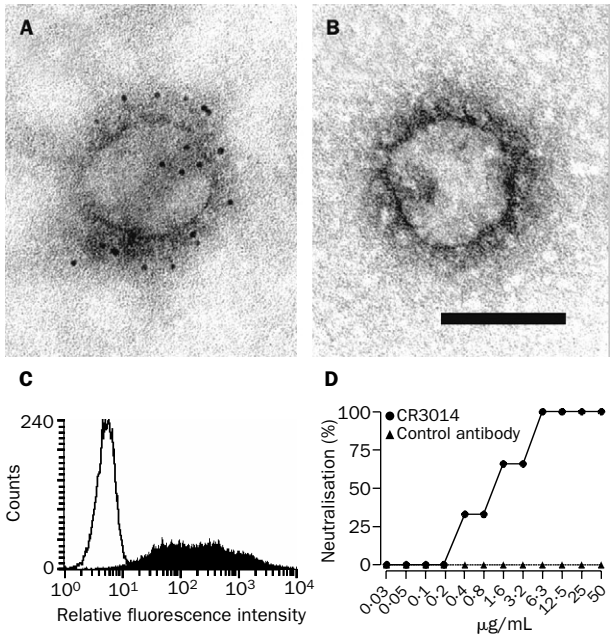

Figure 1: **Binding of CR3014 monoclonal antibody to viral peplomers of SARS coronavirus (A, B) and to HEK293T cells expressing glycoprotein spike of SARS coronavirus (C), and in vitro neutralisation of SARS coronavirus strain HK-39849 (D)**

Incubation with CR3014 led to a dense gold-label of the outer peplomer region of SARS coronavirus strain Frankfurt 1 (A), whereas a control human IgG1 monoclonal antibody did not induce any label (B). Bar indicates 100 nm. (C) Filled histogram=glycoprotein spike of SARS coronavirus, strain Frankfurt 1. Open histogram=control plasmid.