AIDS RESEARCH AND HUMAN RETROVIRUSES
Volume 15, Number 18, 1999, pp. 1631–1638
Mary Ann Liebert, Inc.

Incidence and Predictors of Virologic Failure of Antiretroviral Triple-Drug Therapy in a Community-Based Cohort

DANIEL PARIS, BRUNO LEDERGERBER, RAINER WEBER, JOSEF JOST, MARKUS FLEPP, MILOS OPRAVIL, CHRISTIAN RUEF, and STEFAN ZIMMERLI

ABSTRACT

Highly active antiretroviral therapy fails to reach its recommended goal of sustained suppression of viral replication in a substantial proportion of patients. We analyzed incidence and predictors of virologic failure of the first regimen of a triple-drug combination therapy, including a protease inhibitor and two nucleoside analog reverse transcriptase inhibitors (NRTIs), in 274 HIV-infected patients. Long-term virologic response to combination therapy including salvage regimens was assessed 2.5 years after treatment initiation. During an initial observation period of up to 1.8 years (median, 0.8 years) 152 patients (55%) experienced sustained suppression of HIV-1 RNA to <500 copies/ml. Failure to reduce viral load to <500 copies/ml within 6 months (initial failure) was observed in 51 patients (19%). Independent risk factors for initial failure included higher baseline viral load; addition of a protease inhibitor to an unchanged NRTI regimen; use of saquinavir hard-gel capsules; and longer duration of prior NRTI treatment. Within a median of 7 months viral load rebound above 500 copies/ml occurred in 71 of 223 patients (32%) whose viral load had initially decreased below this threshold. In proportional hazard analysis none of the potential risk factors was significantly associated with viral load rebound. However, in 40 patients (56%) with viral load rebound, incomplete adherence to therapy or treatment interruptions preceded the rebound. Virologic outcome after 2.5 years correlated with initial response to the first regimen: viral load was <500 copies/ml in 88, 55, and 21% of patients with sustained suppression, viral load rebound, and initial failure, respectively.

INTRODUCTION

IMPRESSIVE REDUCTIONS in morbidity and AIDS-associated mortality can be attributed to the use of highly active antiretroviral therapy.[1,2] However, protease inhibitor-containing drug combinations failed to lead to sustained suppression of plasma HIV-1 RNA concentrations to below the limit of quantification in about 50% of patients treated at experienced primary care centers[3–6] and in large clinical trials.[7] Treatment failure may have grim consequences for the individual patient and seriously threatens the initial success of combination therapy in the community, raising concerns about selection and transmission of multiresistant viruses.[8] And, last but not least, treatment failure carries a significant economic burden.

To increase substantially the success rate of antiretroviral combination therapy there is an urgent need to understand better the factors associated with treatment failure. We therefore decided to determine incidence and predictors of virologic failure of the first regimen of highly active antiretroviral therapy in the primary care setting and to assess durability of treatment effect.

MATERIALS AND METHODS

Patients

The focus of our study was 274 participants in the Swiss HIV Cohort Study (SHCS)[2,9]; these patients were treated at our unit, a university-based urban primary care HIV clinic. Included were patients who initiated antiretroviral triple-combination therapy, including a protease inhibitor and two NRTIs, between October 1995 and March 1997, and for whom complete laboratory data sets were available. The latter include (1) an HIV-1 RNA concentration above 500 copies/ml and a $CD4^+$ cell count performed within 6 weeks before the start of combina-

Division of Infectious Diseases and Hospital Epidemiology, Department of Medicine, University Hospital, 8091 Zurich, Switzerland.

tion therapy, (2) at least one HIV-1 RNA assay performed between 2 and 6 months after the start of therapy, and (3) at least one other HIV-1 RNA determination performed more than 6 months after the start of therapy. Patients were protease inhibitor naive at study entry. Baseline patient characteristics, starting date, duration and composition of antiretroviral treatment, as well as laboratory results were retrieved from the SHCS database and verified by detailed chart review. Information on adherence was also retrieved from the charts. In the absence of a precise definition, the assessment of the quality of adherence was at the discretion of the treating physician. The analysis of incidence and predictors of virologic failure of the first regimen and on clinical events is based on information recorded up to December 1997. Assessment of long-term virologic outcome was based on the latest available HIV-1 RNA measurement recorded between January 1998 and May 1999. The limitation to the first treatment regimen was dropped for the analysis of long-term outcome.

HIV-1 RNA measurements

Until January 1998 the Roche Amplicor HIV-1 Monitor test was used to determine levels of plasma HIV-1 RNA concentration (viral load) according to the manufacturer's instructions. The lower limit of quantification was 500 copies/ml. Plasma HIV-1 RNA concentration was measured approximately every 3 months. For the latest available HIV-1 RNA measurements, the ultrasensitive procedure of the Roche Amplicor HIV-1 Monitor test with a lower limit of detection of 50 copies/ml was used.

Antiretroviral drugs

The nucleoside analog reverse transcriptase inhibitors (NRTIs) available during the study period and the doses prescribed included zidovudine (250 mg twice daily), stavudine (40 mg twice daily; 30 mg twice daily in patients weighing less than 60 kg), lamiduvine (3TC, 150 mg twice daily), didanosine (200 mg twice daily), and zalcitabine (0.75 mg three times daily). More than 85% of patients used either zidovudine and 3TC or stavudine and 3TC. Availability and, consequently, use of individual protease inhibitors changed during the study period. While saquinavir hard-gel capsules (600 mg three times daily) were prescribed to more than 60% of the patients prior to March 1996, they were largely replaced by indinavir (800 mg three times daily) or ritonavir (600 mg twice daily) by the second half of 1996. At the end of the study period, 60% of patients were taking indinavir, and 40% were taking ritonavir. In antiretroviral therapy-naive patients both NRTIs and the protease inhibitor were started on the same day or within an interval of less than 14 days.

Statistical analysis

Data were analyzed with SAS (SAS Institute, Cary, NC) software, version 6.11. The Kruskal–Wallis test or Wilcoxon two-sample test was used to analyze differences between groups. The following end points were analyzed: (1) initial treatment failure (no HIV-1 RNA value below 500 copies/ml within the first 6 months after initiating combination therapy); (2) viral load rebound (at least one HIV-1 RNA measurement below 500 copies/ml within the first 6 months of starting combination therapy, followed by at least one HIV-1 RNA value at or above 500 copies/ml); and (3) sustained suppression (at least two HIV-1 RNA measurements below 500 copies/ml; at least one within the first 6 months and at least one after 6 months of initiating combination therapy). Potential predictors of initial treatment failure and viral load rebound were assessed by logistic regression analysis and Cox proportional hazard analysis, respectively. The following variables were entered into the models: age; sex; HIV exposure risk; disease status according to clinical stages A, B, and C of the 1993 definition of the Centers for Disease Control and Prevention; $CD4^+$ cell count; plasma HIV-1 RNA concentration; calendar period when combination therapy was initiated (in quarters); mode of initiation of combination therapy; protease inhibitor; duration of HIV infection; and duration of prior antiretroviral treatment. $CD4^+$ cell count (after adding the value of 1) and HIV-1 RNA concentration were $\log_{10}$ transformed.

RESULTS

Baseline characteristics

We analyzed data from 274 HIV-infected persons who started antiretroviral combination therapy that included a protease inhibitor and two NRTIs between October 1995 and March 1997. Baseline patient characteristics are summarized in Table 1. The only significant difference between study patients and patients not included in the analysis (mainly due to incomplete baseline data) was a higher rate of pretreatment with NRTIs in the study population (73 versus 48%; $p < 0.001$). For patients with plasma HIV-1 RNA concentrations persistently below the limit of quantification, the observation period was at least 6 months.

TABLE 1. BASELINE PATIENT CHARACTERISTICS[a]

Characteristic	*Value*
Median age, years (range)	37 (21–67)
Sex, *n* (%)	
Male	217 (79)
Female	57 (21)
HIV exposure risk, *n* (%)	
Same-sex sexual activity	143 (52)
Injection drug users	72 (26)
Heterosexual	48 (18)
Other	11 (4)
CDC disease stage, *n* (%)	
A	82 (30)
B	88 (32)
C	104 (38)
Median $CD4^+$ cell count, $\times 10^6$/liter (range)	123.5 (0–1234)
Median plasma HIV-1 RNA concentration, $\log_{10}$ copies/ml (range)	4.68 (2.77–6.76)
History of prior NRTI treatment, *n* (%)	200 (73)

[a] $n = 274$.

We observed two general patterns of virologic treatment failure: (1) treatment failed ever to reduce viral load below the limit of quantification within the first 6 months or (2) after an initial decline below 500 copies/ml, plasma HIV-1 RNA concentrations rebounded above this threshold after variable amounts of time. Both patterns were analyzed separately.

Initial treatment failure

For the study of incidence and predictors of virologic failure of the first regimen of combination therapy patients were closely monitored for a median of 0.85 year (range, 0.5–1.65 years). Within 6 months of its initiation, antiretroviral combination therapy failed to reduce plasma HIV-1 RNA concentration to less than 500 copies/ml in 51 of 274 patients (19%) (Table 2). The failure rate was higher in NRTI-experienced patients (46 of 200 [23%]) than in treatment-naive patients (5 of 74 [7%]) (Table 4) and increased with the duration of prior NRTI treatment. Univariate analysis revealed further that the failure rate increased with higher baseline plasma HIV-1 RNA concentrations and lower $CD4^+$ cell counts at baseline. Patients taking saquinavir had significantly higher failure rates than did patients taking indinavir or ritonavir. Failure rates were independent of age, and evenly distributed between both sexes and all HIV exposure categories. Clinical stage and calendar period when combination therapy was initiated had no significant effect on the incidence of treatment failure. In multivariate logistic regression analysis we found the following four variables to be independent predictors of failure of therapy to reduce plasma HIV-1 RNA below the limit of quantification: (1) adding a protease inhibitor to an unchanged NRTI regimen (odds ratio [OR] 5.06 [95% confidence interval, 1.51–17.0]), compared with treatment-naive patients); (2) use of saquinavir (OR 3.06 [95% confidence interval, 1.03–9.06], compared with indinavir); (3) duration of pretreatment with NRTI (OR 1.36 [95% confidence interval, 1.04–1.77], per year); and (4) high baseline plasma HIV-1 RNA concentration (OR 2.41 [95% confidence interval, 1.44–4.04], per $\log_{10}$ copies/ml) as the only treatment-independent variable (Table 2 and Fig. 1). Combination therapy failed to reduce viral load below the limit of quantification in 7 of 68 patients (10%) with less than 10^4 HIV-1 RNA copies/ml and 25 of 95 patients (26%) with more than 10^5 copies/ml. Multivariate analysis also revealed that neither a treatment strategy that introduced at least one new NRTI to an existing regimen when protease inhibitor treatment was started, nor baseline $CD4^+$ cell count, were independently associated with virologic treatment failure.

TABLE 2. INCIDENCE AND PREDICTORS OF FAILURE OF ANTIRETROVIRAL COMBINATION THERAPY TO REDUCE VIRAL LOAD TO <500 COPIES PER MILLILITER[a]

			Univariate analysis		*Multivariate analysis*	
Factor	*Initial success*	*Failure*	*Odds ratio (95% CI)*[b]	p *Value*	*Odds ratio (95% CI)*[b]	p *Value*
Number of patients, *n* (%)	223 (81)	51 (19)	—		—	—
Clinical stage, *n* (%)				0.10		0.64
A	72 (89)	9 (11)	1		1	
B	69 (78)	19 (22)	2.20 (0.93–5.20)		1.58 (0.60–4.13)	
C	82 (78)	23 (22)	2.24 (0.98–5.16)		1.40 (0.53–3.69)	
Median HIV-1 RNA, $\log_{10}$ copies/ml (range)	4.60 (2.77–6.76)	4.99 (2.98–6.57)	1.80 (1.21–2.68)	0.004	2.41 (1.44–4.04)	0.0008
Median $CD4^+$ cell count, cells/mm^3 (range)	133 (0–1234)	72 (0–760)	0.55 (0.33–0.90)	0.017	0.82 (0.42–1.60)	0.56
Calendar period, *n* (%)				0.58		0.58
<3rd quarter, 1996	62 (78)	18 (22)	1.44 (0.71–2.93)		0.68 (0.26–1.80)	
3rd quarter, 1996	99 (83)	20 (17)	1		1	
>3rd quarter, 1996	62 (83)	13 (17)	1.04 (0.48–2.23)		1.21 (0.48–3.05)	
Mode of treatment initiation, *n* (%)				0.003		0.012
ART-naive patients	69 (93)	5 (7)	1		1	
Switch[c]	53 (80)	13 (20)	3.39 (1.14–10.09)		2.36 (0.65–8.61)	
Add-on[d]	101 (75)	33 (25)	4.51 (1.68–12.12)		5.06 (1.51–17.0)	
Protease inhibitor, *n* (%)						
Indinavir	113 (84)	22 (16)	1	—	1	—
Ritonavir	82 (84)	16 (16)	1.00 (0.50–2.03)	1.0	1.20 (0.52–2.74)	0.67
Saquinavir	28 (68)	13 (32)	2.39 (1.07–5.31)	0.03	3.06 (1.03–9.06)	0.04
Median duration of prior NRTI treatment, years (range)	1.17 (0.08–7)	1.82 (0.17–6.5)	1.44 (1.18–1.74)	0.0003	1.36 (1.04–1.77)	0.03
Median observation period, years (range)	0.8 (0.5–1.6)	0.9 (0.5–1.7)	—	—	—	—

Abbreviation: ART, Antiretroviral therapy.

[a] $n = 274$.

[b] Logistic regression model adjusted for sex, age, HIV exposure risk, and duration of HIV infection.

[c] Switch, at least one nucleoside reverse transcriptase inhibitor with which the patient has never before been treated is introduced when protease inhibitor is started.

[d] Add-on, protease inhibitor is added to an unchanged nucleoside reverse transcriptase inhibitor regimen.

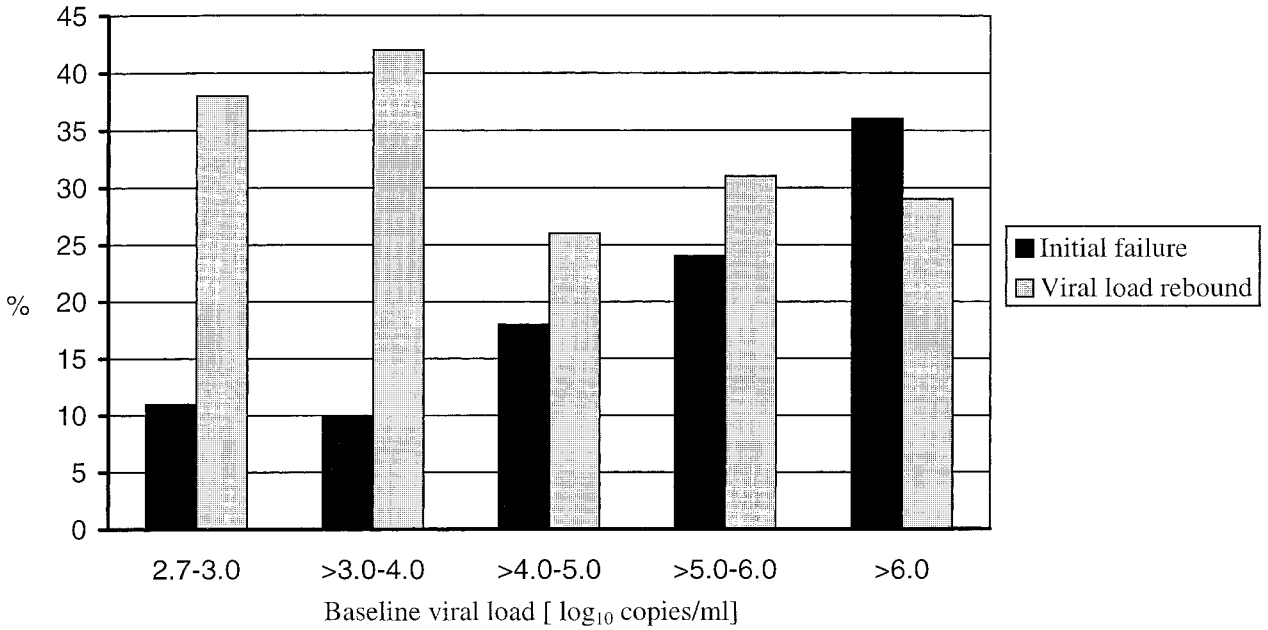

FIG. 1. The influence of viral load on virologic treatment failure. Proportion of patients in the respective baseline viral load stratum failing to suppress viral load below 500 copies/ml of plasma within 6 months of starting combination therapy (initial failure, solid bars), and those with viral load rebound above 500 copies/ml of plasma (gray bars).

Rebound of HIV-1 RNA

In 223 of 274 patients (81%) antiretroviral combination therapy initially succeeded in reducing plasma HIV-1 RNA concentration to undetectable levels. However, during an observation time of at least 6 months after the start of a protease inhibitor-containin g regimen, viral load rebounded to levels above 500 copies/ml of plasma in 71 of 223 patients (32%) (Table 3). Again, the virologic treatment failure rate was higher among NRTI-experienced patients (57 of 154 [37%]) than among treatment-naive patients (14 of 69 [20%]) (Table 4). Overall, combination therapy led to sustained reduction of plasma viral load to less than 500 copies/ml in 55 of 74 treatment-naive patients (74%), and in 97 of 200 pretreated patients (49%). In the univariate Cox proportional hazard model, failure of treatment to continuously suppress viral replication was significantly associated with (1) calendar period of treatment initiation, (2) adding a protease inhibitor to an unchanged NRTI regimen, and (3) CDC stage C disease. In the multivariate model, none of the tested variables was significantly associated with a rebound of HIV-1 RNA concentration above 500 copies/ml of plasma. However, detailed chart review revealed that the treating physician had noted incomplete adherence to therapy to precede viral load rebound in 25 of 71 patients (35%), and that in an additional 15 of 71 patients (21%) treatment interruptions of at least 1 week were documented before virologic failure occurred. In contrast, for none of the patients with sustained suppression of viral replication were difficulties with adherence to the prescribed regimen reported, and only 12 of these 152 patients (8%) had documented treatment interruptions of at least 1 week. A high baseline viral load was not associated with a rebound of HIV-1 RNA concentration above 500 copies/ml of plasma. In contrast, the odds for initial virologic treatment failure increased with baseline plasma HIV-1 RNA concentration (Fig. 1).

Overall, antiretroviral therapy failed to continuously reduce viral load below 500 copies/ml in 122 of 274 patients (45%) (Table 4).

Clinical events during observation period

During the initial 25 months of the study, comprising 235 observation-years among 274 patients, no deaths of patients taking combination therapy were recorded. However, one patient with progressing non-Hodgkin's lymphoma died 4 months after stopping treatment. Nine AIDS-defining events in 8 patients were observed 1 to 5 months after the start of a protease inhibitor-containing regimen ($CD4^+$ cell counts at the time the events occurred are given in parentheses): *Candida* esophagitis in two (23 and 57), herpes simplex virus-associated nonhealing ulcers in two (139 and 424), non-Hodgkin's lymphoma in two (16 and 40), and one case each of cytomegalovirus retinitis (50), cryptosporidiosis (9), and Kaposi's sarcoma (150). Eight of the 9 events occurred in patients with virologic treatment failure; none seems to have been triggered by combination therapy.

Long-term virologic outcome

To assess long-term virologic response to combination therapy including salvage regimens, the latest HIV-1 RNA values available in May 1999 (corresponding to a median follow-up of 2.5 years [range, 1.7–3.4 years] after the start of combination therapy) were compared with the initial treatment response to the first regimen (Table 5). Results of HIV-1 RNA determinations were available for 242 of 274 patients (88%). A total

TABLE 3. INCIDENCE AND PREDICTORS OF FAILURE OF VIRAL LOAD REBOUND AFTER INITIAL REDUCTION OF VIRAL LOAD TO <500 COPIES PER MILLILITER[a]

Factor	Sustained suppression	Rebound[b]	Univariate analysis: Risk ratio (95% CI)[c]	Univariate analysis: p Value	Multivariate analysis: Risk ratio (95% CI)[c]	Multivariate analysis: p Value
Number of patients, *n* (%)	152 (68)	71 (32)	—	—	—	—
Clinical stage, *n* (%)				0.10		0.25
A	57 (79)	15 (21)	1		1	
B	46 (67)	23 (33)	1.68 (0.87–3.23)		1.42 (0.72–2.84)	
C	49 (60)	33 (40)	1.88 (1.02–3.47)		1.77 (0.89–3.49)	
Median HIV-1 RNA, $\log_{10}$ copies/ml (range)	4.5 (2.8–6.8)	4.6 (2.9–6.6)	0.84 (0.63–1.12)	0.24	0.90 (0.62–1.30)	0.57
Median $CD4^+$ cell count, cells/mm^3 (range)	140 (0–1230)	110 (0–930)	1.07 (0.73–1.55)	0.73	1.30 (0.82–2.07)	0.27
Calendar period, *n* (%)				0.017		0.11
<3rd quarter, 1996	27 (44)	35 (56)	1.68 (1.0–2.83)		1.67 (0.89–3.14)	
3rd quarter, 1996	73 (74)	26 (26)	1		1	
>3rd quarter, 1996	52 (84)	10 (16)	0.66 (0.32–1.36)		0.70 (0.32–1.52)	
Mode of treatment initiation, *n* (%)				0.081		0.62
ART-naive patients	55 (80)	14 (20)	1		1	
Switch[d]	36 (68)	17 (32)	1.83 (0.90–3.72)		1.50 (0.63–3.58)	
Add-on[e]	61 (60)	40 (40)	1.91 (1.04–3.53)		1.21 (0.52–2.85)	
Protease inhibitor, *n* (%)						
Indinavir	87 (77)	26 (23)	1	—	1	—
Ritonavir	55 (67)	27 (33)	1.48 (0.86–2.54)	0.15	1.30 (0.73–2.30)	0.38
Saquinavir	10 (36)	18 (64)	1.66 (0.87–3.16)	0.12	0.89 (0.41–1.93)	0.77
Median duration of prior NRTI treatment, years (range)	1.0 (0.08–7)	1.42 (0.08–4.8)	1.11 (0.96–1.30)	0.16	1.06 (0.85–1.32)	0.61
Median observation period, years (range)	0.8 (0.5–1.8)	0.56 (0.2–1.4)	—	—	—	—

[a] $n = 223$.

[b] Intent to treat analysis. Fifty of 71 (70%) patients were still receiving their initial drug regimen at time of rebound.

[c] Cox proportional hazard model for time between initiation of combination therapy and viral load rebound adjusted for sex, age, HIV exposure risk, and duration of HIV infection.

[d] Switch, at least one nucleoside reverse transcriptase inhibitor with which the patient has never before been treated is introduced when protease inhibitor is started.

[e] Add-on, protease inhibitor is added to an unchanged nucleoside reverse transcriptase inhibitor regimen.

of 122 of 139 patients (88%) who initially responded with sustained suppression of viral replication had viral load values below 500 copies/ml; most, 107 of 139 (77%), had viral load determinations below 50 copies/ml. Seventy-five (54%) of them were still on their original treatment regimen. In contrast, only 33 of 60 patients (55%) with initial viral load rebound and 9 of 43 patients (21%) with initial treatment failure achieved a reduction of HIV-1 RNA concentration below 500 copies/ml on a salvage regimen. Overall, 2.5 years after starting combination therapy, 164 of 242 (68%) and 139 of 242 patients (57%) had a viral load below 500 and below 50 copies/ml, respectively.

DISCUSSION

During the initial observation period of this study, the first regimen of a protease inhibitor-containing regimen failed to lead to sustained suppression of viral replication in 45% of patients. We identified two distinct patterns of virologic treatment failure that also differed with respect to the associated risk factors. During the first 6 months of treatment viral load failed to decline below 500 copies/ml in 19% of patients. A high baseline viral load, addition of a protease inhibitor without changing an established NRTI regimen, use of saquinavir hard-gel capsules, and long prior NRTI treatment were independent predictors of this type of treatment failure. In total of 32% of 223 patients who initially achieved an unquantifiable HIV-1 RNA level, viral load later rebounded above 500 copies/ml. None of the potential risk factors studied was significantly associated with viral load rebound in proportional hazard analysis. Chart review revealed that incomplete adherence to therapy preceded viral load rebound in 35% of patients (in contrast, none of the patients with sustained suppression was noted for adherence problems). Treatment interruptions of at least 1 week preceded viral load increase above the limit of quantification in an additional 21% of patients.

At a median of 2.5 years after initiation of a protease inhibitor-containing regimen, highly active antiretroviral therapy including first and salvage regimens had led to suppression of viral replication to levels of 500 and 50 copies/ml in 68 and 57% of patients, respectively.

TABLE 4. INFLUENCE OF NRTI PRETREATMENT ON VIROLOGIC TREATMENT FAILURE

Type of virologic failure	*All patients*	*NRTI-experienced patients*	*NRTI-naive patients*
Failure to reduce viral load below 500 copies/ml, *n* (%)	51/274 (19)	46/200 (23)[a]	5/74 (7)
Viral load rebound above 500 copies/ml, *n* (%)	71/223 (32)	57/154 (37)[b]	14/69 (20)
Overall virologic treatment failure, *n* (%)	122/274 (45)	103/200 (52)[c]	19/74 (26)

[a] χ^2, $p = 0.02$ (naive versus experienced).
[b] χ^2, $p = 0.01$ (naive versus experienced).
[c] χ^2, $p = 0.0002$ (naive versus experienced).

Many of the risk factors associated with failure to achieve unquantifiable HIV-1 RNA levels can be linked to the emergence of resistance, although this was not directly measured. Pretreatment with NRTI did not lead to complete suppression of viral replication in the study population (those with unquantifiable viral load at baseline were excluded from analysis). Ongoing viral replication under treatment has been shown to be strongly associated with resistance development.[10,11] Consequently, failure to change NRTI when the protease inhibitor was introduced may be equivalent to instituting protease inhibitor monotherapy, which has been shown to lead to the development of resistant viruses.[10] Considering the potential of cross-resistance between NRTIs,[12] it may not be surprising that long duration of prior NRTI prescription is a risk factor not only for patients whose NRTI regimen remained unchanged when a protease inhibitor was added but also for patients whose new regimen consisted of at least one new NRTI in addition to the protease inhibitor. The low bioavailability of saquinavir hard-gel capsules has been used to explain its limited potency to suppress viral replication, which may lead directly to the emergence of resistant viruses.[13]

Although studies on the virologic effect of protease-containing regimens in clinical cohorts[3,4,6] did not differentiate between initial failure and viral load rebound, the risk factors for overall treatment failure they identified largely correspond with those we found associated with failure to achieve unquantifiable HIV-1 RNA concentrations. A notable exception is the absence in this study, and that of Casado *et al.*,[6] of an association between baseline $CD4^+$ cell count and failure rate, a finding that contrasts with previous work.[3,4] Differences in the definition of virologic failure and baseline patient characteristics may account for this discrepancy.

In retrospect, it may be surprising that the experience gained with the sequential use of tuberculostatic drugs when they were introduced more than 40 years ago ("never add a single drug to a failing regimen") was not initially applied to antiretroviral therapy. When protease inhibitors first became available, they were often added to previously established NRTI regimens.[7,14] Only when the high failure rates associated with this approach became evident did concomitant rather than sequential introduction of antiretroviral drugs become standard. However, in spite of the strong association between treatment failure and sequential introduction of antiretroviral drugs, our adoption of the new standard, during the course of the study, is not reflected by a significantly better outcome in patients who started combination therapy in the later phases of the study (Table 2).

The first protease inhibitor-containing regimen failed to continuously suppress viral replication below the limit of quantification in 45% of our patients. Similar failure rates ranging from 40 to 55% have been reported for other clinical cohorts[3,4,6] and for clinical trials.[7] Patients failing their first protease inhibitor-containing regimen are commonly switched to new treatment regimens containing drugs the patient has not previously taken. The success rate of these salvage regimens may be low.[4] Our observations at follow-up 2.5 years after initiating combination therapy suggest that salvage regimens may lead to sustained suppression of viral replication in up to 55% of patients with viral load rebound. Salvage regimens, however, were significantly less successful in patients who experienced initial fail-

TABLE 5. VIROLOGIC STATUS AT LAST FOLLOW-UP[a]

	Sustained suppression	*Rebound*	*Failure*
n[b]	152	71	51
Died[c]	5 (3.3%)	4 (5.6%)	4 (7.8%)
Lost to follow-up[c]	8 (5.3%)	7 (9.9%)	4 (7.8%)
Long-term virologic follow-up available	139 (91.4%)	60 (84.5%)	43 (84.3%)
Median follow-up, months (range)	29 (20–40)	31 (24–39)	30 (22–41)
HIV-1 RNA at latest follow-up, copies/ml			
>10,000	7 (5.0%)	10 (16.7%)	18 (41.8%)
>1000–10,000	7 (5.0%)	14 (23.3%)	15 (34.9%)
>500–1000	3 (2.2%)	3 (5.0%)	1 (2.3%)
>50–500	15 (10.8%)	7 (11.7%)	3 (7.0%)
≤50	107 (77.0%)	26 (43.3%)	6 (14.0%)

[a] $n = 242$.
[b] From analyses of short-term effectiveness.
[c] Between January 1998 and May 1999.

ure (only 21% of these patients had a viral load below 500 copies/ml at the latest follow-up).

The durability of effect of a successful first regimen is emphasized by our finding that 88 and 77% of patients with initial success had a viral load of <500 and <50 copies/ml, respectively, 2.5 years after starting combination therapy.

No risk factors for rebounding HIV-1 RNA concentrations other than treatment interruptions of at least 1 week and incomplete adherence to therapy could be identified. Since adherence has been assessed retrospectively on the basis of unstructured and potentially biased notes in the patient charts, this finding needs to be interpreted with caution. However, suboptimal drug levels associated with nonadherence may promote resistance development and consequently therapeutic failure.[10] Treatment interruption can be considered an extreme of nonadherence. Our results suggest that once HIV-1 RNA concentration has become unquantifiable, neither baseline characteristics (such as high baseline viral load) nor inadequate treatment strategies (such as sequential introduction of antiretroviral drugs) that may make reaching this goal difficult determine the continued suppression of viral replication. This finding was confirmed in a study that found only a modest correlation between baseline HIV-1 RNA concentration and duration of viral suppression during protease inhibitor treatment.[5]

To achieve long-term control of HIV-infection, antiretroviral therapy must be aimed at sustained, complete suppression of viral replication. Recent work suggests this may mean to reduce plasma HIV-1 RNA levels to below 20 copies/ml.[11] If this goal is missed, resistance to antiretroviral drugs will eventually develop and virologic failure will ensue.[15] Virologic treatment failure predicted clinical failure in some studies.[16] However, the small number of AIDS-defining events observed in this and other studies[6] in spite of high virologic failure rates, and a report of a lack of clinical progression and increasing $CD4^+$ cells in a subset of patients continuing antiretroviral combination therapy in spite of increasing HIV-1 RNA plasma concentrations,[17] indicate that virologic treatment failure may not necessarily lead to clinical progression.

This study has several methodological limitations. Owing to a selection bias toward pretreated patients included in the study we may have overestimated the overall failure rate of combination therapy. The study still includes 27% treatment-naive patients and therefore allows for valid conclusions on the effect of pretreatment and its duration in the multivariate analyses (Tables 2 and 3). The low number of HIV-1 RNA measurements required for entry into the study and their timing may have led to underestimation of the initial response rate in patients whose HIV-1 RNA declines slowly. However, because 87 and 85% of patients had two or more HIV-1 RNA determinations in the first 6 months and between 6 and 9 months, respectively, after starting combination therapy this potential error is thought to be minor. The choice of protease inhibitor used was not randomized but rather governed by availability. Comparison of the relative contribution of individual protease inhibitors to virologic failure should thus be interpreted with caution.

Our study indicates ways to improve the success rate of antiretroviral combination therapy. Patients with high base line HIV-1 RNA concentrations should receive the most intense treatment tolerated, and may be considered for four-drug regimens (in spite of the limited data indicating their efficacy[18]). When a protease inhibitor is initiated in NRTI-experienced patients, at least one or possibly two NTRIs that the patient has not been taking previously should be included in the new regimen. Saquinavir hard-gel capsules should be combined with another protease inhibitor to increase serum levels, or be replaced by the soft-gel formulation.[19] The finding that antiretroviral-experienced patients had twice the failure rate of NRTI-naive patients emphasizes the importance of a successful first treatment regimen. Given the cross-resistance between antiretroviral drugs, treatment options may be limited in patients who develop resistance to one treatment regimen.

Last but not least, every effort should be taken to maximize adherence to therapy. In our experience, frequent individual discussions and support are invaluable but may need to be complemented by novel strategies to reach their goal.

In summary, we found the first regimen of antiretroviral combination therapy to suppress viral load below the limit of quantification in 55% of patients in a clinical cohort of HIV-infected individuals for at least 6 months. Virologic treatment failure was evident either as failure to achieve HIV-1 RNA levels below 500 copies/ml or as viral load rebound. Both patterns of failure were associated with distinct risk factors. Improved treatment strategies for clinical practice and higher success rates of antiretroviral combination treatment may result when these risk factors are taken into consideration.

REFERENCES

1. Palella FJ Jr, Delaney KM, Moorman AC, Loveless MO, Fuhrer J, Satten GA, Aschman DJ, and Holmberg SD: Declining morbidity and mortality among patients with advanced human immunodeficiency virus infection. HIV Outpatient Study Investigators. N Engl J Med 1998;338:853–860.
2. Egger M, Hirschel B, Francioli P, Sudre P, Wirz M, Flepp M, Rickenbach M, Malinverni R, Vernazza P, and Battegay M: Impact of new antiretroviral combination therapies in HIV infected patients in Switzerland: Prospective multicentre study. Swiss HIV Cohort Study. Br Med J 1997;315:1194–1199.
3. Fatkenheuer G, Theisen A, Rockstroh J, Grabow T, Wicke C, Becker K, Wieland U, Pfister H, Reiser M, Hegener P, Franzen C, Schwenk A, and Salzberger B: Virological treatment failure of protease inhibitor therapy in an unselected cohort of HIV-infected patients. AIDS 1997;11:F113–F116.
4. Deeks SG, Hecht FM, Swanson M, Elbeik T, Loftus R, Cohen PT, and Grant RM: HIV RNA and CD4 cell count response to protease inhibitor therapy in an urban AIDS clinic: Response to both initial and salvage therapy. AIDS 1999;13:F35–F43.
5. Kempf DJ, Rode RA, Xu Y, Sun E, Heath-Chiozzi ME, Valdes J, Japour AJ, Danner S, Boucher C, Molla A, and Leonard JM: The duration of viral suppression during protease inhibitor therapy for HIV-1 infection is predicted by plasma HIV-1 RNA at the nadir. AIDS 1998;12:F9–F14.
6. Casado JL, Perez-Elías MJ, Antela A, Sabido R, Martí-Belda P, Dronda F, Blazquez J, and Quereda Q: Predictors of long-term response to protease inhibitor therapy in a cohort of HIV-infected patients. AIDS 1998;12:F131–F135.
7. Hammer SM, Squires KE, Hughes MD, Grimes JM, Demeter LM, Currier JS, Eron JJ Jr, Feinberg JE, Balfour HH Jr, Deyton LR, Chodakewitz JA, and Fischl MA: A controlled trial of two nucleoside analogues plus indinavir in persons with human immunodeficiency virus infection and CD4 cell counts of 200 per cubic mil-

limeter or less. AIDS Clinical Trials Group 320 Study Team. N Engl J Med 1997;337:725–733.
8. Hecht FM, Grant RM, Petropoulos CJ, Dillon B, Chesney MA, Tian H, Hellmann NS, Bandrapalli NI, Digilio L, Branson B, and Kahn JO: Sexual transmission of an HIV-1 variant resistant to multiple reverse-transcriptase and protease inhibitors. N Engl J Med 1998;339:307–311.
9. Ledergerber B, von Overbeck J, Egger M, and Luthy R: The Swiss HIV Cohort Study: Rationale, organization and selected base-line characteristics. Soz Praventivmed 1994;39:387–394.
10. Condra JH, Schleif WA, Blahy OM, Gabryelski LJ, Graham DJ, Quintero JC, Rhodes A, Robbins HL, Roth E, Shivaprakash M, *et al.*: In vivo emergence of HIV-1 variants resistant to multiple protease inhibitors. Nature (London) 1995;374:569–571.
11. Gunthard HF, Wong JK, Ignacio CC, Guatelli JC, Riggs NL, Havlir DV, and Richman DD: Human immunodeficiency virus replication and genotypic resistance in blood and lymph nodes after a year of potent antiretroviral therapy. J Virol 1998;72:2422–2428.
12. Gao Q, Gu Z, Parniak MA, Cameron J, Cammack N, Boucher C, and Wainberg MA: The same mutation that encodes low-level human immunodeficiency virus type 1 resistance to 2′,3′-dideoxyinosine and 2′,3′-dideoxycytidine confers high-level resistance to the (−) enantiomer of 2′,3′-dideoxy-3′-thiacytidine. Antimicrob Agents Chemother 1993;37:1390–1392.
13. Noble S and Faulds D: Saquinavir. A review of its pharmacology and clinical potential in the management of HIV infection. Drugs 1996;52:93–112.
14. Cameron DW, Heath-Chiozzi M, Danner S, Cohen C, Kravcik S, Maurath C, Sun E, Henry D, Rode R, Potthoff A, and Leonard J: Randomised placebo-controlled trial of ritonavir in advanced HIV-1 disease. The Advanced HIV Disease Ritonavir Study Group. Lancet 1998;351:543–549.
15. Carpenter CC, Fischl MA, Hammer SM, Hirsch MS, Jacobsen DM, Katzenstein DA, Montaner JS, Richman DD, Saag MS, Schooley RT, Thompson MA, Vella S, Yeni PG, and Volberding PA: Antiretroviral therapy for HIV infection in 1998: Updated recommendations of the International AIDS Society—USA panel. JAMA 1998;280:78–86.
16. O'Brien WA, Hartigan PM, Daar ES, Simberkoff MS, and Hamilton JD: Changes in plasma HIV RNA levels and CD4+ lymphocyte counts predict both response to antiretroviral therapy and therapeutic failure. VA Cooperative Study Group on AIDS. Ann Intern Med 1997;126:939–945.
17. Kaufmann D, Pantaleo G, Sudre P, and Telenti A: CD4-cell count in HIV-1-infected individuals remaining viraemic with highly active antiretroviral therapy (HAART). Swiss HIV Cohort Study. Lancet 1998;351:723–724.
18. Mellors J, Japour AJ, Cameron DW, Farthing C, Cohen C, Markowitz M, Poretz D, Follansbee S, Ho D, McMahon D, Berg J, Nieves J, Xu Y, Rode R, Salgo M, Leonard J, and Sun E: Ritonavir (RTV)–saquinavir (SQV) in protease inhibitor-naive patients after 72 weeks. In: *12th World AIDS Conference. Geneva, Switzerland.* Geneva, 1998. [Abstract 12295]
19. Perry CM and Noble S: Saquinavir soft-gel capsule formulation. A review of its use in patients with HIV infection. Drugs 1998; 55:461–468.

Address reprint requests to:
Stefan Zimmerli
Institute of Medical Microbiology
Infectious Diseases
University of Bern
Friedbuehlstrasse 51
CH-3010 Bern, Switzerland

E-mail: szimmerli@imm.unibe.ch