Intermittent antiretroviral therapy in patients with controlled HIV infection

Bruno Marchou[a], Philippe Tangre[b], Isabelle Charreau[b], Jacques Izopet[a], Pierre-Marie Girard[c], Thierry May[e], Jean-Marie Ragnaud[f], Jean-Pierre Aboulker[b] and Jean-Michel Molina[d] for the ANRS 106 Study team

Background: To assess the safety of a drug-sparing treatment regimen in patients with high CD4 cell counts and controlled HIV replication under antiretroviral therapy.

Methods: An open-label, non-inferiority study involving 403 adults with CD4 cell counts of 450×10^6 cells/l or greater and plasma HIV-1-RNA levels less than 200 copies/ml, randomly assigned to switch to an 8-week off, 8-week on regimen or to continue their antiretroviral regimen. The primary endpoint was the proportion of patients reaching a confirmed CD4 cell count less than 300×10^6 cells/l.

Results: Over 96 weeks, the proportion of patients meeting this endpoint was non-inferior in the intermittent group (3.6 versus 1.5%, upper bound of the 95% confidence interval of the difference 5.6%). No AIDS-defining event and two non-HIV-related deaths (intermittent arm) were recorded. The median decrease from baseline in the CD4 cell count was greater in the intermittent arm (-155 versus -8×10^6 cells/l, $P < 0.0001$). Minor HIV-related events, mainly lymphadenopathy and mucosal candidiasis, were more frequent in the intermittent group (14 versus 7%, $P = 0.04$) as were thrombocytopenia. The incidence of grade 3–4 non-HIV-related events and laboratory abnormalities were not statistically different between the groups. At week 96, the proportion of patients with plasma HIV-1-RNA levels less than 400 copies/ml were 81 and 90% in the intermittent (8 weeks after treatment resumption) and continuous groups ($P = 0.02$), respectively, with similar patterns of HIV resistance genotypes.

Conclusion: Despite some limitations, an 8-week off and on intermittent treatment regimen appeared clinically safe over 96 weeks while sparing half of the drug exposure.
© 2007 Lippincott Williams & Wilkins

AIDS 2007, **21**:457–466

Keywords: antiretroviral therapy, HIV, intermittent therapy, structured treatment interruption

Introduction

The use of combination antiretroviral therapy has dramatically improved the prognosis of HIV-1 infection, but continuous lifelong therapy is required to maintain virological suppression [1–4]. In patients with full suppression of HIV-1 replication and high CD4 cell counts, the benefits conferred by antiretroviral therapy

From the [a]Hôpital Purpan, Toulouse, France, the [b]INSERM SC 10, Villejuif, France, the [c]Assistance-Publique Hôpitaux de Paris, Hôpital Saint-Antoine, and [d]Hôpital Saint-Louis, Paris, France, the [e]Hôpital Brabois, Nancy, France, and the [f]Hôpital Pellegrin, Bordeaux, France.

Correspondence and requests for reprints to Bruno Marchou, Department of Infectious and Tropical Diseases, Hôpital Purpan, Toulouse 31059, France.

E-mail: marchou.b@chu-toulouse.fr

The investigators participating in the Window study are listed in the Appendix.

Received: 23 June 2006; revised: 10 October 2006; accepted: 31 October 2006.

ISSN 0269-9370 © 2007 Lippincott Williams & Wilkins

Copyright © Lippincott Williams & Wilkins. Unauthorized reproduction of this article is prohibited.

can be tempered by the required high rate of adherence to therapy, and by the occurrence of drug-related lipodystrophy and metabolic abnormalities [5,6]. Initially, scheduled treatment interruption (STI) trials aimed at stimulating HIV-specific immune responses to allow a spontaneous control of HIV replication. Failure to reach this goal led to the hypothesis that intermittent therapy could be a drug-sparing strategy, potentially reducing drug-related adverse events and costs while maintaining CD4 cell counts at a level high enough to prevent the risk of disease progression [7–10].

In this study, we assessed the safety of a 50% drug-sparing strategy in patients with high CD4 cell counts and viral suppression under antiretroviral therapy, using a fixed intermittent 8-week off, 8-week on regimen, in the setting of a randomized 96-week trial.

Methods

Patients

Eligible patients were HIV-1-infected adults who were tolerating combination antiretroviral therapy well, who had a nadir pretreatment CD4 cell count of 100×10^6 cells/l or more, who had a CD4 cell count above 450×10^6 cells/l at screening, and whose plasma HIV-1-RNA levels had been less than 200 copies/ml for at least the previous 6 months. Exclusion criteria were pregnancy, current treatment with abacavir or nevirapine, chronic hepatitis B infection, a history of splenectomy, and a history of an AIDS-defining event in the previous 18 months. A history of virological failure under previous antiretroviral therapy was not an exclusion criterion.

Study design

The study was a multicentre, randomized, open-label, non-inferiority trial carried out at 39 centres in France. Randomization was performed after screening by a centralized procedure and was stratified on the use of efavirenz, the CD4 cell count at entry (above or below 600 cells), and the nadir CD4 cell count (above or below 200 cells).

Patients were randomly assigned 1 : 1 to switch to a fixed intermittent treatment strategy starting with 8 weeks off therapy and followed by 8 weeks on therapy, or to maintain their current treatment strategy. In the intermittent group, patients underwent six cycles off and on therapy, and those on efavirenz were asked to stop efavirenz 7 days before the other drugs to limit the risk of resistance because of its longer half-life.

Patients were allowed to change drugs during the study because of adverse reactions, or patient or investigator decision, providing that they remained under the assigned treatment strategy.

The protocol was approved by the ethics committee of Toulouse University Purpan Hospital and by the Agence Nationale de Recherches sur le SIDA et les Hépatites Virales (ANRS). All patients gave written informed consent. The study is registered at clinicaltrials.gov, no. NCT 00122551.

Study monitoring

Patients were assessed at baseline, then every 8 weeks until they completed the 96 weeks of follow-up. At each visit, clinical data were collected through an interim medical history and physical examination, and blood specimens were obtained. Routine analyses were performed at each site throughout the follow-up period and included a complete blood count, CD4 cell count, measurement of plasma HIV-1 RNA, and tests of liver, kidney, muscle and pancreatic function. Furthermore, at baseline and weeks 48 and 96, fasting glucose, triglycerides, total cholesterol, and high and low-density lipoprotein cholesterol levels were measured. Safety was assessed through the reporting of adverse events and laboratory abnormalities, the severity of which was assessed with use of the ANRS toxicity grading scale (http://www.anrs.fr/index.php/article/articleview/1358/1/44).

When a CD4 cell count of less than 300×10^6 cells/l was recorded at a visit, the patient had to be retested 2 weeks later for confirmation of immunological failure. In such a case, a switch to a continuous treatment was recommended. Other reasons for treatment strategy discontinuation included disease progression, a significant adverse event, laboratory abnormality, virological failure and patient or physician decision.

At baseline and at weeks 48 and 96, patients were also assessed for morphological changes related to body-fat abnormalities by their physician using a directed questionnaire as described [11]. No objective measurement of body composition was performed.

Virological substudy

A virological substudy was planned among the first 200 patients enrolled in the study to analyse the virological consequences of treatment interruptions. Frozen samples were centralized for genotypic resistance analyses performed according to the consensus method of the ANRS [12,13]. Genotypic resistance testing was performed in peripheral blood mononuclear cell DNA from all patients at baseline and week 96, and in the plasma of all patients whenever the HIV-RNA level was above 1000 copies/ml after at least 6 weeks of therapy. HIV-1 mutations associated with drug resistance were defined according to the International AIDS Society – USA panel drug resistance mutations group definition [14].

Copyright © Lippincott Williams & Wilkins. Unauthorized reproduction of this article is prohibited.

Study endpoints

The primary study endpoint was the cumulative proportion of patients throughout the study reaching a confirmed CD4 cell count of less than 300×10^6 cells/l, defined as immunological failure. Secondary endpoints included the proportion of patients with a CD4 cell count above 450×10^6 cells/l at week 96, the proportion of patients who experienced any new Centers for Disease Control and Prevention category C event [15], and the proportion of patients with a plasma HIV-RNA level of less than 400 copies/ml at week 96. Safety endpoints included the proportion of patients remaining under their assigned treatment strategy, the proportion of patients with grade 3–4 adverse events and laboratory abnormalities, and the occurrence of metabolic and body-fat abnormalities. An independent Data Safety Monitoring Board was responsible for the safety and integrity of the patients throughout the study.

Statistical analysis

The primary endpoint was analysed on an intention-to-treat basis, including observed data of all randomized patients who started the assigned treatment strategy, regardless of whether this strategy was prematurely discontinued.

A second 'on-treatment strategy' analysis was also performed, in which only observed data from patients who remained under their assigned treatment strategy were considered for analysis. Data from patients who withdrew consent, were lost to follow-up, or discontinued treatment strategy were censored. Discontinuation of the treatment strategy was defined in both groups as the discontinuation of all antiretroviral agents for 6 weeks or more during a period on treatment, or in the intermittent group as the use of drugs for 6 weeks or more during a period off treatment.

The sample size was calculated on the basis of the primary immunological endpoint, to establish non-inferiority of the intermittent group compared with the continuous group. The limit for non-inferiority was set at 7% for the upper limit of the 95% confidence interval for the difference of proportions, a difference lower than 7% being considered as clinically acceptable. Based upon an expected immunological failure rate of 5% in the continuous group, a total of 200 patients per group was determined, which would give a statistical power of 90% to demonstrate non-inferiority with a one-sided alpha level of 0.05 (Newcombe–Wilson score method for confidence interval limits). The log-rank test was used to compare the time to treatment strategy discontinuation between groups. Chi-square or Fisher's exact tests were used to compare qualitative variables, and the Wilcoxon's rank-sum test was used to compare continuous variables. Comparisons were made with use of a two-sided alpha level of 0.05. Statistical analysis was performed with the use of SAS software version 8.2 (SAS Institute Inc., Cary, North Carolina, USA).

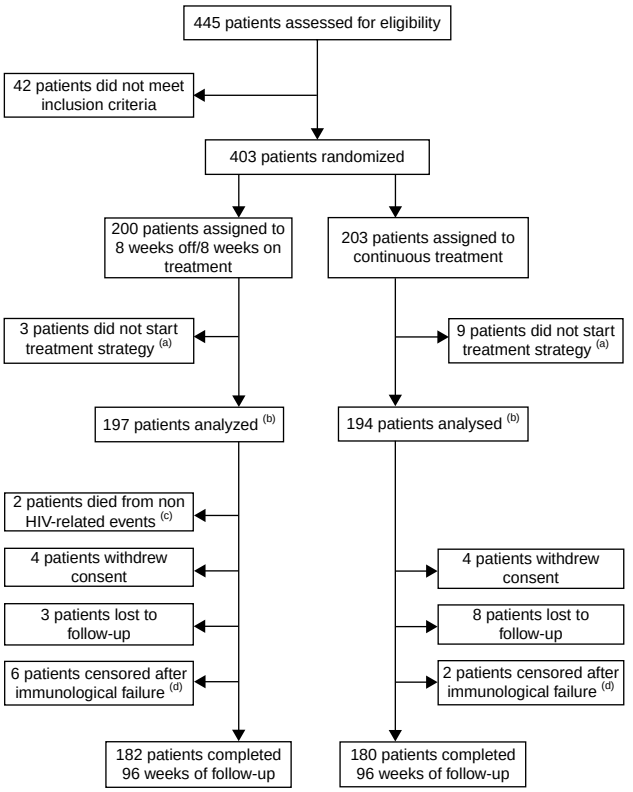

Fig. 1. Profile of patient enrolment and disposition over 96 weeks of follow-up. [a]Patients were disappointed by their randomization group, did not start the assigned treatment strategy and were not included in the analysis. [b]Twenty-seven patients in the intermittent group and 16 patients in the continuous group discontinued their assigned treatment strategy during follow-up ($P = 0.09$ by the log-rank test). [c]Two patients died, one was a sudden death 20 months after randomization and the other died of hepatic failure 11 months after discontinuation of antiretroviral therapy. These two deaths were considered not to be related to HIV disease. [d]Two other patients experienced immunological failure; one in the intermittent arm (who died of hepatic failure 11 months after discontinuation of antiretroviral therapy), one in the continuous arm at week 96.

Results

Population

Between December 2001 and June 2003, 403 patients were randomly assigned to the study (Fig. 1). Three patients in the intermittent group and nine patients in the continuous group withdrew consent at baseline and were not included in the analysis. The baseline characteristics of the remaining 391 patients were well balanced among the groups (Table 1). Ninety-two per cent of the patients in the intermittent group and 93% in the continuous group completed 96 weeks, for a total duration of follow-up of 705 patient-years (354 and 351 in the intermittent and continuous groups, respectively). Forty-three patients (11%) discontinued their treatment strategy during the study, 27 (14%) in the intermittent group and

Copyright © Lippincott Williams & Wilkins. Unauthorized reproduction of this article is prohibited.

Table 1. Baseline characteristics of the patients.

Characteristic	Intermittent group ($N=197$)	Continuous group ($N=194$)
Age (years)		
Median	42	41
Interquartile range	36–48	36–48
Male sex, no. (%)	159 (81)	153 (79)
Route of HIV infection, no. (%)		
Male homosexuals	104 (53)	96 (50)
Heterosexuals	62 (31)	63 (32)
Injection drug users	12 (6)	11 (6)
Others or unknown	19 (10)	24 (12)
CDC stage, no. (%)		
A	128 (65)	134 (69)
B	54 (27)	44 (23)
C	15 (8)	16 (8)
Pretreatment nadir CD4 cell count ($\times 10^6$ cells/l)		
Median	274	288
Interquartile range	203–371	208–370
CD4 cell count ($\times 10^6$ cells/l)		
Median	739	748
Interquartile range	600–887	614–926
Antiretroviral therapy[a]		
Nucleoside reverse-transcriptase inhibitors, no. (%)	196 (99)	194 (100)
Lamivudine	160 (81)	157 (81)
Emtricitabine	3 (2)	5 (3)
Zidovudine	85 (43)	90 (46)
Stavudine	85 (43)	83 (43)
Didanosine	65 (33)	62 (32)
Tenofovir	4 (2)	9 (5)
Zalcitabine	1 (1)	1 (1)
Protease inhibitors, no. (%)	94 (48)	85 (44)
Nelfinavir	35 (18)	31 (16)
Indinavir	25 (13)	34 (18)
Saquinavir	17 (9)	7 (4)
Ritonavir alone	5 (3)	3 (2)
Lopinavir/ritonavir	14 (7)	10 (5)
Other combinations with low-dose ritonavir	27 (14)	27 (14)
Non-nucleoside reverse-transcriptase inhibitors, no. (%)	86 (44)	83 (43)
Efavirenz	86 (44)	82 (42)
Nevirapine	0 (0)	1 (1)
Duration of antiretroviral therapy (years)		
Median	5.2	5.1
Interquartile range	4.1–6.6	3.9–6.5
First-line antiretroviral therapy, no. (%)	35 (18)	37 (19)
Second-line antiretroviral therapy, no. (%)	50 (25)	39 (20)
Third-line or more antiretroviral therapy, no. (%)	112 (57)	118 (61)
Previous history of treatment failure, no. (%)	77 (39)	74 (38)
Lipodystrophy[b] (all grades)	126 (65)	124 (66)
Lipoaccumulation	19 (10)	27 (14)
Lipoatrophy	44 (23)	49 (26)
Mixed syndrome	63 (32)	48 (25)

CDC, Centers for Disease Control and Prevention.
[a]Patients could receive more than one drug.
[b]Eight questionnaires were missing at baseline, respectively, three and five in the intermittent and continuous groups.

16 (8%) in the continuous group, but time to treatment strategy discontinuation was not statistically different between the two groups ($P=0.09$, log-rank test, data not shown). Adverse events and laboratory abnormalities, other than immunological failure, were the most common reason for treatment strategy discontinuation (11 patients in the intermittent group compared with eight in the continuous group), followed by patient choice (six patients in each group). Overall, the median duration of time under antiretroviral treatment was 51.5% [interquartile range (IQR) 49.5–55.7] in the intermittent group and 100% (IQR 100–100) in the continuous group.

Study outcomes

During the 96 weeks of follow-up, in an intention-to-treat analysis, seven patients (3.6%) in the intermittent group and three (1.5%) in the continuous group

Copyright © Lippincott Williams & Wilkins. Unauthorized reproduction of this article is prohibited.

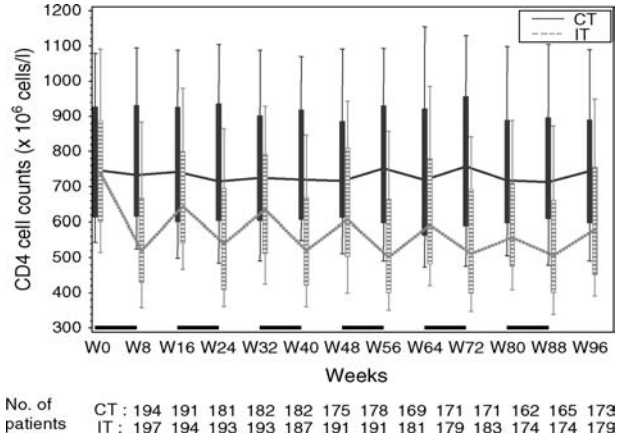

Fig. 2. Median CD4 cell counts (interquartile range, 10th and 90th percentiles) over 96 weeks in patients randomly assigned to intermittent or continuous therapy. Bold bars on the time axis represent periods off therapy. This figure shows observed data obtained within 2 weeks before or after a scheduled visit. At week 96 data were not available for three and seven patients in the intermittent (IT) and continuous (CT) group, respectively. — Continuous therapy; ---- intermittent therapy.

experienced an immunological failure, with the upper bound of the 95% confidence interval of the difference at 5.6%. In these 10 patients the median value of the nadir of the CD4 cell count was 206×10^6 cells/l (range 126–275). In the on-treatment strategy analysis, six patients (3.5%) in the intermittent group and one patient (0.6%) in the continuous group experienced immunological failure, with the upper bound of the 95% confidence interval for the difference at 6.5%, again below the predefined threshold of 7% for non-inferiority. Primary outcomes assessed by baseline or nadir CD4 cell strata, or by baseline efavirenz use indicated consistent results between groups and across strata (data not shown).

Figure 2 shows the median CD4 cell counts in both groups throughout the study. At week 96, median changes from baseline in CD4 cell counts showed a decrease of 155×10^6 cells/l in the intermittent group compared with 8×10^6 cells/l in the continuous group ($P < 0.0001$, Wilcoxon test). The median decrease in CD4 cell counts from baseline to week 8 (end of first treatment interruption) was significantly higher in the intermittent group (196 versus 12×10^6 cells/l, $P < 0.0001$). In contrast, from week 8 to week 88 (end of last treatment interruption), the median changes in CD4 cell counts were not significantly different between the groups (-48 and $+9 \times 10^6$ cells/l in the intermittent and continuous groups, respectively, $P = 0.19$). At week 96, the proportion of patients with a CD4 cell count above 450×10^6 cells/l was 75% in the intermittent group compared with 92% in the continuous group ($P < 0.0001$, chi-square test). Median CD8 cell counts remained almost unchanged in the continuous group during follow-up, but mirrored CD4 cell changes in the intermittent group (data not shown).

No patient experienced any AIDS-defining event during the study (95% confidence interval 0–1.5%). Two patients randomly assigned to the intermittent group died of non-HIV-related events. One patient had a sudden death of unknown origin 80 weeks after randomization, and the other with alcoholic cirrhosis died of hepatic failure, 48 weeks after the discontinuation of antiretroviral therapy. Forty-one patients experienced 52 HIV-related clinical events, 27 (14%) in the intermittent group and 14 (7%) in the continuous group (Table 2, $P = 0.04$, chi-square test) with no clinical progression. Lymphadenopathy and mucosal candidiasis accounted for most of these events.

In the intermittent group, plasma viral rebound was observed at the end of each treatment interruption, with a median peak of 4.6 $\log_{10}$ copies/ml of HIV RNA (data not shown) and at week 96, 8 weeks after treatment was resumed, the proportion of patients with plasma HIV-RNA levels of less than 400 copies/ml was lower than in the continuous group (81 versus 90%, respectively, $P = 0.02$, chi-square test).

Among patients enrolled in the virological substudy, baseline characteristics were well balanced between groups, and were similar to those of the whole patient population (data not shown). Resistance DNA genotype patterns performed at baseline and week 96 using DNA from peripheral blood mononuclear cells were similar in both groups (Table 3). The proportion of patients with plasma HIV-RNA levels above 1000 copies/ml after 6 weeks of treatment or more to week 96 was also similar in the intermittent and continuous groups (17 versus 14%, $P = 0.56$, chi-square test), as were plasma HIV resistance genotypes (Table 3).

Safety and tolerability

The overall incidence of grade 3–4 non-HIV-related events or laboratory abnormalities was similar in both groups (Table 2). Twelve per cent of patients in each group experienced grade 3–4 clinical adverse events. Of note is the fact that cardiovascular events were reported in three patients in the control group (myocardial infarction, angina pectoris, transient ischaemic attack) and in only one patient in the intermittent group (pulmonary embolism).

A similar proportion of patients in each group also experienced grade 3–4 laboratory abnormalities (Table 2). However, grade 3–4 thrombocytopenia occurred more frequently in the intermittent group (4.6 versus 1.0%, respectively, $P = 0.03$, chi-square test). Eighteen episodes of severe thrombocytopenia were observed after the discontinuation of antiretroviral

Copyright © Lippincott Williams & Wilkins. Unauthorized reproduction of this article is prohibited.

Table 2. HIV-related clinical events, non-HIV-related grade 3–4 clinical adverse events and laboratory abnormalities[a] over 96 weeks of follow-up.

	Intermittent group ($N=197$)	Continuous group ($N=194$)	P Chi-square test
HIV-related clinical events	34	18	
AIDS-defining event	0	0	
Acute retroviral syndrome[b]	3	0	
Persistent lymphadenopathy	10	3	
Oral/vaginal candidiasis	9	6	
Herpes simplex infection	4	2	
Herpes zoster	2	3	
Thrombocytopenia-related bleeding[c]	2	0	
Pneumonia	1	1	
Other events	3	3	
No. of patients with HIV-related clinical events[d] (%)	27 (14)	14 (7)	0.04
Clinical adverse events not HIV related	31	32	
Infectious	8	8	
Psychiatric	6	7	
Respiratory[e]	4	1	
Musculoskeletal	3	1	
Gastrointestinal	2	2	
Nervous system[f]	3	1	
Renal	2	1	
Cardiac[g]	0	2	
Other	3	9	
No. of patients with non-HIV-related clinical events[d] (%)	23 (12)	24 (12)	0.83
Laboratory abnormalities	32	26	
Neutrophils ($<750/\mu l$)	2	1	
Platelets ($<50\,000/\mu l$)[h]	9	2	
Aminotransferases (>5 ULN)	9	4	
Lipase (>5 ULN)	1	1	
Creatine phosphokinase (>5 ULN)	8	10	
Triglycerides (>8.5 mmol/l or 745 mg/dl)	2	4	
Cholesterol (>7.5 mmol/l or 298 mg/dl)	1	1	
Glucose (>16.5 mmol/l or 297 mg/dl)	0	3	
Total number of patients (%)	29 (15)	22 (11)	0.32

[a]According to Agence Nationale de Recherches sur le SIDA toxicity grading scale (http://www.anrs.fr/index.php/article/articleview/1358/1/44).
[b]Only one patient switched to a continuous regimen; the other two maintained an intermittent regimen without any recurrence of acute retroviral syndrome.
[c]Two patients with grade 4 thrombocytopenia experienced mild haemorrhagic symptoms: one had haematoma at venous puncture sites, and the other had mild gynaecological bleeding.
[d]Patients could have more than one event. Only the first occurrence of an event was reported in the Table.
[e]Respiratory events included one case of pulmonary embolism in the intermittent group, two cases of asthma and two cases of other lung disorders.
[f]Nervous system disorders included one case of transient ischaemic attack in the continuous group and three cases (paresthesia one, sciatalgia one, lipothymia one) in the intermittent group.
[g]Cardiac events included a myocardial infarction and an acute coronary heart event.
[h]The difference between arms was statistically significant ($P=0.03$, chi-square test).

therapy in 11 patients overall, nine of whom reported a history of thrombocytopenia. Five patients in the intermittent group switched to continuous treatment because of thrombocytopenia, although only two patients had haemorrhagic symptoms and these symptoms were mild. No patient experienced grade 3–4 anaemia or an increase in the creatinine level throughout the study period. Also, at week 96, median (IQR) haemoglobin levels were similar in both groups [14.2(13.5–15)versus14.3(13.4–15.2) g/dl,respectively,$P=0.48$], as were median (IQR) plasma creatinine levels [85 (75–94) versus 80 (72–92) μmol/l, respectively, $P=0.13$]. Median changes from baseline for fasting plasma glucose, triglycerides, and total cholesterol levels were not significantly different among the treatment groups throughout the study (data not shown). Also, the proportions of patients with lipodystrophy at week 96 were not different in the two groups (intermittent treatment arm 60%, continuous treatment arm 64%, $P=0.49$, chi-square test).

Discussion

In order to maintain the CD4 cell count at a level high enough to avoid the risk of AIDS-defining events during treatment interruptions, the primary study endpoint of this study was chosen as the proportion of patients with immunological failure defined as a confirmed CD4 cell count of less than 300×10^6 cells/l. Our study showed that in HIV-infected patients with high CD4 cell counts and suppression of viral replication under antiretroviral treatment, the proportion of patients

Copyright © Lippincott Williams & Wilkins. Unauthorized reproduction of this article is prohibited.

Table 3. HIV genotypic resistance patterns among patients enrolled in the virological substudy and assigned to intermittent or continuous antiretroviral therapy for 96 weeks.

Characteristic	Intermittent group ($N=98$)	Continuous group ($N=98$)
Patients with 'virological failure'[a], no. (%)	17 (17)*	14 (14)*
Patients with plasma genotype available, no.	14	10
Wild-type virus	5	2
Resistance mutations	9	8
Any NRTI-associated	7	6
M184V	6	5
Any major PI-associated	2	2
Any NNRTI-associated	2	6
K103N	1	5
Patients with viral DNA available in PBMC[b]	72	81
No. with wild-type virus at baseline (%)	49 (68)	57 (70)
At week 96 (%)[c]	47 (65)	48 (59)
No. with any NRTI mutations at baseline	20	22
At week 96	18	29
No. with M184V at baseline	9	10
At week 96	8	13
No. with any major PI mutations at baseline	2	2
At week 96	4	7
No. with any NNRTI mutations at baseline	5	5
At week 96	6	4
No. with K103N at baseline	1	1
At week 96	4	1

NNRTI, non-nucleoside reverse transcriptase inhibitor; NRTI, nucleoside reverse transcriptase inhibitor; PBMC, peripheral blood mononuclear cell; PI, protease inhibitor.
[a]'Virological failure' was defined as a plasma HIV-RNA level above 1000 copies/ml after at least 6 weeks of continuous antiretroviral therapy, and resistance was assessed whenever virological failure occurred. For subjects with multiple genotypic assessments, mutations detected in each isolate were considered in the analysis.
[b]Among patients who had peripheral PBMC DNA available both at baseline and week 96.
[c]The proportions of patients who had a plasma HIV-RNA level less than 400 copies/ml at week 96 were 87 and 79% in patients with, at week 96, wild-type viral DNA and mutated viral DNA (≥ 1 mutation), respectively ($P=0.15$, chi-square test).
*$P=0.56$, chi-square test.

with immunological failure over 96 weeks was not inferior by more than the predefined limit of 7% in the group assigned to a fixed intermittent 8 weeks off, 8 weeks on treatment compared with the continuous group.

Also, no AIDS-defining event and two deaths not related to HIV were reported during the study, both in the intermittent group. These results differ from those of the TRIVACAN and SMART CD4 cell-guided interruption trials, in which a higher incidence of disease progression and death was observed in the intermittent groups [16,17]. According to the incidence density of 3.3 events (clinical disease progression or death) per 100 patient-years of follow-up reported in the intermittent arm of the SMART study, 12 clinical disease progression events or deaths were to be expected in the intermittent group in our study instead of the two observed. One explanation for this difference could be that in both of these CD4 cell count-guided trials with a low CD4 cell threshold of 250×10^6 CD4 cells/l to resume antiviral therapy, patients could have persistently low CD4 cell counts. In the Staccato trial, where a higher threshold of 350 instead of 250×10^6 CD4 cells/l was used to resume therapy, no increased risk of clinical progression was reported [18]. Another explanation could be that in CD4 cell count-guided STI trials, patients may remain off treatment and exposed to persistent viraemia for months, whereas in the time-guided strategies patients remain off treatment exposed to viraemia for a predefined, limited period of time.

However, in our study, whereas the median CD4 cell count remained stable in the continuous group to week 96, a median drop of 155×10^6 CD4 cells/l was recorded in the intermittent group (Fig. 2); at week 96, 75% of patients in the intermittent group maintained CD4 cell counts above 450×10^6 cells/l, a proportion significantly lower than in the continuous group (92%). The kinetics of CD4 cell counts in the intermittent group had a biphasic decline, with a steep decrease after the first cycle of intermittent therapy, but less pronounced decreases during the following cycles, similar to reports in patients who stopped antiretroviral therapy [19].

Even though the incidence of HIV-related events was higher in the intermittent group compared with the continuous group (14 versus 7%, $P=0.04$), most events were limited to lymphadenopathy and mucosal candidiasis, with only three acute retroviral syndromes (two of which did not recur during further treatment interruptions).

Contrary to what was observed in the SMART study and to a lesser extent in the TRIVACAN study, the incidence of grade 3–4 non-HIV-related clinical adverse events reported

Copyright © Lippincott Williams & Wilkins. Unauthorized reproduction of this article is prohibited.

in our study was not higher in the intermittent arm (12% in both groups). Of note is the fact that cardiovascular events were reported in three patients in the control group and in only one patient in the intermittent group.

Overall, the incidence of grade 3–4 laboratory abnormalities was similar in the two groups. No grade 3–4 anaemia or increase in plasma creatinine level was observed in this study. However, thrombocytopenia occurred more frequently in the intermittent group. Five patients in this group switched to continuous treatment because of thrombocytopenia, although only two had mild haemorrhagic symptoms. Thrombocytopenia after the discontinuation of antiretroviral therapy has been reported in a number of studies, among patients with a previous history of thrombocytopenia: these patients should be discouraged to interrupt therapy [18,20]. No benefit of treatment interruptions on metabolic abnormalities and body-fat distribution was seen in our study. Also, no change in body-fat distribution was observed in either group during follow-up, with the limitation that no objective measure of body-fat composition was performed. Although the convenience of the intermittent regimen was mentioned by a number of patients, we currently have no data available to support that the quality of life of patients receiving this strategy was improved.

The proportion of patients with plasma HIV-RNA levels below 400 copies/ml was high in both groups at week 96 in our study, although significantly lower in the intermittent group. However, these patients had resumed antiretroviral therapy only 8 weeks earlier, and this was probably too short a time to control HIV replication fully. It is likely that with a longer duration of antiretroviral therapy, a higher proportion of patients in the intermittent group would also have suppressed viral replication, as described in other STI studies [18,21].

Previous STI studies have underlined the risk of selecting drug-resistance mutations in plasma during interruptions, especially to drugs with a low genetic barrier to resistance [22–26]. In this study, the proportion of patients enrolled in the virological substudy who experienced a plasma HIV-RNA level above 1000 copies/ml after 6 weeks of antiretroviral therapy or more was similar in both groups over 96 weeks of follow-up, with the same pattern of HIV genotypic resistance mutations. Interestingly, patterns of genotypic resistance were also similar in both groups in peripheral blood mononuclear cell DNA at week 96, with no increase in the number of patients with resistant strains in the intermittent group, although 43 and 85% of these patients received a non-nucleoside reverse transcriptase inhibitor or a lamivudine/emtricitabine-based regimen, respectively, and although 39% of these patients had a history of previous treatment failure. These results are similar to those of the Staccato trial in which very few resistance mutations were detected [18]. Other studies, however, have shown a higher rate of detection of resistance mutations, but these mutations were often already present at baseline and did not always impact on the short-term virological response to subsequent antiretroviral therapy [27–30].

Our study has, however, several limitations. First, the results obtained in the intermittent arm cannot be generalized to patients different from those we studied, i.e. mostly men in their forties, with median CD4 cell counts of 274×10^6 cells/l at nadir and of 739×10^6 cells at baseline after 5 years of antiretroviral therapy. Second, because of the longer half-life of efavirenz, we stopped this drug 7 days before the others. However, the clearance of efavirenz varies greatly between individuals [31]. Although not observed in this study, the risk of the emergence of resistance after the interruption of an efavirenz (or nevirapine)-based treatment remains a concern. Third, the results we obtained are limited to the 96-week duration of the study, and it is uncertain that this treatment strategy could be used lifelong as a CD4 cell count decline was observed over time. Also, it is uncertain whether the level of clinical support needed to ensure proper adherence to an intermittent treatment strategy could be offered in clinical practice.

In summary, an intermittent 8-week off and on treatment strategy offers the possibility of short treatment holidays with a low risk ($< 7\%$) of CD4 cell counts falling below 300×10^6 cells, without AIDS-defining events and without strategy-induced resistance. However, this strategy was associated with a CD4 cell count decline, a higher risk of minor HIV-related events, and a lower proportion of patients resuppressing viral replication after retreatment. More studies are therefore needed to elucidate the place of time-guided intermittent antiretroviral therapy in clinical practice.

Sponsorship: This study was supported by the Agence Nationale de Recherches sur le SIDA et les Hépatites Virales (ANRS), ANRS 106 trial – Window.

References

1. Palella FJ Jr, Delaney KM, Moorman AC, Loveless MO, Fuhrer J, Satten GA, *et al*. **Declining morbidity and mortality among patients with advanced human immunodeficiency virus infection.** *N Engl J Med* 1998; **338**:853–860.
2. Mocroft A, Ledergerber B, Katlama C, Kirk O, Reiss P, d'Arminio Monforte A, *et al*. **EuroSIDA study group. Decline in the AIDS and death rates in the EuroSIDA study: an observational study.** *Lancet* 2003; **362**:22–29.
3. Davey RT Jr, Bhat N, Yoder C, Chun TW, Metcalf JA, Dewar R, *et al*. **HIV-1 and T cel dynamics after interruption of highly active antiretroviral therapy (HAART) in patients with a history of sustained viral suppression.** *Proc Natl Acad Sci U S A* 1999; **96**:15109–15114.
4. Hatano H, Vogel S, Yoder C, Metcalf JA, Dewar R, Davey RT, *et al*. **Pre-HAART burden approximates post-HAART viral levels following interruption of therapy in patients with sustained viral interruption.** *AIDS* 2000; **14**:1357–1363.
5. Carr A, Cooper DA. **Adverse effects of antiretroviral therapy.** *Lancet* 2000; **356**:1423–1430.

Copyright © Lippincott Williams & Wilkins. Unauthorized reproduction of this article is prohibited.

6. Dubé MP, Stein JH, Aberg JA, Fichtenbaum CJ, Gerber JG, Tashima KT, *et al*., Adult AIDS Clinical Trials Group Cardiovascular Subcommittee; HIV Medical Association of the Infectious Disease Society of America. **Guidelines for the evaluation and management of dyslipidemia in human immunodeficiency virus (HIV)-infected adults receiving antiretroviral therapy: recommendations of the HIV Medical Association of the Infectious Disease Society of America and the Adult AIDS Clinical Trials Group.** *Clin Infect Dis* 2003; **37**:613–627.
7. Fagard C, Oxenius A, Gunthard H, Garcia F, Le Braz M, Mestre G, *et al*., Swiss HIV Cohort Study. **A prospective trial of strategic treatment interruptions in human immunodeficiency virus infection.** *Arch Intern Med* 2003; **163**:1220–1226.
8. Oxenius A, Price DA, Gunthard HF, Dawson SJ, Fagard C, Perrin L, *et al*. **Stimulation of HIV-specific cellular immunity by structured treatment interruption fail to enhance viral control in chronic HIV infection.** *Proc Natl Acad Sci U S A* 2002; **99**:13747–13752.
9. Walmsley S, Loutfy M. **Can structured treatment interruptions (STIs) be used as a strategy to decrease total drug requirements and toxicity in HIV infection?** *J Int Assoc Physicians AIDS Care* 2002; **1**:95–103.
10. D'Arminio Monforte A, Cozzi-Lepri A, Phillips A, De Luca A, Murri R, Mussini C, *et al*. **Interruption of highly active antiretroviral therapy in HIV clinical practice.** *J Acquir Immune Defic Syndr* 2005; **38**:407–416.
11. Chene G, Angelini E, Cotte L, Lang JM, Morlat P, Rancinan C, *et al*. **Role of long-term nucleoside-analogue therapy in lipodystrophy and metabolic disorders in human immunodeficiency virus-infected patients.** *Clin Infect Dis* 2002; **34**:649–657.
12. Pasquier C, Millot N, Njouom R, Sandres K, Cazabat M, Puel J, *et al*. **HIV-1 subtyping using phylogenetic analysis of pol gene sequences.** *J Virol Methods* 2001; **94**:45–54.
13. Descamps D, Delaugerre C, Masquelier B, Ruffault A, Marcelin AG, Izopet J, *et al*. **Repeated HIV-1 resistance genotyping external quality assessments improve virology laboratory performance.** *J Med Virol* 2006; **78**:153–160.
14. Johnson VA, Brun-Vezinet F, Clotet B, Conway B, Kuritzkes DR, Pillay D, *et al*. **Update of the drug resistance mutations in HIV-1: fall 2005.** *Top HIV Med* 2005; **13**:125–131.
15. Centers for Disease Control and Prevention. **1993 Revised classification system for HIV infection and expanded surveillance case definition for AIDS among adolescents and adults**. *MMWR Recomm Rep* 1992; **41**:1–19.
16. Danel C, Moh R, Minga A, Anzian A, Ba-Gomis O, Kanga C, *et al*. **CD4-guided structured antiretroviral treatment interruption strategy in HIV-infected adults in west-Africa adults (Trivacan ANRS 1269 trial): a randomized trial.** *Lancet* 2006; **367**:1981–1989.
17. El-Sadr W, Neaton J, for the SMART study investigators. The strategies for management of antiretroviral therapy (SMART) study group. **CD4+ count-guided interuption of antiretroviral treatment.** *NEJM* 2006; **355**:2283–2296.
18. Ananworanich J, Gayet-Ageron A, Le Braz M, Prasithsirikul W, Chetchotisakd P, Kiertiburanakul S, *et al*. **CD4-guided scheduled treatment interruptions compared with continuous therapy for patients infected with HIV-1: results of the Staccato randomised trial.** *Lancet* 2006; **368**:459–465.
19. Fagard C, Bandelier CY, Ananworanich J, Le Braz M, Gunthard H, Perneger T, *et al*., Staccato/SSITT Study Group. **Biphasic decline of CD4 cell count during scheduled treatment interruptions.** *AIDS* 2005; **19**:439–441.
20. Ananworanich J, Phanuphak N, Nuesch R, Apateerapong W, Rojnuckarin P, Ubolyam S, *et al*. **Recurring thrombocytopenia associated with structured treatment interruption in patients with human immunodeficiency virus infection.** *Clin Infect Dis* 2003; **37**:723–725.
21. Garcia F, Plana M, Vidal C, Cruceta A, O'Brien WA, Pantaleo G, *et al*. **Dynamics of viral load rebound and immunological changes after stopping effective antiretroviral therapy.** *AIDS* 1999; **13**:F79–F86.
22. Martinez-Picado J, Morales-Lopetegi K, Wrin T, Prado JG, Frost SD, Petropoulos CJ, *et al*. **Selection of drug-resistant HIV-1 mutants in response to repeated structured treatment interruptions.** *AIDS* 2002; **16**:895–899.
23. Dybul M, Nies-Kraske E, Daucher M, Hertogs K, Hallahan CW, Csako G, *et al*. **Long-cycle structured intermittent versus continuous highly active antiretroviral therapy for the treatment of chronic infection with human immunodeficiency virus: effects on drug toxicity and on immunologic and virologic parameters.** *J Infect Dis* 2003; **188**:388–396.
24. Yerly S, Fagard C, Gunthard HF, Hirschel B, Perrin L. **Drug resistance mutations during structured treatment interruptions.** *Antivir Ther* 2003; **8**:411–415.
25. Metzner KJ, Bonhoeffer S, Fischer M, Karanicolas R, Allers K, Joos B, *et al*., the Swiss HIV Cohort Study. **Emergence of minor populations of human immunodeficiency virus type 1 carrying the M184V and L90M mutations in subjects undergoing structured treatment interruptions.** *J Infect Dis* 2003; **188**:1433–1443.
26. Palmisano L, Giuliano M, Bucciardini R, Andreotti M, Fragola V, Galluzzo C, *et al*., Italian ISS PART Clin Ctrs. **Final results of a randomized controlled trial of structured treatment interruptions vs continuous HAART in chronic HIV-infected subjects with persistent suppression of viral replication**. In: *13th Conference on Retroviruses and Opportunistic Infections*. Denver, CO, USA, 5–8 February 2006 [Abstract 103].
27. Papasavvas E, Grant RM, Sun J, Mackiewicz A, Pistilli M, Gallo C, *et al*. **Lack of persistent drug-resistant mutations evaluated within and between treatment interruptions in chronically HIV-1-infected patients.** *AIDS* 2003; **17**:2337–2343.
28. Papasavvas E, Kostman JR, Mounzer K, Grant RM, Gross R, Gallo C, *et al*. **Randomized, controlled trial of therapy interruption in chronic HIV-1 infection.** *PLoS Med* 2004; **1**:e64.
29. Nuesch R, Ananworanich J, Sirivichayakul S, Ubolyam S, Siangphoe U, Hill A, *et al*. **Development of HIV with drug resistance after CD4 count-guided structured treatment interruptions in patients treated with highly active antiretroviral therapy after dual-nucleoside analogue treatment.** *Clin Infect Dis* 2005; **40**:728–734.
30. Arnedo-Valero M, Garcia F, Gil C, Guila T, Fumero E, Castro P, *et al*. **Risk of selecting de novo drug-resistance mutations during structured treatment interruptions in patients with chronic HIV infection.** *Clin Infect Dis* 2005; **41**:883–890.
31. Ribaudo H, Haas D, Tierney C, Kim R, Wilkinson G, Guilck R, *et al*. **Pharmacogenetics of plasma efavirenz exposure after treatment discontinuation: an adult AIDS Clinical Trials Group study.** *Clin Infect Dis* 2006; **42**:401–407.

Appendix

The members of Window ANRS 106 Study Team were as follows: Trial chair, B. Marchou; Trial co-chair, J.M. Molina; Trial coordinator and monitors, P. Tangre, J.P. Aboulker; Trial statistician, I. Charreau; Trial virologists, J. Izopet; Scientific committee, B. Marchou, J. Izopet, P. Tangre, J.P. Aboulker, P.M. Girard, T. May, J.M. Molina; Data safety and monitoring board, M. Seligmann, F. Brun-Vezinet, A. Laplanche; Coordinating trial centre: INSERM SC10, J.P. Aboulker, P. Tangre, A. Uludag, M. Saouzanet, V. Eliette, C. Lascoux, S. Delmas, S. Gueguen, A. Polaert, I. Charreau (statistics), B Guillon and Y Saïdi (data management).

Participating centres and investigators (all in France): Hôpital Debré, Reims: Remy, Strady, Rouger, Beguinot, Berger, Brodard, Tabary, Gourdier; Hôpital Lagny, Marne la Vallée: Lagarde, David-Ouaknine, Simon, Froguel, Le Rudulier, Costa, Louin; Hôpital Avicenne, Bobigny: Guillevin, Jarrousse, Krivitzki, Lortholary, Bentata, Klutse, Djebbar, Makki, Belarbi, Obenga, Honoré, Touam, Mansouri, Baazia, Feuillard, Soreda; Hôpital Bellevue, St Etienne: Lucht, Fresard, Cazorla, Ronat, Saoubin, Lambert,

Copyright © Lippincott Williams & Wilkins. Unauthorized reproduction of this article is prohibited.

Simoens; Hôpital St Jacques, Besançon: Estavoyer, Hoen, Pichon, Coquet, Bettinger, Tiberghien, Legalery; Hôpital Pellegrin, Bordeaux: Dupon, Ragnaud, Raymond, Dutronc, Ochoa, Lafabie, Neau, Chambon, Garrigue, Moreau, Dupin; Hôpital Bicêtre, Kremlin-Bicêtre: Delfraissy, Goujard, Robquin, Segeral, Quertainmont, Guillet, Lambert, Rannou, Taburet, Bocquentin, Idri, Gubler; Hôpital Lariboisière, Paris: Bergmann, Sellier, Diemer, Bendenoun, Rami, Parinello, Mazeron, Boval, Roux; Hôpital R. Poincaré, Garches: Perronne, De Truchis, Melchior, Salomon, Berthe, Mathez, Paillet; Hôpital P. Brousse, Villejuif: Vittecoq, Teicher, Smadja, Bergerol, Merad, Minozzi, Bolliot, Mallet, Bensidhoum, Mackiewicz, Kara Terki, Rudant; Hôpital G. Pompidou, Paris: Kazatchkine, Weiss, Gonzales-Canali, Haddadi, Piketty, Karhaman, Karmochkine, Bengrait, Si Mohamed, Caccavelli, Sabatier; Hôpital Bichat, Paris: Yeni, Vilde, Leport, Bouvet, Al Kaied, Charlois, Jestin, Benabdelmoumen, Fournier, Hadjoudj, Phung, Gaudebout, Le Gac, Pahlavan, Zeng, Chams, Elbim, Certain; Hôpital Cochin, Paris: Sicard, Salmon-Ceron, Spiridon, Krivine, Lacombe, Guerin; Hôpital Foch, Suresnes: Bletry, Zucman, Majerholc, Honderlick, Drupt, Bessard; Hôpital Pitié-Salpêtrière, Paris: Bricaire, Herson, Katlama, Simon, Tubiana, Ghosn, Duvivier, Boubezari, Schoen, Curjol, Kouadio, Amirat, Bonmarchand, Capitaine, Lambert, Brançon, Carcelain, Samri, Hrichi, Fievet; Hôpital St Antoine, Paris: Girard, Meynard, Meyohas, Bollens, Roussard, Lefebvre, Besse, Berriot, Gaujour, Lagneau, Lupin, Morand-Joubert, Rosenzwajg, Charrois, Archi, Daguenel-Nguyen; Hôpital St Louis, Paris: Molina, Ponscarme, Tourneur, Fournier, Ferret, Balkan, Bani Sadr, De Castro, Garrait, Goguel, Lafaurie, Neuville, Furco, Perignon, Maslo, Colin de Verdière, Rachline, Loze, Schnell, Palmer, Rabian, Madelaine; Hôpital St André, Bordeaux: Beylot, Morlat, Lacoste, Bernard, Malvy, Nouts, Pertusa, Bonarek, Thibault, Garrigue, Moreau, Pedeboscq; Hôpital E. Herriot, Lyon: Touraine, Livrozet, Jeanblanc, Makhloufi, Brunel, Palmer, Tardy, Malcus, Nageotte; Hôpital Hôtel-Dieu, Lyon: Trepo, Augustin-Normand, Benmakhlouf, Cotte, Bailly, Miailhes, Schlienger, Thoirain, Brochier, Ritter, Bataillard; Hôpital Ste Marguerite, Marseille: Gastaut, Poizot-Martin, Frixon-Marin, Dinh, Dignat-Georges, Tamalet, Penot-Ragon; Hôpital Gui de Chauliac, Montpellier: Reynes, Atoui, Baillat, Lotthe, Siffert, Le Moing, Merle de Boever, Vidal, Tramoni, Montes, Pages, Giraudon; Hôpital Hôtel-Dieu, Nantes: Raffi, Billaud, Milpied, Bugnon, Bonnet, Hue, Ferre, Poirier, Audrain, Lepelletier; Hôpital Les Oudairies, La Roche sur Yon: Perre, Aubry, Desailly-Chanson, Ferre, Berruchon; Hôpital L'Archet, Nice: Dellamonica, Cassuto, Ceppi, Rahelinirina, Poirée, Bagot, Cottalorda, Benhamou, Ticchioni, Rigault; CHRU de Strasbourg: Lang, Cheneau, Hess, Kempf, Priester, Rey, Fischer, Ebel, Schmitt, Uring-Lambert, Hutt; Hôpital Bretonneau, Tours: Choutet, Bastides, Besnier, Nau, Vauthier, Dailloux, Guerois, Barin, Thibault, Chartrin; Hôpital Purpan, Toulouse: Massip, Marchou, Cuzin, Obadia, Bicart-See, Bonnet, Khatibi, Alvarez, Labau, Picot, Balsarin, Jaafar, Izopet, Kuhlein, Peyranne; CHU de Clermont-Ferrand: Beytout, Laurichesse, Jacomet, Henquell, Chassagne, Coudert; CHU de Grenoble: Leclercq, Blanc, Trappo, Dufresnes, Schmuck, Jacob, Boitard; Centre Hospitalier de Tourcoing: Mouton, Cheret, Dos Santos, De la Tribonnière, Vandamme, Labalette, Bocket, Dubar; CHU Brabois, Nancy: May, Boyer, Ahmed Khalifa, Kolopp-Sarda, Le Faou, Demore; Hôpital Croix-Rousse, Lyon: Peyramond, Boibieux, Braun, Lippmann, Cozon, Ritter, Charpiat; Hôpital La Grave, Toulouse: Viraben, Aquilina, Cuzin, Abboud, Lucas, Prevoteau du Clary, Barone, Pomies.

Author contributions: As chair and co-chair, Drs Marchou and Molina had full access to the data in this study, and take responsability for the integrity of the data and for the accuracy of the data analyses. Study concept and design: Marchou, Aboulker, Molina. Acquisition of data: Tangre, Marchou, Molina, Aboulker. Analysis and interpretation of data: Marchou, Charreau, Tangre, Izopet, Girard, May, Aboulker. Critical revision of the manuscript for important intellectual content: Marchou, Charreau, Tangre, Izopet, Girard, Ragnaud, May, Aboulker. Statistical expertise: Charreau, Aboulker. Study supervision: Marchou, Molina, Aboulker, Izopet, Tangre.

Copyright © Lippincott Williams & Wilkins. Unauthorized reproduction of this article is prohibited.