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Introduction

Some rare HIV-1 infected individuals, referred to as "elite controllers" (ECs), have persistently undetectable plasma viral load in the absence of therapy. Data from human ECs remains inconclusive, but several lines of evidence suggest that certain individuals make successful natural immune responses to HIV. Vigorous CD4+ T-cell and CD8+ T-cell cellular immune responses against the virus have been observed in controllers, suggesting that these responses are actively containing infection. *HLA* polymorphism has been shown to play a role in protection against HIV infection and several studies examining *HLA* associations with disease progression have suggested that certain *HLA* class I alleles, such as *HLA-B*57*, are associated with low set-point viremia and delayed onset of AIDS.

The aims of this study were to determine the *HLA* allele distribution in ECs who were recently infected in Sao Paulo, Brazil. Brazilian patients express a mixture of *HLA* alleles reflecting the diverse groups of individuals that populated this country. Additionally, there are several different strains of viruses circulating in South America.

Methods

A cohort of recently HIV-1 infected subjects identified by serologic testing algorithm for recent human immunodeficiency virus seroconversion (STARHS) was prospectively enrolled since 2002. All participants signed the IRB approved informed consent and the entry time was set when the first serology was performed and clinical evaluation were performed every three months thereafter. Venous blood was collected, PBMC was isolated by Ficoll-Hypaque density centrifugation and the DNA was obtained from whole blood using the QIAamp DNA blood kit following the manufacturer's guidelines. The *HLA* class I molecular typing was carried out using sequence-specific probe PCR (PCR-SSP) (Figure 1). We also analyzed T cell counts by flow cytometry (Becton Dickinson) and HIV-1 viral load by PCR using an Amplicor Monitor (Roche Diagnostics Systems, Branchburg, NJ), with a lower detection limit of 400 copies/mL.

Nonparametric analyses were performed, significance threshold was defined at $p < 0.05$.

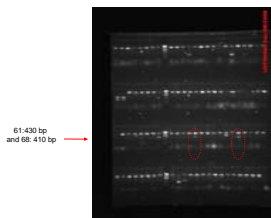


Figure 1 : Electrophoresis of a *HLA-B*5701* positive sample. 2 positive wells (61 and 68)

Results

Of the 197 participants, 92.1% were men and their median age was 31.5 years (25-75% IQR, 25.2-37.0). As depicted in Table 1, median CD4+ T cell count was 535 cells/mm³ (IQR, 420-699), reflecting the relatively preserved cellular immunity, compatible with recently acquired HIV-1 infection, together with higher CD8+ T cell counts. The viral load was 18,600 HIV-RNA copies/ml (IQR, 4,115-63,350), also compatible with recent infection.

Among the enrolled volunteers, we were able to identify seven (3.5%) who have been consistently presenting with undetectable viral loads despite the fact that they have never been exposed to any antiretroviral therapy. We have defined this small group as "elite controllers" (Table 2). These individuals, have also maintained high levels of CD4+ T lymphocyte counts (viral load <400 copies/mL and CD4+ T cell counts >400 cells per μ L) (Figure 2) based on viral load assessments throughout their follow-up (median 429.7 days).

The most common *HLA* alleles were *HLA-A*0201* (46%), *-B*1510* (18%) and *-B*3520* (18%), and *-Cw*0701* (34%). Five (71.4%) ECs were *HLA-B*5701* positive, (table 3), whereas only three of the normal progressors were positive for this allele (N=43). Overall, the *HLA-B*5701* allele was strongly correlated with EC status ($p < 0.001$).

Table 1 : Selected cohort characteristics at enrollment.

Variable	Cohort participant N (%)
Age, in years: median (IQR)	31.5 (25.2, 37.0)
Sex: male	180 (92.3%)
Racial background	
White	118 (62.1%)
Mulatto	36 (18.9%)
Black	15 (7.9%)
Other (indian, asian, not defined)	21 (11.1%)
CD4+ T cells (per μ l): median (IQR)	527 (403, 698)
CD8+ T cells (per μ l): median (IQR)	903 (609, 1,190)
Viral load (copies/ml): median (IQR)	19,800 (4,115, 69,050)

Table 2 : Characteristics of "elite controllers"

Number	Age (years)	Gender	Days in follow-up	CD4+ T cells range (per μ l of blood)	Chronic phase Highest viral load (copies/ml)
1034	23.9	Male	861	516 – 1135	<400
1060	39.4	Male	567	728 – 913	<400
1068	46.6	Male	420	727 – 856	<400
1073	36.6	Male	377	433 – 762	<400
1098	30.3	Male	664	654 – 879	<400
1103	25.7	Male	370	711 – 813	<400
2017	40.1	Male	562	518 – 662	<400

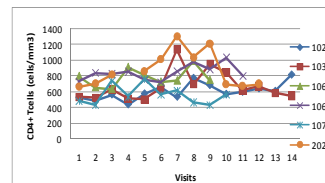


Figure 2: CD4+ T lymphocyte counts in the individuals controlling HIV viral load.

Table 3: *HLA* class I molecular typing of ECs

Code	<i>HLA</i> alleles		
	A	B	C
1034	0201/0301	1510/ 5701	0202/0801
1060	3001/3201	1402/ 5701	0802/1801
1068	0301/2901	0702/5801	0702/0729
1073	0101/3401	3501/ 5701	0413/0707
1098	0301/2601	2708/ 5701	0102/1801
1103	0101/0201	3505/ 5701	0401/0602
2017	0201/1101	3901/5303	0401/0702

Conclusions

1 A considerable frequency (71%) of patients identified as elite controllers was observed in a cohort of patients who were recently infected with HIV-1.

2 We have identified 5 *HLA-B*5701* positive ECs. This allele was highly enriched in our ECs, suggesting that *HLA-B*5701* plays an important role in controlling viral replication, even in heterogeneous populations infected with diverse strains of HIV-1.

Notes

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