

AIDS restriction HLA allotypes target distinct intervals of HIV-1 pathogenesis

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An effective acquired immune response to infectious agents mediated by HLA-restricted T-cell recognition can target different stages of disease pathogenesis. We show here that three distinct HLA alleles known to alter the overall rate of AIDS progression act during distinct intervals after HIV-1 infection. The discrete timing of HLA allele influence suggests alternative functional mechanisms in immune defense against this dynamic and chronic immunosuppressive disease.

*HLA-B*27* and *HLA-B*57* show robust protection against AIDS progression relative to all other *HLA* class I alleles, whereas certain *HLA-B*35* subtypes (termed *B*35-Px*) show strong influence on susceptibility to developing AIDS rapidly¹. HIV-1-specific CD8⁺ T-cell responses restricted by *B*27* and *B*57* provide a probable mechanism for the epidemiological protection shown in carriers of these alleles. *B*27* mediates an immunodominant response against a viral peptide derived from a conserved region of HIV p24 Gag^{2,3}, termed KK10, which can lead to the selection of cytotoxic T lymphocyte (CTL) escape variants (R264K/G/T), generally years after initial infection^{2,4,5}. The basis for *B*57* protection also seems to involve, in part, a highly conserved immunodominant epitope in Gag, termed TW10, a response to which seems to confer protection early during acute infection⁶. Mutation of TW10 may considerably attenuate viral fitness based on the observed reversion back to the wild-type sequence within months after transmission of the virus to *B*57*⁻ individuals⁷. Reversion of the KK10 mutation upon transmission of the virus from *B*27*⁺ to *B*27*⁻ individuals does not occur nearly as rapidly^{8,9}, if at all. Susceptibility mediated by *B*35-Px* remains the most enigmatic, but differential correlations of CTL activity with HIV-1 viral load suggest that the peptide-binding signatures of *B*35-Px* may affect the quality of HIV-specific CTL activity¹⁰.

Survival analyses of *HLA* class I data from 1,089 seroconverter individuals (**Supplementary Methods** online) show that *HLA-B*35-Px*, *HLA-B*57* and *HLA-B*27* associate with rate of progression from seroconversion to several of four outcomes (**Fig. 1a** and **Supplementary Table 1** online): (i) CD4 < 200 cells/μl blood (CD4 < 200), (ii) CD4 < 200 and/or AIDS-defining illness (1993 definition¹¹), (iii) AIDS-defining illness (1987 definition¹²) and (iv) death. *B*35-Px* and *B*57* associate with progression to all four outcomes, whereas *B*27* associated with the three later outcomes, but not time to CD4 < 200. These data suggest that *HLA-B* alleles may show temporal differences in their effects on disease progression. Using an expanded dataset of 2,627 individuals, we tested the alleles for effects on progression at distinct intervals after infection: seroconversion to CD4 < 200 (for which only seroconverters can be tested); CD4 < 200 to an AIDS-defining illness (seroconverters and seroprevalent individuals); and AIDS-defining illness to death (seroconverters and seroprevalent).

*B*57*-mediated protection occurs early after infection (seroconversion to CD4 < 200; relative hazard (RH) = 0.41, $P < 0.0001$; **Table 1** and **Fig. 1a**), associating with and probably responsible for delayed CD4 T-cell decline. Once the CD4 cell counts drop to 200 cells/μl, the protective effect of *B*57* begins to subside (CD4 < 200 to an AIDS-defining illness; RH = 1.08, $P = 0.8$, seroconverters only; RH = 0.69, $P = 0.01$, seroconverters plus seroprevalent individuals). *B*57* protection during the acute phase of infection was observed previously⁶, correlating with the timing of the epidemiological effects described here. The rate of viral escape from CTL-mediated immune pressure is determined by a delicate balance between the strength of the immune pressure and the costs in viral fitness resulting from the escape¹³. Relative to other HLA allotypes, *B*57* seems to exert strong selection pressure on the virus, as escape from the *B*57*-mediated CTL response can occur within months after infection¹⁴ (T. Allen & M.A., unpublished data). Positive selection of the TW10 mutants within a host was also documented based on ratios of nonsynonymous to synonymous nucleotide substitutions⁷. But mutation of the TW10 epitope must occur at a sizeable fitness cost to the virus, as reversion of a common *B*57* escape mutation, Gag T242N, back to wild-type occurs after transmission to *B*57*⁻ recipients⁷. Thus, the early *B*57* effect observed in the genetic analyses may be the result of rapid viral escape from a strong *B*57*-restricted TW10 CTL response leading to a predominant TW10 mutant virus that is less fit relative to TW10 wild-type virus (**Fig. 1b**), resulting in the maintenance of high CD4 counts for many years.

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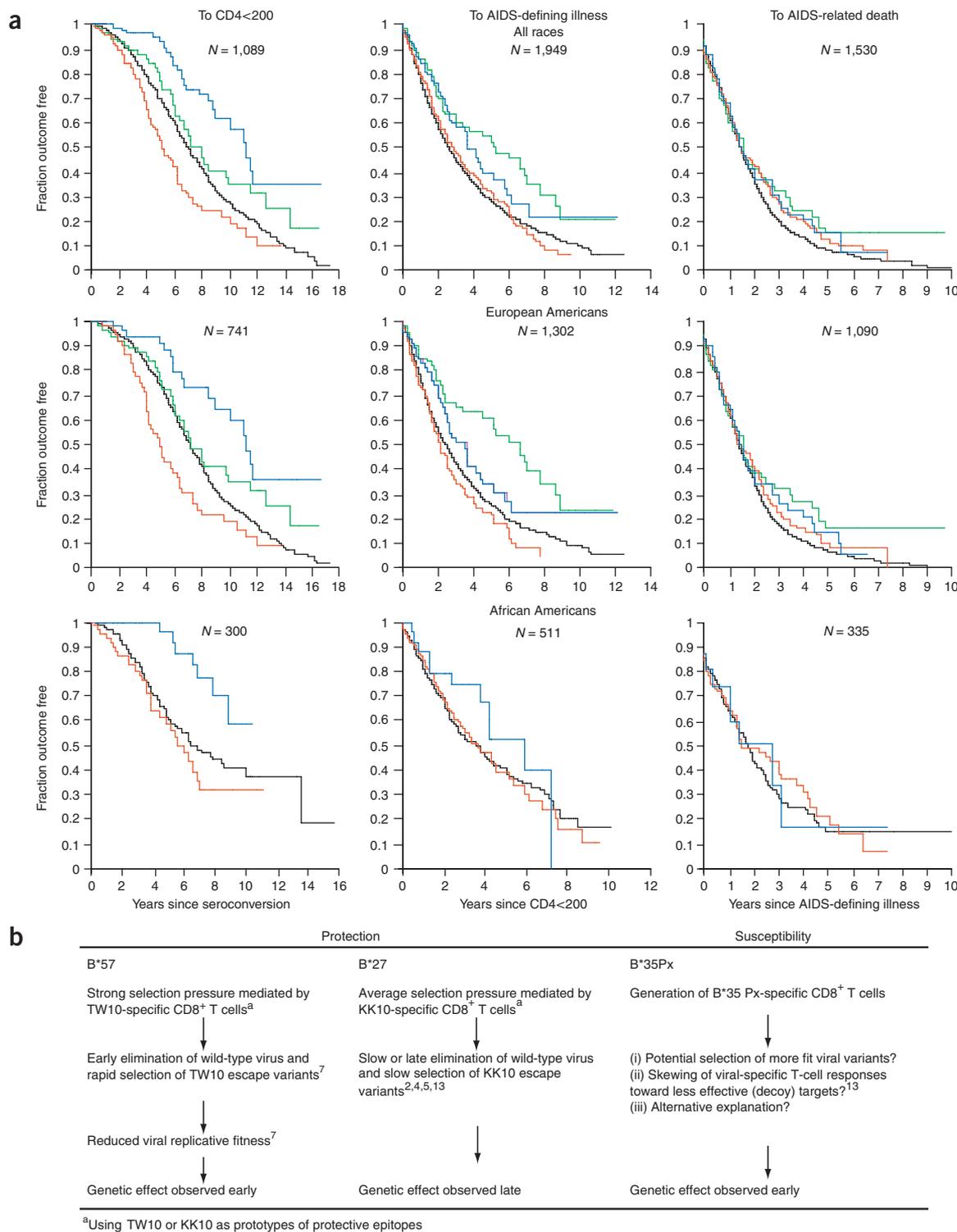


Figure 1 Effect of *HLA* alleles on different stages of AIDS progression. **(a)** Effect of *HLA* alleles on the rate of progression to AIDS. Green, *B*27*; red, *B*35-Px*; blue, *B*57*; black, others. **(b)** Possible models explaining the distinct temporal genetic effects of selected *HLA-B* alleles on AIDS progression.

In sharp contrast, *B*27* shows no significant protection against progression to CD4 < 200 relative to most other *HLA* alleles, but rather delays progression to an AIDS-defining illness primarily after CD4 counts have dropped (RH = 0.54, *P* = 0.01, seroconverters only; RH = 0.53, *P* < 0.0001, seroconverters plus seroprevalent individuals; **Table 1**). We propose that the selection pressure exerted on the virus

to mutate the *B*27*-restricted KK10 epitope is not as great as that as that exerted by *B*57* and in fact is not greater than most other *HLA* allotypes in the early stages of infection before CD4 decline (CD4 < 200) has occurred. A fitness cost upon KK10 mutation combined with the moderate selection pressure on this epitope may explain the observed late escape. We hypothesize that *B*27* may show

Table 1 Effect of *HLA-B*27*, *B*57* and *B*35-Px* on progression to AIDS at distinct intervals after infection

	Seroconversion to CD4 < 200				CD4 < 200 to AIDS-defining illness				AIDS-defining illness to death			
	N = 1,089 ^a				N = 1,949 ^a				N = 1,530 ^a			
	RH	P	n	Percent	RH	P	n	Percent	RH	P	n	Percent
<i>B*27</i> ^b	0.77	0.12	83	7.6	0.53	<0.0001	103	5.3	0.75	0.05	68	4.4
<i>B*57</i> ^b	0.41	<0.0001	99	9.1	0.69	0.01	107	5.5	0.85	0.26	67	4.4
<i>B*35-Px</i> ^b	1.43	0.005	140	12.9	1.08	0.40	282	14.5	0.92	0.35	219	14.3

^aThe total number of individuals in the study equals 2,627, of whom 1,727 are European American, 736 are African American and 164 are of other racial backgrounds. ^bThe phenotypic frequencies of the *B*27*, *B*57* and *B*35-Px* HLA alleles for the entire cohort are 5.8%, 7.0% and 13.6%, respectively.

protection against HIV during the early stages of infection (but not to any greater extent than most other allotypes), however, a considerable protective effect is only seen late during infection because the virus escapes late and the mutation in the KK10 epitope affects fitness to a greater extent than mutations in epitopes for most other HLA allotypes (except for *B*57*).

Late escape from a *B*27*-mediated response has been documented previously, where mutation in the KK10 epitope was observed before a sharp rise in viral load⁵. Notably, early viral escape from a *B*27*-mediated response occurred in an individual who had been vaccinated with p24 Gag and who was known to have a strong *B*27*-mediated response to the KK10 epitope before HIV-1 infection¹⁵. Perhaps strong, immediate *B*27*-mediated elimination of wild-type virus resulted in an atypically rapid increase in the frequency of the KK10 mutant virus, leading to accelerated disease progression relative to most nonvaccinated *B*27*⁺ HIV-1-infected individuals. Thus, late viral escape from a *B*27*-mediated KK10 response may allow enduring restriction of HIV long after escape from most other HLA allotypes has occurred (Fig. 1b), effectively delaying time to AIDS.

*B*35-Px* shows an early susceptibility effect (seroconversion to CD4 < 200; RH = 1.43, *P* = 0.005; Table 1), but *B*35* influence is undetectable once CD4 < 200 is reached, suggesting that *B*35-Px*-mediated rapid progression to AIDS is a function of early decline in CD4 counts. Whether a particular *B*35-Px*-restricted peptide(s) is involved in this activity is unknown. Viral escape from a *B*35-Px*-mediated response may occur at a particularly rapid rate compared to escape from other HLA allotypes, leading to rapid CD4 decline. If *B*35-Px* behaves as a null allele, then heterozygotes for *B*35-Px* would be the functional equivalents of homozygotes at the *HLA-B* locus. But the rate of disease progression amongst heterozygotes for *B*35-Px* is much faster than that observed for true *HLA-B* homozygotes¹, suggesting that *B*35-Px* alleles mediate an actively negative effect in AIDS pathogenesis. More plausible explanations for the early *B*35-Px* effect are suggested in Fig. 1b.

The genetic data presented here, in combination with multiple functional studies reported previously, support the concept that the dynamics by which distinct HLA allotypes render protection against a given infectious pathogen are not necessarily uniform, but rather may differ from one another depending on the nature of the antigenic epitopes recognized by those allotypes.

Note: Supplementary information is available on the Nature Medicine website.

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COMPETING INTERESTS STATEMENT

The authors declare that they have no competing financial interests.

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