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# Effects of Plasma HIV RNA, CD4+ T Lymphocytes, and the Chemokine Receptors CCR5 and CCR2b on HIV Disease Progression in Hemophiliacs

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**Summary:** We have investigated the effects of plasma HIV RNA, CD4+ T lymphocytes and chemokine receptors CCR5 and CCR2b on HIV disease progression in hemophiliacs. We prospectively observed during follow-up 207 HIV-infected hemophiliacs in the Hemophilia Growth and Development Study. Plasma HIV RNA was measured on cryopreserved plasma from enrollment using the Chiron Corporation bDNA (version 2.0) assay. Genoytpe variants *CCR2b-64I* and *CCR5-Δ32* were detected using standard molecular techniques. Those with the mutant allele for *CCR2b*, and to a lesser extent *CCR5*, had lower plasma HIV RNA, and higher CD4+ T lymphocytes than did those without these genetic variants. After controlling for the effects of plasma HIV RNA and CD4+ T lymphocytes, those with the *CCR2b* mutant allele compared with those wild-type, had a trend toward a lower risk of progression to AIDS, adjusted relative hazard of 1.94 (95% confidence interval [CI], 0.9–4.18; $p = .092$), and AIDS-related death, relative hazard 1.97 (95% CI, 0.98–4.00; $p = .059$). We conclude that plasma HIV RNA, CD4+ T lymphocytes, and *CCR* genotypes are correlated, and the protective affect of *CCR2b* against HIV disease progression is not completely explained by plasma HIV RNA or CD4+ T-lymphocyte number. **Key Words:** Genetic resistance—Host factors—Markers—Viral load—Chemokine receptors.

The rate of disease progression in HIV-1–infected individuals is heterogeneous. Several virologic and host factors have been shown to influence the rate of disease progression from the time of infection to AIDS and

death. Virologic factors include biologic phenotype, in particular the ability to induce syncytium in vitro (1–3) and plasma HIV RNA concentration. Longitudinal studies have demonstrated the relationship between ongoing viral replication and the pathogenesis of HIV disease (4–6), with a single plasma HIV RNA measurement being a potent predictor of disease progression in various different cohorts (7–12). In addition, host factors have received increasing attention for their role in determining clinical outcomes. Younger age at the time of HIV infection and select HLA profiles have been associated with delayed disease progression (13–15). More re-

cently, the identification of genotypic variants for select chemokine receptors, namely *CCR5* and *CCR2b,* has been shown to be protective against progression to AIDS and death. In several studies, the 32 amino acid deletion in *CCR5, CCR5-Δ32,* has been associated with improved clinical outcomes (16–21). Similarly, a mutation in the N-terminus of *CCR2b, CCR2b-64I,* has been associated with delayed progression to AIDS and death in seroincident (22–25), but not seroprevalent cohorts (23,26–28). The mechanism by which genetic variants for these chemokine receptors delay clinical progression has not been delineated. Our study defines the effect and interaction between these chemokine receptor variants, plasma HIV RNA concentration, and CD4$^+$ T-lymphocyte number on HIV disease progression in a cohort of infected hemophiliacs.

## MATERIALS AND METHODS

### Study Population and Samples Stored

The Hemophilia Growth and Development Study (HGDS) is a multicenter, U. S. study that enrolled 207 hemophiliacs who acquired HIV infection from exposure to blood products, in most cases between 1982 and 1983. Enrollment occurred in 1989 and 1990 with study subjects ranging in age from 6 to 19 years. Eligibility for enrollment was based on age (date of birth between September 1970 and September 1982), clotting factor usage and fluency in the English language, given that many neuropsychological test instruments were only written in English. A census was taken at participating centers with a total of 2105 children and adolescents identified. Of the 481 eligible study subjects, 69.2% enrolled. Of those who refused to enroll, most cited the burden of participation as the reason, only 3.5% did so because of being too ill. The racial and ethnic composition of the group was 73% white, 15% Hispanic, 10% African American, and 2% other, similar to that of the general U. S. hemophiliac population (29). Loss to vital status follow-up in this cohort was only 0.48% during the 7 years of follow-up. Although enrolled subjects necessarily excluded those who progressed to death before enrollment (estimated mean infection period, 7 years), there is little other evidence of bias introduced into the study due to selection or loss to follow-up of cohort participants. Additional details of recruitment and the characteristics of this cohort have been reported previously (30). The human subjects' committees of all collaborating institutions approved the HGDS, and informed consent was obtained from parents or legal guardians, and informed consent or assent was obtained from all participants. Every 6 months, a history was obtained, physical examination performed, and blood was taken for lymphocyte subsets and processed within 24 hours for cryopreservation of plasma and mononuclear cells. Lymphoblastoid cell lines were developed by Epstein-Barr virus transformation as a source of cellular DNA.

For the current investigation, enrollment plasma HIV RNA represents either the study enrollment visit or the first visit for which stored plasma was available for viral quantitation. For the current analysis, participants were categorized as having progressed to AIDS or having an AIDS-related death if they met the 1987 Centers for Disease Control and Prevention (CDC) surveillance definition (31), or died during the first 7 years of follow-up, respectively.

### Measurements of Plasma HIV RNA Concentrations

Heparinized plasma stored at −70°C was used for plasma HIV RNA measurements using a sensitive branched DNA (bDNA) assay (version 2.0, Chiron Corporation, Emeryville, CA, U.S.A.). The assay has a lower quantification limit of 500 copies/ml and is linear to concentrations as high as $1.6 \times 10^6$ copies/ml (one copy of HIV RNA = one molecule of HIV RNA) (32,33). The volume of stored plasma was limited; therefore insufficient specimens were available to perform all assays in duplicate. A pilot study was performed on 26 samples in duplicate to ensure reliability of the replicates, demonstrating that reproducibility was high with a mean coefficient of variation between duplicates of 13.9%, and a correlation coefficient between plasma HIV RNA concentration and CD4$^+$ T-lymphocyte number of −0.42 ($p = 0.03$). An additional 174 samples were available for testing as singlicates, with quantification performed on a total of 205 of the 207 enrolled subjects.

### Genotype Variants

The *CCR5-Δ32* allele was detected using nested polymerase chain reaction (PCR) methods previously described (16). The *CCR2b-64I* polymorphism was identified using both single-strand conformation polymorphism/heteroduplex mobility assay and sequence-directed PCR–restriction fragment length polymorphism assays (22).

### Study Variables

We analyzed enrollment plasma HIV RNA concentration, CD4$^+$ T-lymphocyte numbers, and chemokine receptor genotypes for *CCR5* and *CCR2b.* Plasma HIV RNA measurements and genotype data were available on 205 and 169 of the 207 seropositive hemophiliacs, respectively. Thirty-eight individuals were missing *CCR5* and/or *CCR2b* genotype data with 32 missing both, 3 missing just *CCR5,* and 3 missing just *CCR2b.* To account for the missing data, a logistic discriminant function with baseline age and ethnicity (white versus other) as covariates was used to classify participants into appropriate categories (34). For 27 individuals, plasma HIV RNA were collected after they developed AIDS or at the time of their last known AIDS-free date, and therefore these persons were excluded from the analyses involving time to AIDS. Twelve individuals had not met the 1987 AIDS definition at their time of death ($n = 11$), or died of unknown causes ($n = 1$), and therefore were considered censored at time of death.

Genotype categories included *CCR5+/+, CCR5+/Δ32, CCR2b+/+, CCR2b+/64I* and *CCR2b64I/64I.* Disease progression was determined as time to development of AIDS according to the 1987 CDC definition (31), and time to AIDS-related death.

### Statistical Methods

The proportion of study subjects who progressed to AIDS or death, stratified by plasma HIV RNA category (<1000, 1000–9999, and ≥10,000 copies/ml) and CD4$^+$ T-lymphocyte category (<200, 200–499, ≥500 cells/mm$^3$), and chemokine receptor genotype (*CCR2b+/+* and *CCR5+/+* versus *CCR2b+/64I* or *CCR5+/Δ32*) were estimated using the Kaplan-Meier method. Cox proportional hazards regression models were also fitted to assess the independent and joint effects of plasma HIV RNA, CD4$^+$ T lymphocytes, *CCR2b* and *CCR5* genotypes on progression to AIDS or death. Shoenfeld residual plots were used to inspect the assumption of proportional hazards (35). The relationships between chemokine receptor genotype and plasma HIV RNA or CD4$^+$

**TABLE 1.** *The distribution of patients by baseline plasma HIV RNA and CD4+ T-lymphocyte category*

| Marker | N (%) | AIDS[a] N (%) | Death[a] N (%) | Median time to AIDS (y) | Median time to death (y) |
|---|---|---|---|---|---|
| HIV RNA (copies/ml) | | | | | |
| <1000 | 56 (27.3) | 10 (17.9) | 8 (14.3) | 6.27 | 7.01 |
| 1000–9999 | 101 (49.3) | 37 (36.6) | 30 (29.7) | 5.32 | 7.01 |
| ≥10,000 | 48 (23.4) | 32 (66.7) | 34 (70.8) | 2.62 | 3.75 |
| CD4+ T cells (/mm³) | | | | | |
| ≥500 | 76 (37.1) | 17 (22.4 | 9 (11.8) | 6.53 | 7.01 |
| 200–499 | 71 (34.6) | 19 (26.8) | 17 (23.9) | 5.03 | 6.85 |
| <200 | 58 (28.3) | 43 (74.1) | 46 (79.3) | 1.93 | 3.18 |

[a] Participants were categorized as having AIDS if they met the 1987 U.S. Centers for Disease Control and Prevention surveillance definition within the first 7 years of follow-up, and all deaths occurring up to 7 years postenrollment were used in the analysis.

T lymphocytes were examined by comparing the log-transformed plasma HIV RNA concentrations and square root transformed CD4+ T lymphocytes between different genotype groups using two-sample unequal variance *t*-tests.

## RESULTS

At the time of enrollment of the 207 seropositive individuals, mean age was 13.2 years, median CD4+ T-lymphocyte count was 400/mm³, and the median plasma HIV RNA concentration was 3102 copies/ml, with 32 (15.6%) having <500 copies/ml. Enrollment plasma HIV RNA and CD4+ T lymphocytes were significantly correlated with a Spearman rank correlation of −0.46 ($p$ = .0001). By 7 years of follow-up, 79 (38.5%) had progressed to AIDS, and, in total, 72 AIDS-related deaths were reported. The median time from baseline to AIDS and death were 5.01 and 6.47 years, respectively. The distribution of total patients, as well as those who progressed to 1987 AIDS and AIDS-related death by plasma HIV RNA and CD4+ T-lymphocyte category are sum-

marized in Table 1. Table 2 shows the unadjusted and adjusted relative hazard (RH) for progression to AIDS and AIDS-related death, showing that those with plasma HIV RNA concentrations ≥10,000 copies/ml and CD4+ T lymphocytes <200/mm³ were significantly different than the most favorable category in each group. Figure 1A and B show unadjusted data as Kaplan-Meier survival curves by plasma HIV RNA and CD4+ T-lymphocyte category, with Figure 2A and B showing unadjusted AIDS-free survival based on these markers.

Proportional hazards model was fitted with plasma HIV RNA concentrations and CD4+ T-lymphocyte numbers treated as continuous variables, with and without adjustment for CD4+ T-lymphocyte or plasma HIV RNA category. For every threefold increase in plasma HIV RNA, there was an unadjusted RH of progression to AIDS and death of 1.79 and 1.86, respectively. The adjusted RH for AIDS was 1.45 and death 1.41. Similarly, every 100-cell decrease in CD4+ T lymphocytes had an unadjusted RH for progression to AIDS and death of 2.97 and 4.62, respectively, with an adjusted RH for

**TABLE 2.** *Unadjusted and adjusted relative hazard by baseline plasma HIV RNA and CD4+ T-lymphocyte categories*

| Marker | AIDS Unadjusted RH (95% CI) | AIDS Adjusted RH[a] (95% CI) | Death Unadjusted RH (95% CI) | Death Adjusted RH[a] (95% CI) |
|---|---|---|---|---|
| HIV RNA (copies/ml) | | | | |
| <1000 | 1.0 | 1.0 | 1.0 | 1.0 |
| 1000–9999 | 3.33[b] (1.29–8.61) | 2.52 (0.95–6.71) | 2.11 (0.97–4.61) | 1.26 (0.56–2.81) |
| ≥10,000 | 7.74[c] (2.84–21.08) | 4.40[c] (1.51–12.82) | 9.16[c] (4.22–19.90) | 4.04[c] (1.77–9.23) |
| CD4+ T cells(/mm³) | | | | |
| ≥500 | 1.0 | 1.0 | 1.0 | 1.0 |
| 200–499 | 1.57 (0.77–3.18) | 1.22 (0.59–2.53) | 2.26 (1.01–5.08) | 1.78 (0.77–4.10) |
| <200 | 7.16[c] (3.56–14.39) | 4.60[c] (2.18–9.71) | 18.06[c] (8.72–37.39) | 12.92[c] (5.97–27.99) |

[a] Relative hazard adjusted for CD4+ T-lymphocytes or plasma HIV RNA categories.
[b] $p < .05$.
[c] $p < .01$.
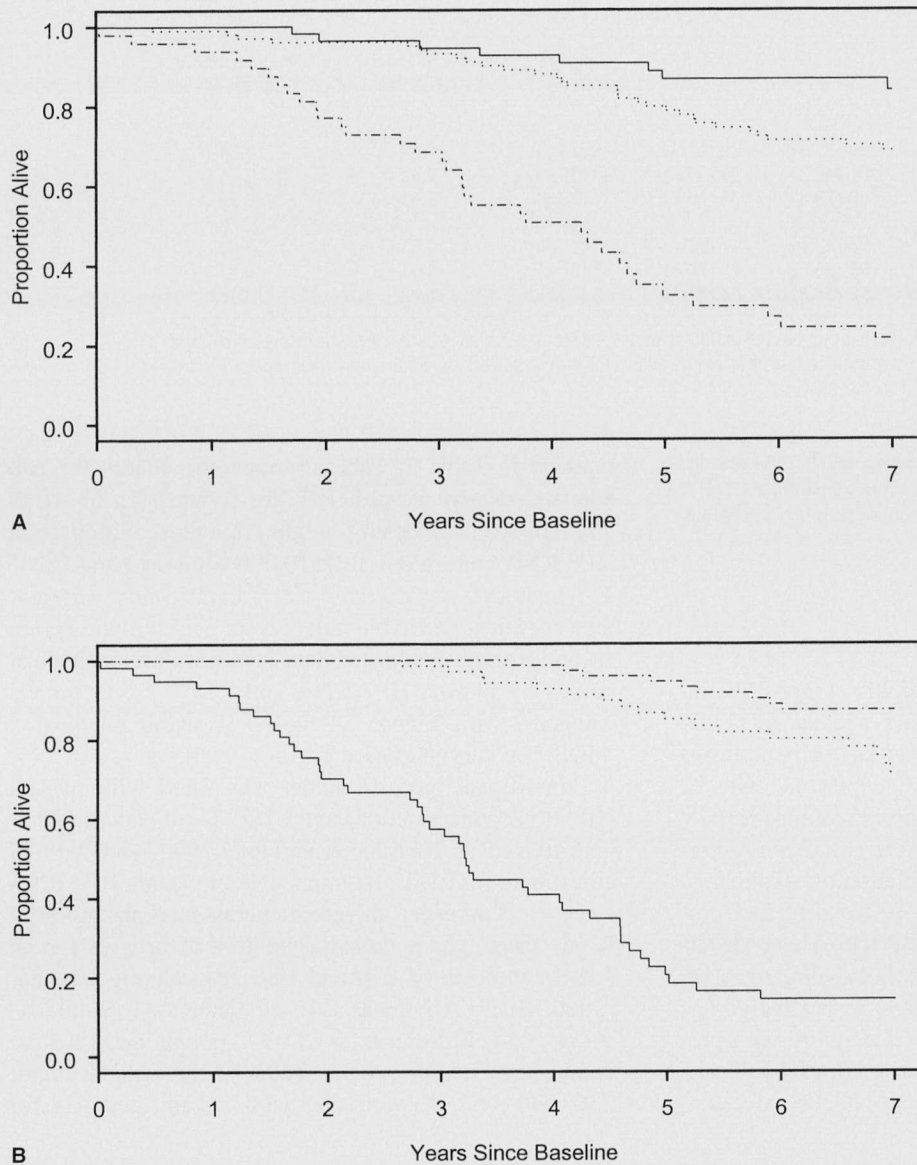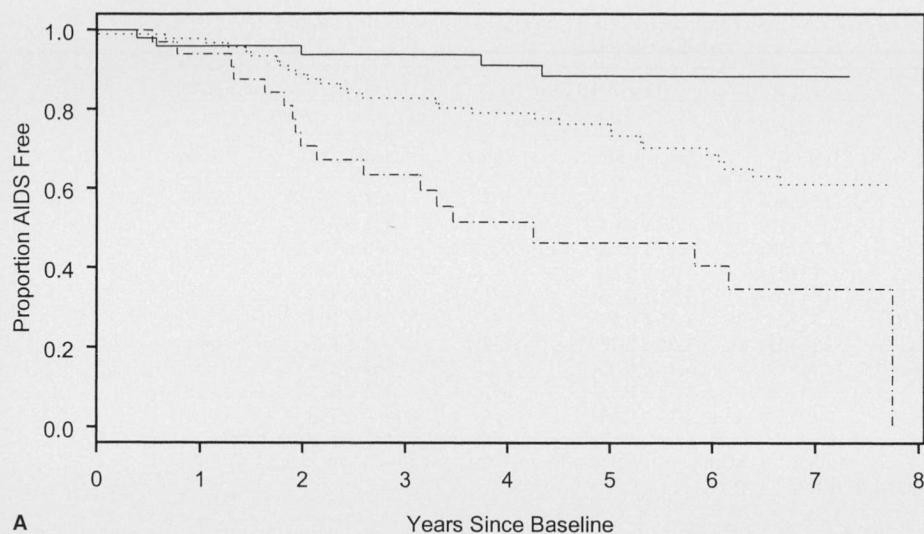RH, relative hazard; CI, confidence intervals.

**FIG. 1.** Kaplan-Meier estimates for progression to AIDS-related death by baseline (**A**) plasma HIV RNA and (**B**) CD4⁺ T lymphocyte category. *Solid line,* <1000 copies/ml and <200 cells/mm³; *dotted line,* 1000–9999 copies/ml and 200–499 cells/mm³; *dots-dashes line,* >10000 copies/ml and >500 cells/mm³. Relative hazards and *p* values are summarized in Table 2.

AIDS of 2.19 and 4.05 for death. All RH were statistically significant at $p < .01$.

*CCR5* and *CCR2b* genotypes for the HGDS study subjects have been previously described as part of larger studies (16,22). We characterized the study subjects in this cohort to determine the interactions between enrollment CD4⁺ T-lymphocyte number, plasma HIV RNA concentration, and chemokine receptor genotype. Table 3 summarizes the distribution of genotype variants for the cohort, and their relationship with enrollment plasma HIV RNA and CD4⁺ T-lymphocyte number. These analyses show that there are lower mean plasma HIV RNA and higher mean CD4⁺ T-lymphocyte numbers in all groups containing a *CCR2b* or *CCR5* mutant allele. In

fact, statistically higher mean CD4⁺ T-lymphocyte counts were found in all groups with the *CCR2b-64I* allele ($p < .05$), with nonsignificant differences observed in the *CCR5-Δ32* containing study subjects. Of the viral comparisons, there were trends toward lower plasma HIV RNA in the groups with the *CCR2b-64I* variant ($p = .07$ to .09) but not in those with the *CCR5-Δ32* allele compared with wild-type ($p \geq .4$). We further determined the interaction between chemokine receptor genotype, plasma HIV RNA concentration, CD4⁺ T-lymphocyte number, and disease progression. Kaplan-Meier survival curves comparing study subjects heterozygous for either chemokine receptor genotype variant to that wild-type for both for progression to AIDS
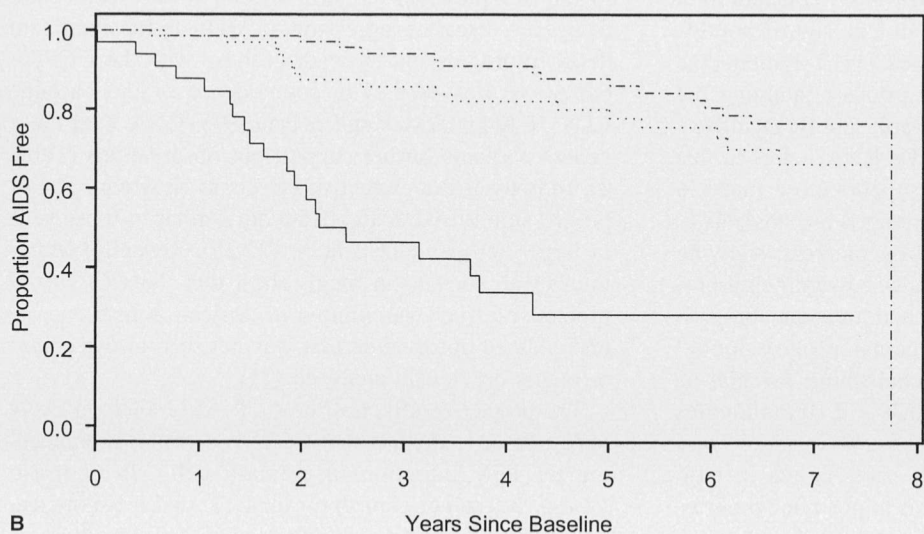
**FIG. 2.** Kaplan-Meier estimates for progression to the 1987 U.S. Centers for Disease Control and Prevention case definition of AIDS by baseline (**A**) plasma HIV RNA and (**B**) CD4$^+$ T lymphocyte category. *Solid line,* <1000 copies/ml and <200 cells/mm$^3$; *dotted line,* 1000–9999 copies/ml and 200–499 cells/mm$^3$; *dots-dashes line,* >10000 copies/ml and >500 cells/mm$^3$. Relative hazards and *p* values are summarized in Table 2.

and AIDS-related death (Fig. 3). When the effect of each of these markers was assessed simultaneously, threefold increases in plasma HIV RNA were associated with a 1.41 to 1.45 times increased risk of progression to AIDS ($p \leq .004$) and a 1.37 to 1.41 times increased risk of progression to AIDS-related death ($p \leq .008$). Similarly, 100-cell decreases in CD4$^+$ T-lymphocyte number were associated with a 2.19 to 2.31 times increased risk of progression to AIDS ($p \leq .002$) and 4.09 to 4.26 times increased risk of progression to death ($p < .001$).

The Unadjusted and adjusted RH for HIV disease progression by chemokine receptor genotype is summarized in Table 4. These analyses show that although no statistically significant protection from progression to AIDS

($p = .77$) or death ($p = .55$) is seen in those with the *CCR5-Δ32* allele after adjusting for plasma HIV RNA and CD4$^+$ T-lymphocyte number, there is a trend toward delayed progression to AIDS in those with the *CCR2b-64I* variant ($p = .092$). A somewhat stronger relationship was seen for survival in the *CCR2b-64I* group compared with the wild-type ($p = .059$) group and a significant protective effect for AIDS-related death seen in other comparisons between *CCR2b-64I*–containing groups ($p < .05$).

## DISCUSSION

This study shows that CD4$^+$ T-lymphocyte number and plasma HIV RNA concentration predict HIV disease

**TABLE 3.** *Plasma HIV RNA concentration and CD4⁺ T-lymphocyte number by chemokine receptor genotype*

| Genotype | N (%) | Log$^{10}$ (HIV RNA) (copies/ml) Mean ± SD | p Value | CD4⁺ T lymphocytes (/mm$^3$) Mean ± SD | p Value |
|---|---|---|---|---|---|
| *CCR5+/+* | 174 (85) | 3.48 ± 0.71 | .404 | 405 ± 321 | .646 |
| *CCR5+/Δ32* | 31 (15) | 3.37 ± 0.64 | | 403 ± 239 | |
| *CCR2b+/+* | 160 (78) | 3.50 ± 0.72 | .070 | 380 ± 304 | .023 |
| *CCR2b+/64I* | 45 (22) | 3.31 ± 0.60 | | 496 ± 318 | |
| *CCR5+/+, CCR2b+/+* | 134 (65) | 3.52 ± 0.73 | .071 | 379 ± 315 | .045 |
| *CCR5+/Δ32, CCR2b+/64I* | 71 (35) | 3.35 ± 0.61 | | 455 ± 295 | |
| *CCR5+/+, CCR2b+/+* | 134 (65) | 3.52 ± 0.72 | .437 | 379 ± 315 | .614 |
| *CCR5+/Δ32, CCR2b+/+* | 26 (13) | 3.41 ± 0.63 | | 384 ± 239 | |
| *CCR5+/+, CCR2b+/+* | 134 (65) | 3.52 ± 0.73 | .092 | 379 ± 315 | .038 |
| *CCR5+/+, CCR2b+/64I* | 40 (20) | 3.33 ± 0.59 | | 494 ± 330 | |

+/+, homozygote for wild-type allele for chemokine receptors; +/−, heterozygote for mutant alleles *CCR5-Δ32* or *CCR2b-64I;* SD, standard deviation.

progression in a cohort of infected children and adolescents (Table 2; Figs. 1 and 2). These analyses demonstrate that the *CCR5* and *CCR2b* chemokine receptor genotypes are related to plasma HIV RNA concentration and CD4⁺ T-lymphocyte number. In fact, significant differences were observed between the CD4⁺ T-lymphocyte number in all comparisons with groups containing the *CCR2b-64I* allele and trends toward significant differences were seen in the plasma HIV RNA levels in this group (Table 3). These findings support those made in other cohorts. Furthermore, we extended our analysis to define the interaction between these markers, showing that the *CCR2b* mutant allele trended toward being protective from progression to AIDS and, in some comparisons, significantly protective against progression to AIDS-related death, even after controlling for plasma HIV RNA concentration and CD4⁺ T lymphocytes (Table 4).

The *CCR5+/Δ32* genotype has been shown in individual studies and in a metaanalysis to postpone progression to AIDS (16–19,21) and to be associated with lower plasma HIV RNA and higher CD4⁺ T lymphocytes (17, 19,36). Similarly, the presence of the *CCR2b-64I* mutant allele has been shown to postpone progression to AIDS and death by 2 to 4 years in seroincident (22–25), but not in seroprevalent cohorts (23,26–28,36). The lack of effect seen in previous studies using seroprevalent cohorts is likely to be a result of the exclusion of rapid progressors and imprecise estimation of the timing of seroconversion. Thus, it has been suggested that such cohorts not be used for studies to assess the protective effect of genetic variants. Such conclusions probably apply to seroprevalent cohorts of homosexual men, in whom the timing of infection varies. In contrast, hemophiliac cohorts are unique in that infection occurred during a relatively narrow window of time, between 1982 and 1983, allow-

ing for a fairly precise estimation of the timing of seroconversion. Because of this, and despite the relatively small cohort and that rapid progressors are not included in our seroprevalent cohort, a trend toward protection from HIV disease progression to AIDS and a significant delay in progression to death in those with the *CCR2b-64I* allele was seen. The concordance of the baseline CD4⁺ T lymphocytes and plasma HIV RNA with these genetic variants further support our observations (Table 3). Moreover, the protective effects as shown by RH of progression to AIDS and death are similar to those seen in larger seroincident cohorts (22,25). Recently, Mummidi et al. showed in their cohort that the *CCR2b-64I* protective effect was limited to African Americans, an unexplained observation that was not seen in our cohort or others previously analyzed (25).

The protective effect of the *CCR5-Δ32* allele is likely to be related to the fact that *CCR5* is a principal coreceptor for HIV infection of primary cells. In contrast, *CCR2b* is a minor coreceptor for HIV, and it is not clear why such a pronounced effect on disease progression would occur. Proposed mechanisms for protection include the role of this receptor in the transition of non–syncytium-inducing to syncytium-inducing isolates (37–40), the possible effect on the cellular expression of *CCR5* (41,42), or even indirect effects on HIV replication (38). In addition, Kostrikis et al. recently suggested that the effect could be related to a polymorphism identified in the *CCR5* promoter region of all study subjects with the *CCR2b-64I* allele (23). Although none of these mechanisms have been established in vivo, the protective effect of the *CCR2b-64I* allele is apparent. From a clinical perspective, it is most important to establish whether the presence of genotypic variants provide additional prognostic information to other established markers, such as plasma HIV RNA and CD4⁺ T-lymphocyte num-
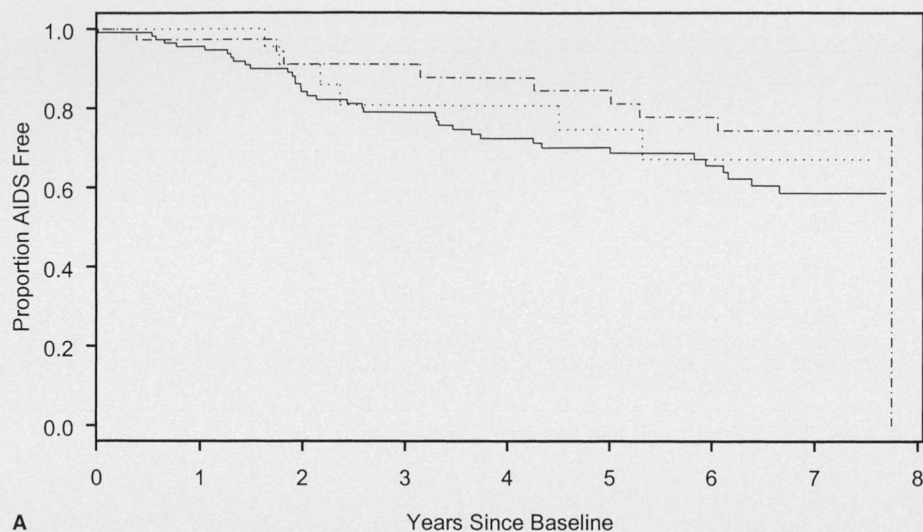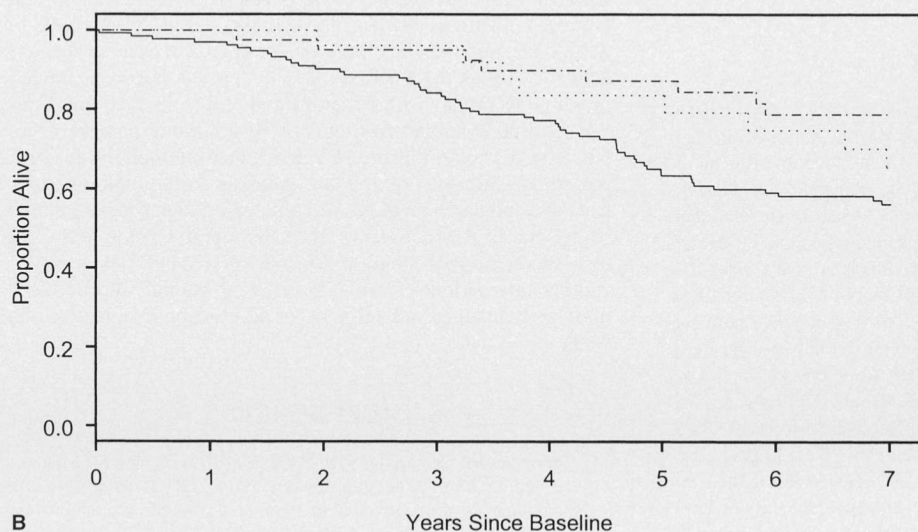
**FIG. 3.** Kaplan-Meier estimates of proportion of patients (**A**) AIDS-free and (**B**) surviving when comparing those with either the CCR2b+/64I or CCR5+/Δ32 genotypes with those wild-type for both (CCR2b+/+ and CCR5+/+). Solid line, CCR2b+/+ and CCR5+/+; dotted line, CCR2b+/+ and CCR5+/Δ32; dots-dashes line, CCR2b+/64I and CCR5+/+. Relative hazard and p values are summarized in Table 4.

ber. Recent studies have described a relationship between the CCR2b-64I allele, CD4+ T-lymphocyte decline and plasma HIV RNA, suggesting that the beneficial effects of this genetic variant is mediated by its effect on these markers (23,24). In the current study, we show a similar relationship between these markers. Moreover, we extended our analysis to show that the protective effect of the CCR2b-64I allele from progression to AIDS and particularly AIDS-related death appears to persist even after controlling for plasma HIV RNA and CD4+ T-lymphocyte number. Together, these findings may provide new insight into the pathogenesis of HIV disease as well as establishing this genotypic variant as an independent predictor of disease progression.

Various interacting virologic and host factors determine the rate of disease progression in each HIV-infected individual. Although the CCR2b and CCR5 chemokine receptor variants have been shown to be protective, they do not explain all heterogeneity observed in the natural history of HIV infection. The inability to control for other known and unknown factors that influence progression to AIDS and death is likely to explain conflicting results across studies (43). Nevertheless, previously reported studies, which include multiple cohorts, have affirmed the significant effects of these chemokine receptor variants in the background of multiple confounders, such as race, HLA, age and undiscovered cofactors (16,22). Further defining the mechanisms by which host

**TABLE 4.** *Unadjusted and adjusted relative hazards of AIDS and death by chemokine receptor genotype*

| | AIDS | | | | Death | | | |
|---|---|---|---|---|---|---|---|---|
| Genotype | Unadjusted RH (95% CI) | p Value | Adjusted RH[a] (95% CI) | p Value | Unadjusted RH (95% CI) | p Value | Adjusted RH[a] (95% CI) | p Value |
| CCR5+/+ | 1.42 | .416 | 1.14 | .767 | 1.34 | .417 | 1.24 | .549 |
| CCR5+/Δ32 | (0.61–3.34) | | (0.48–2.70) | | (0.66–2.69) | | (0.61–2.53) | |
| CCR2b+/+ | 1.91 | .094 | 1.94 | .092 | 2.26 | .022 | 1.97 | .059 |
| CCR2b+/64I | (0.90–4.07) | | (0.90–4.18) | | (1.12–4.54) | | (0.98–4.00) | |
| CCR5+/+, CCR2b+/+ | 1.71 | .089 | 1.62 | .131 | 1.98 | .014 | 1.88 | .027 |
| CCR5+/Δ32, CCR2b+/64I | (0.92–3.16) | | (0.87–3.04) | | (1.15–3.42) | | (1.07–3.28) | |
| CCR5+/+, CCR2b+/+ | 1.33 | .522 | 1.16 | .739 | 1.51 | .276 | 1.60 | .228 |
| CCR5+/Δ32, CCR2b+/+ | (0.56–3.14) | | (0.49–2.77) | | (0.72–3.17) | | (0.75–3.41) | |
| CCR5+/+, CCR2b+/+ | 1.78 | .142 | 1.86 | .119 | 2.44 | .018 | 2.35 | .026 |
| CCR5+/+, CCR2b+/64I | (0.83–3.82) | | (0.85–4.06) | | (1.16–5.13) | | (1.11–4.99) | |

[a] Relative hazards adjusted for plasma HIV RNA CD4$^+$ T lymphocytes, and chemokine receptor genotype.
+/+, homozygote for wild-type allele for chemokine receptors; +/−, heterozygote for mutant alleles; RH, relative hazard; CI, confidence intervals.

factors delay HIV disease progression will not only advance our understanding of HIV immunopathogenesis, but it may also lead to novel therapeutic approaches in the future (44).

## APPENDIX

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