Correlation between Human Immunodeficiency Virus Genotypic Resistance and Virologic Response in Patients Receiving Nelfinavir Monotherapy or Nelfinavir with Lamivudine and Zidovudine

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The relationship between detectable human immunodeficiency virus (HIV) genotypic resistance and virologic response was compared in patients receiving nelfinavir as monotherapy (16 weeks) or in combination with lamivudine and zidovudine (48 weeks). Two patient groups were defined on the basis of the presence or absence of substitutions associated with nelfinavir, a protease (PR) inhibitor, and/or a reverse transcriptase (RT) inhibitor. HIV RNA levels <50 copies/mL were achieved in 17 (85%) of 20 combination-therapy patients without genotypic resistance (PR-RT- versus only 1 (17%) of 6 patients with genotypic resistance (PR-RT+). PR-RT- patients exhibited greater and more durable virus suppression compared with PR-RT+ patients. All 6 PR-RT+ patients had virus with M184V (lamivudine resistance); 3 isolates also contained D30N (nelfinavir resistance). M184V preceded D30N in all determinable instances. In this study, suppression of HIV replication to <50 copies/mL was associated with durable response and reduced incidence of resistance. Results also indicate that combination regimens can fail despite the absence of detectable genotypic PR resistance.

Administration of antiretroviral regimens containing protease (PR) inhibitors (PIs) in combination with reverse transcriptase (RT) inhibitors (RTIs) has resulted in significant and sustained reductions in plasma HIV RNA levels and increases in CD4 cell counts among patients infected with human immunodeficiency virus (HIV) [1]. Despite the potency of such highly active treatment regimens, some patients experience a loss of suppression of viral replication, which has led to the emergence of resistant HIV variants. Such variants contain amino acid substitutions in PR and/or RT and have demonstrated cross-resistance to all classes of antiretroviral agents currently approved for treatment of patients infected with HIV [2, 3]. In vitro studies have confirmed the association of the majority of these PR and RT substitutions with reduced PI and RTI susceptibility [4].

Nelfinavir mesylate is a peptidomimetic inhibitor of HIV PR with potent in vitro activity against laboratory, clinical, and RTI-resistant HIV strains [5, 6]. In clinical trials, nelfinavir treatment is associated with prolonged viral suppression and immunologic improvement and has thus become an integral component of highly active antiretroviral therapeutic regimens [7–11]. In patients experiencing virologic relapse while being treated with nelfinavir-containing regimens, the predominant amino acid substitution in viral PR associated with phenotypic resistance to nelfinavir occurs at residue 30 (D30N) [12, 13]. This substitution has occasionally been observed together with additional changes in PR (e.g., E35D, M36I, M46I, A71V/T, V77I, and N88D/S). Substitutions that have been shown to confer resistance to other PIs, including saquinavir, ritonavir, indinavir, and amprenavir, however, have either never (G48V, I50V, V82F/T, and I84V) or only occasionally (L90M) been observed in patients treated with nelfinavir [12,13].

Specific substitutions conferring resistance to the RTIs lamivudine and zidovudine have also been detected in HIV RT genes isolated from patients treated with these drugs either as monotherapy or in combination with other antiretroviral agents [14–16]. A single substitution at residue 184 (M184V) has been shown to confer high-level resistance to lamivudine. This substitution arises as early as 2 months after patients begin receiving lamivudine as monotherapy or in combination with zidovudine [17–19]. In contrast, the pattern of amino acid substitutions in RT during treatment with zidovudine is more complex and involves substitutions at residues 41, 67, 70, 215, and 219 that arise in patient isolates in a treatment duration-dependent manner [20, 21].

Although critical substitutions associated with PI and RTI resistance have been detected in isolates obtained from patients who experience virologic rebound during antiretroviral therapy [1, 2, 12–21], comprehensive analyses correlating the appear-
ance of these substitutions with virologic response and/or failure have only recently been undertaken. In this study, we evaluated the relationship between detectable viral genotypic changes and virologic response among patients who received nelfinavir either as monotherapy or in combination with lamivudine and zidovudine in 2 separate phase II/III controlled clinical trials.

Materials and Methods

Patients. Patients in this study were drawn from 2 nelfinavir-containing phase II/III controlled clinical trials. They included all 32 patients who received 750 mg nelfinavir 3 times daily for 12 or 16 weeks as monotherapy (study AG1343-505) [9] and a randomly selected group of 26 patients (every fourth patient from a total of 99) who received 750 mg nelfinavir 3 times daily in combination with lamivudine and zidovudine for periods ≤48 weeks (study AG1343-511) [8]. Entry criteria for patients in both studies included plasma HIV RNA ≥15,000 copies/mL by HIV branched-DNA (bDNA) or polymerase chain reaction (PCR) analysis. Patients (10 of 32) originally assigned to receive placebo in study AG1343-505 were reassigned to the nelfinavir (750-mg, 3 times daily) treatment arm after 4 weeks. Patients enrolled in study AG1343-505 were PI-naive but could be RTI-experienced, whereas patients in study AG1343-511 were PI- and RTI-naive (zidovudine for <1 month).

HIV genotype analysis. Population-based nucleotide sequences of HIV PR and RT, obtained from patient plasma virus, were determined by Professional Genetics Laboratory (Uppsala, Sweden) [22], at 16 weeks for patients from study AG1343-505 and at 48 weeks for patients from study AG1343-511. Population-based sequencing was also performed by Agouron Pharmaceuticals (San Diego) at selected time points among patients from study AG1343-511. In brief, a cDNA template was generated by RT-PCR. A 2-step PCR was then used to separately amplify the entire PR gene or the 5′ region (687 nucleotides or the first 229 amino acid residues) of RT. Alternatively, a 2-step PCR was used to amplify a 1.5-kb fragment encompassing the entire PR and the first 330 amino acid residues of RT. PCR-amplified PR and RT gene products were directly sequenced using the AutoLoad (Amersham Pharmacia Biotech, Piscataway, NJ) solid-phase sequencing kit with T7 polymerase and/or using the ThermoSequenase Cycle Sequencing kit (Amersham Pharmacia Biotech) and Cy5-labeled sequencing primers. The 1.5-kb PR-RT PCR product was sequenced using the ThermoSequenase Cycle Sequencing kit and Cy5-labeled primers. Sequencing assays were performed using the ALFExpress automated DNA sequencer (Amersham Pharmacia Biotech) and were analyzed with Mutation Analyzer (Amersham Pharmacia Biotech), DNASTAR analysis software (DNASTAR, Madison, WI), and visual evaluation. Specific amino acid substitutions were identified by comparison with the HIV North American clade B sequence as master sequence [23]. Baseline DNA sequence analysis was determined for 2 patients (PR”) who carried L90M after 12 weeks of nelfinavir monotherapy (study AG1343-505). Baseline DNA sequence analysis was determined for 6 patients (PR-RT”) who developed genotypic resistance after 20 weeks (n = 1) or 48 weeks (n = 5) of treatment with nelfinavir in combination with lamivudine and zidovudine (study AG1343-511). GenBank accession numbers are AF247855–AF247932.

Clonal analysis of viral PR and RT sequences. The linkage relationship between PR and RT substitutions was confirmed through sequence analysis of multiple molecular clones generated by Agouron Pharmaceuticals. In brief, cDNA synthesis and 2-step PCRs were performed in independent triplicate reactions to generate a 1.5-kb PCR product containing PR and the first 330 amino acid residues of RT as described above. Aliquots of each PCR product were ligated into a pGEM-T Easy plasmid vector (Promega, Madison, WI). After transformation, single independent recombinant plasmids containing PR and RT sequences on a contiguous DNA molecule were chosen for sequence analysis as described above.

HIV RNA quantitation. The amount of plasma HIV RNA present in samples obtained from patients who received nelfinavir monotherapy was determined using the bDNA Quantiplex assay (Chiron, Emeryville, CA) with a lower limit of detection (LOD) of 500 HIV bDNA copies/mL. Quantitation of HIV RNA in plasma samples from patients who received nelfinavir in combination with lamivudine and zidovudine was performed using both a Roche PCR assay (Roche Diagnostic Systems, Branchburg, NJ), which has an LOD of 400 HIV RNA copies/mL, and the Ultra-sensitive HIV PCR (Roche Diagnostic Systems), which has an LOD of 50 HIV RNA copies/mL.

Statistical analyses. Determination of significance in this study was made using analysis of variance and a log rank test.

Results

Incidence of genotypic nelfinavir resistance in patients who received nelfinavir monotherapy (study AG1343-505). To determine the incidence of nelfinavir resistance–associated genotypic changes, DNA sequence analysis was performed on the entire PR gene amplified from plasma HIV RNA from all 32 patients who had received 12 or 16 weeks of nelfinavir monotherapy. PR genes amplified from 20 of 32 (62.5%) patient isolates contained either the D30N substitution (n = 18 [56%]) or the L90M substitution (n = 2 [6%]; PR” patients), whereas PR genes from 32 (37.5%) of 32 patient isolates contained no primary resistance–associated substitutions (PR’ patients). The L90M substitution was present before nelfinavir therapy in 1 of the 2 patients, which suggests prior therapy with another PI. Other predominant substitutions selected after nelfinavir therapy were detected at residues 46 (M46I or M46L) and 88 (N88D or N88S) in 6 (19%) of 32 and 8 (25%) of 32 patients, respectively. With the exception of 2 patient isolates that contained the N88S substitution alone, substitutions at residues 46 and/or 88 were always associated with D30N and/or L90M. Substitutions associated with resistance to other PIs (e.g., at residues 48, 50, 82, and 84) were not detected. A range of 1–11 secondary genotypic changes (e.g., at residues 10, 13, 15, 19, 36, 37, 41, 62, 63, 64, 77, and 93), which are described elsewhere as polymorphisms [13], were detected in all patient isolates examined. PR sequences could not be PCR amplified from the plasma of 4 (13%) of 32 patients. These 4 patients were included in the PR patient group.
Genotypic resistance correlates with virologic response in patients who received nelfinavir monotherapy (study AG1343-505). Subsequent analyses evaluated the relationship between detectable genotypic resistance and virologic response among the 32 patients (table 1). Although a significantly higher baseline virus load was observed for the PR\(^+\) patient group when compared with the PR\(^-\) patient group (5.1 vs. 4.7 \(\log_{10}\) bDNA copies/mL; \(P = .05\)), no significant difference in baseline CD4 cell counts was observed (\(P = .27\)). Additional analyses revealed comparable levels of initial antiviral response among both patient groups as reflected by similar mean maximum reductions in plasma HIV RNA (\(-1.6 [\text{PR}^+]\) vs. \(-1.5 [\text{PR}^-] \log_{10}\) bDNA copies/mL; \(P = .59\); data not shown) and in the time required to achieve maximum HIV RNA reduction (\(P = .42\); data not shown). The emergence of virus containing D30N or L90M substitutions in PR\(^+\) patients was, however, associated with a significantly greater loss in virus suppression at 16 weeks when compared with PR\(^-\) patients whose isolates contained no critical genotypic changes (\(-0.4 \text{ vs. } -1.0 \log_{10}\) bDNA copies/mL; \(P = .01\); table 1). Virologic rebound (defined as achieving 2 consecutive time points of HIV bDNA 0.5 log\(_{10}\) above the nadir value within the first 24 weeks of treatment; data not shown) was also observed for 5 of 12 PR\(^+\) patients (2 patients whose isolates contained the N88S substitution and 3 patients whose isolates contained 1–6 secondary genotypic changes at residues 15, 37, 41, 57, 63, 64, 77, or 93).

No significant difference in CD4 cell count increases at 16 weeks was observed between the 2 patient groups (\(P = .28\)).

Incidence of genotypic resistance in patients who received nelfinavir in combination with lamivudine and zidovudine (study AG1343-511). To determine the incidence of nelfinavir resistance–associated genotypic changes in PR, and of lamivudine and zidovudine resistance–associated genotypic changes in RT, DNA sequence analysis of PR and RT genes was performed for 26 randomly selected patients who received nelfinavir in combination with lamivudine and zidovudine. Twenty-two of the 26 patients received this nelfinavir-containing regimen for 48 weeks. DNA sequence analysis was performed at 16, 20, 28, or 46 weeks for 4 of the 26 patients who discontinued treatment at these time points. Results indicated that virus isolates from 6 (23\%) of 26 patients contained substitutions in PR and/or RT (PR-RT\(^+\); table 2). Virus isolates from all 6 of these patients contained the M184V lamivudine resistance–associated amino acid substitution in RT. Three of the 6 patient isolates also carried the D30N substitution in PR in association with other secondary genotypic changes. Two patients also had viruses that contained zidovudine resistance–associated genotypic changes M41L/M or K219K/R; 1 change was associated with M184V, and 1 change was associated with both D30N and M184V. Secondary PR substitutions (not present at baseline) that were selected after nelfinavir combination therapy included I15V, M36I/L, and R41K (patient 2), I62I/V (patient 3), and M46L, I54I/L, N88I/S, and I93L (patient 4). Twenty (77\%) of 26 patients were designated as PR-RT\(^+\) because their isolates contained either no primary resistance–associated genotypic changes (\(n = 3\) for PR and \(n = 1\) for RT) or were negative by PCR analysis (\(n = 17\) for PR and \(n = 19\) for RT). The L90M substitution in PR, or other zidovudine resistance–associated substitutions (e.g., at residues 67, 70, or 215 in RT), was not detected in any of these 26 patients after 16–48 weeks of nelfinavir combination therapy.

Linkage relationship between D30N and M184V amino acid substitutions (study AG1343-511). To determine whether D30N and M184V substitutions detected in the 3 patients described above existed on a single viral genomic RNA molecule or were instead single substitutions that occurred within individual viral genomes, sequence analysis was performed using independently generated, subcloned PCR-amplified DNAs that

### Table 1. Baseline and 16-week plasma human immunodeficiency virus (HIV) RNA and CD4 cell counts in patients who received nelfinavir monotherapy.

<table>
<thead>
<tr>
<th>Time, parameter</th>
<th>PR(^+) ((n = 20))</th>
<th>PR(^-) ((n = 12))</th>
<th>(P^a)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baseline HIV RNA (\log_{10}) bDNA copies/mL</td>
<td>5.1 (4.3–6.1)</td>
<td>4.7 (4.0–5.3)</td>
<td>.05</td>
</tr>
<tr>
<td>CD4 count cells/(\mu L)</td>
<td>257 (55–601)</td>
<td>319 (121–521)</td>
<td>.27</td>
</tr>
<tr>
<td>16 Weeks HIV RNA reduction (\log_{10}) bDNA copies/mL</td>
<td>-0.4 (-1.8–0.1)</td>
<td>-1.0 (-2.2–0.0)</td>
<td>.01</td>
</tr>
<tr>
<td>CD4 count increase cells/(\mu L)</td>
<td>90 (-38–320)</td>
<td>60 (-86–165)</td>
<td>.28</td>
</tr>
</tbody>
</table>

**NOTE.** Data are mean (range). Clinical responses were evaluated for patients whose isolates contained nelfinavir-associated genotypic changes (PR\(^+\)) and for patients whose isolates did not (PR\(^-\)), when genotyped after 12 or 16 weeks of nelfinavir monotherapy (AG1343-505). PR, protease; bDNA, branched DNA.

\(^a\) Significance determined by analysis of variance.

### Table 2. Summary of protease (PR) and reverse transcriptase (RT) inhibitor–associated genotypic changes in patients who received nelfinavir in combination with lamivudine and zidovudine

<table>
<thead>
<tr>
<th>Patient</th>
<th>PR D30(^a)</th>
<th>M41</th>
<th>M184</th>
<th>K219</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>N</td>
<td>V</td>
<td>V</td>
<td>V</td>
</tr>
<tr>
<td>2</td>
<td>D/N</td>
<td>M/L</td>
<td>V</td>
<td>V</td>
</tr>
<tr>
<td>3</td>
<td>N</td>
<td>V</td>
<td>V</td>
<td>K/R</td>
</tr>
<tr>
<td>4</td>
<td>M46L</td>
<td>M/I</td>
<td>M/I</td>
<td>M/I</td>
</tr>
<tr>
<td>5</td>
<td>PCr</td>
<td>V</td>
<td>V</td>
<td>V</td>
</tr>
</tbody>
</table>

**NOTE.** Sequence analysis was performed on human immunodeficiency virus (HIV) PR and RT genes obtained from plasma samples derived from patients who received 48 weeks (patients 1–6) or 20 weeks (patient 5) of nelfinavir (750 mg 3 times daily) with lamivudine and zidovudine (study AG1343-511). N, D30N; V, M184V; D, D30D; M, M41M; L, M41L; K, K219K; R, R219R; M, M184V; I, M184I; PCR, polymerase chain reaction.

\(^a\) Although the entire HIV PR gene was sequenced, only substitutions at residue 30 are indicated. Seventeen (65\%) of 26 patients were PCR negative. Secondary PR substitutions that were not present at baseline but that were selected after nelfinavir combination therapy were as follows: patient 2, H55V, M36I/L, and R41K; patient 3, I62I/V; patient 4, M46L, I54I/L, N88I/S, and I93L.

\(^b\) Although the first 687 nucleotides (229 amino acid residues) of RT were sequenced, only substitutions at residues 41, 184, and 219 are indicated. Nineteen (73\%) of 26 patients were PCR negative.
spanned both the PR and the RT genes. Nine of 10, 10 of 10, and 10 of 10 subcloned sequences (data not shown) amplified from plasmas of these patients (patients 1, 2, and 3, table 2; figure 1A, B, and C, respectively) carried both D30N and M184V substitutions. The K219R substitution was detected in only 1 patient (patient 4, table 2; figure 1D) and was linked with M184V in 1 of 9 clones sequenced (9 of 9 contained M184V; data not shown).

Temporal appearance of PR and RT genotypic changes

To determine the temporal appearance of genotypic changes in PR and RT, DNA sequence analysis was performed on plasma samples obtained from the 6 PR-RT patients (patients 1–6, table 2; figure 1A–F). Results indicate that the lamivudine resistance–associated RT substitution M184V was detected in the absence of, or before, the appearance of the D30N substitution in all patients (4 of 6) for whom a temporal relationship could be determined (patients 1, 3, 4, and 5, table 2; figure 1A, C, D, and E). The M184V substitution

Figure 1. Plasma human immunodeficiency virus (HIV) RNA in 6 patients who received the protease (PR) inhibitor nelfinavir in combination with lamivudine and zidovudine and whose isolates contained D30N and/or genotypic changes in reverse transcriptase (RT). A, Patient 1. B, Patient 2. C, Patient 3. D, Patient 4. E, Patient 5. F, Patient 6. Patient plasma HIV RNA levels were measured at the indicated times (●) using a polymerase chain reaction (PCR) assay with a lower limit of quantitation of 400 (2.6 log₁₀) HIV RNA copies/mL, as indicated by the solid line. DNA sequence analysis of PR and RT genes was performed as described in Materials and Methods. WT, wild type.
Table 3. Baseline and 48-week plasma human immunodeficiency virus (HIV) RNA and CD4 cell counts in patients who received nelfinavir in combination with lamivudine and zidovudine.

<table>
<thead>
<tr>
<th>Time, parameter</th>
<th>PR-RT⁺ (n = 20)</th>
<th>PR-RT⁻ (n = 6)</th>
<th>P*</th>
</tr>
</thead>
<tbody>
<tr>
<td>HIV RNA (log₁₀, copies/mL)</td>
<td>5.2 (4.6–5.9)</td>
<td>5.5 (4.6–5.9)</td>
<td>.44</td>
</tr>
<tr>
<td>CD4 count (cells/mm³)</td>
<td>206 (58–500)</td>
<td>309 (10–662)</td>
<td>.26</td>
</tr>
<tr>
<td>48 Weeks:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HIV RNA reduction (log₁₀)</td>
<td>−1.6 (−4.1–0)</td>
<td>−2.4 (−3–0.41)</td>
<td>.02</td>
</tr>
<tr>
<td>CD4 count increase (cells/mm³)</td>
<td>155 (51–196)</td>
<td>177 (−155–430)</td>
<td>.72</td>
</tr>
</tbody>
</table>

NOTE. Data are mean (range). Clinical responses were evaluated for patients whose isolates contained genotypic changes associated with the protease (PR) inhibitor nelfinavir and/or a reverse transcriptase (RT) inhibitor (PR-RT⁻) and for patients whose isolates did not (PR-RT⁺). Patient samples were genotyped at 16, 20, 28, 46, or 48 weeks. Of 6 PR-RT⁻ patients, 1 discontinued treatment at 20 weeks. Of 20 PR-RT⁺ patients, 3 discontinued treatment at 16 weeks, 28 weeks, and 46 weeks, respectively. CD4 count and plasma HIV type 1 RNA levels at these time points were included in these analyses (AG1343-511).

* Significance determined by analysis of variance.

Discussion

In this study we evaluated the relationship between detectable genotypic resistance and virologic response in patients treated with nelfinavir alone or in combination with lamivudine and zidovudine. In patients receiving nelfinavir monotherapy, a correlation between high baseline HIV RNA and the presence of HIV isolates that subsequently contained critical amino acid substitutions was demonstrated. A larger virus pool at baseline could provide more opportunities for errors during reverse transcription [5, 6, 24–26], resulting in greater numbers of HIV variants that are capable of replication in the presence of nelfinavir. The higher virus load observed at baseline is also consistent with the significantly greater loss of virus suppression seen at 16 weeks in PR⁻ patients compared with PR⁺ patients. Results from DNA sequence analyses of PR genes from these patients are also consistent with data from in vitro selection experiments and in vivo studies. As seen in patients treated with other PIs, numerous secondary substitutions can be detected in patients receiving nelfinavir, although D30N appears to be the predominant nelfinavir resistance-associated substitution [5, 6, 12, 13]. Likewise, the L90M substitution, which previously was rarely observed in nelfinavir-treated patients, was selected after nelfinavir monotherapy in only 1 of 32 patients at 16 weeks and was not detected at 48 weeks in any patient who received nelfinavir in combination with lamivudine and zidovudine. Data describing incidences of genotypic resistance in patients receiving ritonavir, saquinavir, or amprenavir as monotherapy for similar treatment periods have not been shown to exhibit this finding.

Figure 2. Percentage of patients with and without genotypic changes who demonstrated virologic response after treatment with nelfinavir in combination with lamivudine and zidovudine. Patients were categorized as having D30N and/or genotypic changes in reverse transcriptase (RT) genes (PR-RT⁻) or as lacking such genotypic changes (PR-RT⁺). Response is defined as achieving 2 consecutive time points with plasma human immunodeficiency virus (HIV) RNA <400 HIV RNA copies/mL or <50 HIV RNA copies/mL.
of time between the first of 2 consecutive time points of plasma HIV RNA ≤400 copies/mL and the first of 2 consecutive time points of plasma HIV RNA >400 copies/mL. Significance of difference between PR-RT− and PR-RT+ groups’ duration of response was determined by log rank test (P = .0001), PR, protease.

Results of this study clearly demonstrate a correlation between detectable genotypic changes and virologic response at 48 weeks among patients receiving nelfinavir as monotherapy, and they demonstrate a similar correlation at 48 weeks among patients receiving nelfinavir in combination with lamivudine and zidovudine. Results of this study further indicate that potent suppression of HIV replication (to <50 copies/mL) in patients receiving nelfinavir in combination with lamivudine and zidovudine is associated with a more durable response, leading to a reduction in emergence of resistant HIV strains. Our analysis also demonstrates that the drug-resistant HIV variants that too low to detect by current techniques, the relative fitness and resulting contribution to plasma HIV RNA on the part of such minor populations must be considered negligible in the face of durable plasma virus suppression.

Although the analysis of the relationship between nelfinavir-associated substitutions and virologic response in patients who received nelfinavir monotherapy in our study may be straightforward, the association of specific resistance-associated substitutions and loss of virus suppression in patients receiving nelfinavir in combination with lamivudine and zidovudine is more complex. Samples from all 6 patients whose viruses showed detectable genotypic changes during nelfinavir-combination therapy contained the lamivudine resistance-associated substitution M184V. Only a subset of these patients, however, had HIV isolates with genotypic changes associated with resistance to nelfinavir (3 of 6) and/or zidovudine (2 of 6). Furthermore, in the 4 patients for whom a temporal change in genotype could be determined, the M184V substitution occurred 4–40 weeks before D30N, M41M/L, or K219K/R substitutions. The appearance of M184V in these patients, either alone or before substitutions in PR, is consistent with recent data from patients who received other PIs (indinavir or amprenavir) in combination with lamivudine and zidovudine [31, 32].

Results of earlier studies [31, 32], together with the data presented here, strongly suggest that the drug-resistant variant that first emerges during such regimens is resistant to lamivudine rather than to the PI component of the drug combination. It is unclear why genotypic RTI resistance precedes PI resistance in these patients; however, the preexistence of lamivudine-resistant HIV variants in untreated patients is supported by the report describing pretreatment variants containing the precursor lamivudine resistance–associated substitution M184I [33]. Initiation of treatment with lamivudine could therefore serve to rapidly select for lamivudine-resistant viruses that contain M184V. In vitro studies comparing the relative fitness of some recombinant RTI- or PI-resistant viruses with that of wild-type viruses have also been reported [34–42]. Although data from these studies have shown that viruses with resistance-associated substitutions in PR or RT have a replicative disadvantage relative to wild-type virus, studies comparing the relative fitness levels of variants containing PI- and RTI-resistance–associated substitutions have not been reported.

Results of this study clearly demonstrate a correlation between detectable genotypic changes and virologic response at 16 weeks among patients receiving nelfinavir as monotherapy, and they demonstrate a similar correlation at 48 weeks among patients receiving nelfinavir in combination with lamivudine and zidovudine. Results of this study further indicate that potent suppression of HIV replication (to <50 copies/mL) in patients receiving nelfinavir in combination with lamivudine and zidovudine is associated with a more durable response, leading to a reduction in emergence of resistant HIV strains. Our analysis also demonstrates that the drug-resistant HIV variants that
first emerge in patients who lose the ability to suppress virus while receiving nelfinavir in combination with lamivudine and zidovudine are resistant to lamivudine rather than to nelfinavir or zidovudine. These findings may carry important implications regarding which components of specific regimens may be modified after failure of virologic suppression during the course of combination antiretroviral treatment. The findings also support the potential relevance of genotyping in the course of clinical management of HIV infection for selected patients.

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References