

# Toxicity and drug exposure in a quadruple drug regimen in HIV-1 infected patients participating in the ADAM study

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**Objective:** To study the relationship between toxicity and the exposure to nelfinavir and saquinavir as part of a quadruple drug regimen.

**Design:** The ADAM study is a randomized study to investigate the feasibility of induction-maintenance therapy in HIV-1 infection.

**Methods:** HIV-1-infected patients with no prior use of antiretroviral treatment started induction therapy consisting of stavudine + lamivudine + nelfinavir + saquinavir for a period of 26 weeks. Data regarding toxicity of the quadruple regimen and exposure to the protease inhibitors were collected.

**Results:** Seven of the 65 patients enrolled had to switch therapy for reasons of toxicity within the first 26 weeks. Diarrhoea was frequently reported (49 of 65, one discontinuation), but could be relieved by using antidiarrhoeal agents. Laboratory monitoring revealed elevated liver enzymes (leading to four discontinuations) and mild to moderate elevations of triglycerides and cholesterol (nine and 23 of 65, respectively). The exposure to saquinavir and nelfinavir was lower than expected. Abdominal pain was associated with a higher exposure to nelfinavir or saquinavir. The association of nausea and abdominal distension with drug exposure appeared to vary over time.

**Conclusions:** The quadruple drug regimen was quite well tolerated. Diarrhoea was frequently reported but could be relieved by the use of antidiarrhoeal agents. In comparison with other protease inhibitor combinations, lipid abnormalities in plasma were infrequent and mild. With the exception of diarrhoea, all gastrointestinal complaints observed were found to be associated with the level of exposure to nelfinavir or saquinavir. The exposure to the protease inhibitors was relatively low, although the virologic efficacy of the regimen used was satisfactory.

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## Introduction

A durable suppression of viral replication in HIV-1 infection can be achieved by giving a combination of three antiretroviral agents [1–3]. Highly active antiretroviral therapy (HAART) can result in clinical benefit in terms of a prolonged (disease-free) survival [4,5]. The use of more than three agents or a higher exposure to a specific antiretroviral agent might even increase the potency of an antiretroviral regimen [6–9]. However, the occurrence of side effects may hamper the use of intensified treatment regimens [10,11].

Protease inhibitor combinations are pharmacokinetically favourable in antiretroviral therapy [12,13]. All protease inhibitors show mild to moderate gastrointestinal side effects in addition to a rather unique safety and tolerability profile. Recently, the association between the use of protease inhibitors and the occurrence of a new onset of diabetes mellitus, hyperlipidaemia or peripheral lipodystrophy has been described [14]. However, there is only limited data regarding the safety profiles of PI combinations, such as zidovudine or zalcitabine plus saquinavir [15–19].

A quadruple induction regimen, consisting of stavudine, lamivudine, nelfinavir, and saquinavir, was used for at least 26 weeks in antiretroviral therapy-naïve patients in the Amsterdam Duration of Antiretroviral Medication (ADAM) study [20]. In this study, the feasibility of an induction-maintenance strategy in HAART was investigated. The preliminary results of the efficacy of this induction-maintenance regimen have already been described [20]. In this paper, we focus on the toxicity of the quadruple induction regimen used in the ADAM study and its relationship with the exposure to nelfinavir and saquinavir.

## Materials and methods

### Patients

The enrolment of HIV-1-infected patients in this open-label randomized-controlled study started in March 1997 and was prematurely discontinued on April 6, 1998, as the interim analysis demonstrated increased viral replication in patients on maintenance therapy [20]. Patients, aged 18 years or more, were eligible for participation if they had a CD4+ cell count of at least  $200 \times 10^6/l$  in their peripheral blood, 1000 or more HIV-1 RNA copies/ml in plasma, and if they were antiretroviral therapy-naïve. Exclusion criteria were the existence of an active opportunistic infection, active hepatitis C or presence of the hepatitis B surface antigen, women who were breast-feeding or pregnant, the use of immunomodulatory drugs or investigational drugs up to 1 month prior to the start of the study

medication, and certain laboratory parameters [Hb < 7 mmol/l (male) or < 6.5 mmol/l (female), neutropenia <  $0.75 \times 10^9/l$ , aspartate-amino transferase (ASAT)/alanine-amino transferase (ALAT) > 5 × upper limit of normal (ULN), serum creatinine > 1.5 × ULN]. Drugs such as rifampin or ketoconazole, which have a strong pharmacokinetic interaction with protease inhibitors, were not allowed. Written informed consent was obtained from all patients. The Institutional Review Boards of all participating centres approved the study protocol.

### Study design

All patients started therapy with a quadruple drug regimen consisting of stavudine (40 mg twice daily, or 30 mg twice daily for those with body weight < 60 kg), lamivudine (150 mg twice daily), nelfinavir (750 mg three times a day) and saquinavir hard-gelatin-capsules (saquinavir-HGC, 600 mg three times a day). When saquinavir soft-gelatin-capsules (saquinavir-SGC) became available (1 November 1997), all patients using saquinavir-HGC switched to saquinavir-SGC (800 mg three times a day). Roche (Roche NL, Mijdrecht, the Netherlands) provided both nelfinavir and saquinavir-SGC. Patients were instructed to take their medication with food.

### Follow-up

During the induction phase, patients were scheduled to visit the outpatient clinic for clinical assessment and routine laboratory monitoring at the start of treatment and at weeks 1, 2, 4, 8, 16, 24, 25 (for plasma HIV-1 RNA concentration assessment only) and 26. Laboratory monitoring included plasma HIV-1 RNA concentration [Nuclisens HIV-1 RNA QT assays (Organon Teknika, Boxtel, The Netherlands)], CD4+ and CD8+ cell count and plasma concentrations of nelfinavir and saquinavir. At baseline no plasma concentrations of nelfinavir and saquinavir were assessed.

### Toxicity grading

The occurrence of side effects (signs, symptoms, or laboratory abnormalities) was assessed during each study visit. The severity of each event was graded according to the World Health Organization (WHO) classification (grades 1 to 4; Appendix 1) and the probability of a relationship with the study medication used was indicated (see also Appendix 1) [21]. As the abnormalities in plasma cholesterol were not graded in the WHO classification, a more recently defined grading system by the AIDS Clinical Trial Group (ACTG) was used for the analysis of this parameter [22]. The probability of a relationship with the study medication used was also indicated and the use of concomitant medication to relieve side effects was recorded.

## Assessment of drug exposure

The quantification of plasma concentrations of nelfinavir and saquinavir was performed using a validated and sensitive reverse-phase high-performance liquid chromatography (RP-HPLC) assay [23]. For each sample the drug plasma concentration was divided by the expected drug concentration at the corresponding time-point, to adjust for the time interval between drug ingestion and the drawing of the sample. The expected concentrations of nelfinavir or saquinavir at different time points were obtained from full (8 h) pharmacokinetic profiles of both drugs assessed in 18 patients participating in the ADAM study [24]. These plasma concentration ratios were used as a measure of exposure to both protease inhibitors.

## Treatment failure and discontinuations

After attaining a plasma HIV-1 RNA concentration below the quantification limit of the ultrasense assay (< 50 copies/ml), a plasma HIV-1 RNA concentration above 400 copies/ml at two consecutive time points was considered as a treatment failure.

In case of a grade 4 toxicity (WHO classification) or grade 3 toxicity with no improvement after temporary discontinuation (maximum 2 weeks), or the recurrence of a grade 3 toxicity after re-challenge, permanent discontinuation of the study medication was obligatory [21]. Further therapy was at the discretion of the investigator after treatment failure or discontinuation of the study medication.

## Analysis

The baseline values and the changes from baseline to week 4, 8, 16 and 26 for plasma HIV-1 RNA concentration, and CD4+ and CD8+ cell counts of all patients were assessed. Patients with and without a specific side effect were compared for baseline values of these parameters and the change from baseline to week 26 (t-test).

The proportion of patients experiencing a specific side effect was assessed. In addition, the duration of the side effect within the first 6 months was calculated. Only side effects with an incidence of > 10% and a possible relationship with the study medication used were taken into account for analysis.

The correlation coefficient between nelfinavir and saquinavir per patient was assessed. For each patient, the median plasma concentration ratios of nelfinavir and saquinavir over time were calculated. To evaluate whether the plasma concentration ratio of nelfinavir and saquinavir varied over time, a generalized linear model with mixed effects was estimated (Mixed Models procedure of the statistical package SAS 6.12 for Windows, SAS Institute, Cary, North Carolina, USA).

The median plasma concentration ratios of nelfinavir and saquinavir of patients on treatment with or without a specific side effect were compared (Wilcoxon test). In addition to this comparison between groups of patients, the odds ratio for experiencing a specific side effect at a specific time point for the corresponding drug exposure was estimated by using the generalized estimating equations (GEE) method, taking into account the 'within patient' correlation between time points [25]. Time (as a factor) and plasma concentration ratio (as a covariate) were included in the model as main effects, as well as the interaction between these two variables. The analysis was based on the logistic link function and the exchangeable working correlation matrix. Calculations were performed by using the GENMOD procedure of the statistical package SAS 6.12 for Windows. To calculate *P*-values from the results of the GENMOD procedure, a SAS macro was written using the Wald statistic.

## Results

### Patients and discontinuations

In total, 65 patients (61 males, 94%) were enrolled. The baseline characteristics of these patients are summarized in Table 1. Two patients were lost to follow-up (one for logistic reasons, one withdrew from further medical follow-up after moving to another city). One patient discontinued his study medication due to virologic failure at week 24. Of the remaining 62 patients, seven patients discontinued the study medication due to side effects within the first 26 weeks (see 'Toxicity').

### CD4+ and CD8+ cell count and plasma HIV-1 RNA concentrations during therapy

The median CD4+ cell count rose from  $410 \times 10^6$  at baseline to  $560 \times 10^6$  cells/l at week 26. The median CD8+ cell count decreased from  $1050 \times 10^6$  at baseline to  $970 \times 10^6$  cells/l at week 26. The median baseline plasma HIV-1 RNA concentration was 4.51  $\log_{10}$  copies/ml. At weeks 4, 8, 16 and 26, the median

**Table 1.** Baseline characteristics.

Characteristic	All patients (n = 65)
Male <sup>a</sup>	61 (94)
Age (years) <sup>b</sup>	39 ± 8
CDC classification [26]	
A <sup>a</sup>	47 (72)
B <sup>a</sup>	16 (25)
C <sup>a</sup>	2 (3)
CD4+ cell count ( $\times 10^6$ cells/l) <sup>c</sup>	410 (320–510)
CD8+ cell count ( $\times 10^6$ cells/l) <sup>c</sup>	1050 (910–1460)
$\log_{10}$ HIV-1 RNA (copies/ml) <sup>c</sup>	4.51 (4.18–5.04)

Values are <sup>a</sup>number (%); <sup>b</sup>mean ± SD; <sup>c</sup>median (interquartile range).

plasma HIV-1 RNA concentration declined to 2.58, 2.42, 2.25 and 1.54 log<sub>10</sub> copies/ml, respectively.

### Toxicity

The frequency of the occurrence of side effects, the median of the total duration of the side effect per patient in this population and the percentage with a grade 1, 2, 3, or 4 severity are summarized in Table 2. If the use of loperamide was longer than the duration of diarrhoea, the duration of diarrhoea was extended for the period during which loperamide was used and the severity was graded as 1.

### Clinical side effects

Forty-nine of the 65 patients suffered from diarrhoea, mostly consisting of loose or watery stools, two or three times daily. Loperamide was used in 30 patients for a mean duration of 128 days (SD: 61) to relieve the complaint. Diarrhoea led to discontinuation of the study medication in one patient, after 8 weeks. Other side effects leading to discontinuation of the study medication were headache (one patient, week 22), and peripheral neuropathy, probably related to the use of stavudine (one patient, week 24). In addition to gastrointestinal side effects such as abdominal pain, nausea and abdominal distension, fatigue and headache were frequently reported. No peripheral lipodystrophy was observed within the first 26 weeks of treatment.

### Abnormalities of laboratory values

Mild to severe elevations of the liver enzymes occurred in 12 patients. In four of these patients the elevated liver enzymes led to a discontinuation of the study medication (at weeks 4, 8, 16, and 20, respectively). The occurrence of elevated cholesterol in plasma was frequent; 23 out of 65 patients (35%). However, the cholesterol level rose above 7.7 mmol/l in only six of these 23 patients. Hypertriglyceridaemia was less frequent and did not exceed grade 2 severity. No hyperglycaemia or glycosuria was observed within the first 26 weeks of treatment.

### Toxicity and patient characteristics

Patients with or without a specific complaint did not differ with respect to their baseline characteristics. Only patients complaining about fatigue had a lower baseline CD4+ cell count and a higher plasma HIV-1 RNA concentration compared to patients without this complaint (t-test,  $P = 0.02$  and  $P = 0.03$ , respectively). The patients complaining of abdominal pain had a greater decrease in their plasma HIV-1 RNA concentration at week 26 than patients without abdominal pain (t-test,  $P = 0.02$ ; data not shown).

### Drug exposure

The correlation coefficient between the nelfinavir and saquinavir plasma concentrations per patient was calculated. The mean correlation coefficient in this group of patients was 0.55 [95% confidence interval (CI) 0.41–0.67]; calculation of the confidence intervals was based on the Fisher transformation of the mean. As a result of unknown time intervals (e.g. either an unknown time of medication intake and/or the drawing of the sample), plasma drug concentration ratios could only be calculated at two or more time points in 56 patients. The median plasma concentration ratio over time per patient for nelfinavir and saquinavir varied between 0.11 and 1.31 for nelfinavir [median 0.60; inter-quartile range (IQR) 0.44–0.86] and between 0.11 and 6.50 for saquinavir (median 0.75; IQR 0.42–1.37). For both nelfinavir and saquinavir, the median plasma drug concentration ratios were significantly lower than 1 (95% CI 0.59–0.93 and 0.51–0.68, respectively); calculation of the confidence intervals was based on the geometric mean. The plasma concentration ratios of nelfinavir and saquinavir in these patients did not change over time (Fig. 1; analysis of repeated measures  $P = 0.76$  and  $P = 0.68$  for nelfinavir and saquinavir, respectively).

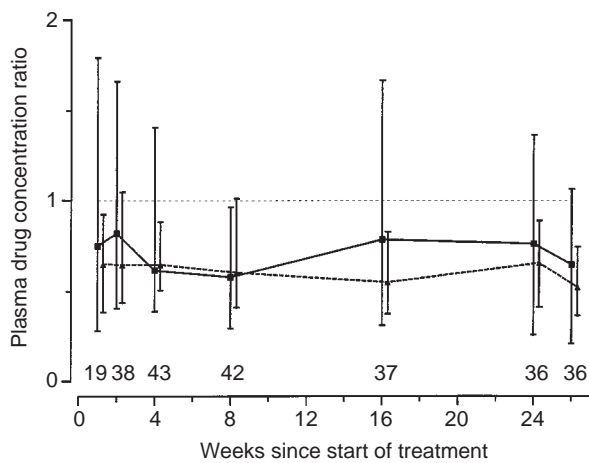
### Toxicity and drug exposure

The median plasma drug concentration ratios over time of patients with and without a specific complaint, are

**Table 2.** Toxicity.

Side effect	Frequency n (%)	Duration (days) Median (IQR)	Severity (%) <sup>a</sup>			
			1	2	3	4
<b>Clinical</b>						
Diarrhoea <sup>b</sup>	49 (75)	98 (42–164)	51	43	6	–
Nausea	13 (20)	12 (4–162)	77	23	–	–
Abdominal pain	9 (14)	40 (13–122)	57	29	14	–
Abdominal distension	8 (12)	63 (19–107)	75	25	–	–
Fatigue	18 (27)	55 (21–167)	72	28	–	–
Headache	11 (17)	44 (30–127)	73	18	–	–
<b>Laboratory</b>						
Elevated liver enzymes	12 (18)	27 (14–56)	50	17	17	16
Elevated cholesterol	23 (35)	14 (14–67)	–	88	12	–
Elevated triglycerides	9 (14)	70 (14–168)	–	100	–	–

<sup>a</sup>See also Appendix 1 [21,22]; <sup>b</sup>Corrected for the use of loperamide. IQR, interquartile range.



**Fig. 1.** Median plasma concentration ratios of nelfinavir (dashed line) and saquinavir (solid line) over time. The numbers at the bottom of the figure represent the number of samples available for the different time points. Bars represent interquartile ranges.

tabulated in Table 3. Patients with abdominal pain appeared to have higher plasma concentration ratios for nelfinavir and saquinavir than patients without this complaint (Wilcoxon test,  $P = 0.02$  and  $P = 0.05$ , respectively). The patients with diarrhoea had lower saquinavir plasma concentration ratios than patients without diarrhoea (Fig. 2; Wilcoxon test,  $P = 0.04$ ). No differences in drug exposure were found for any of the other side effects.

Using the GEE method, the occurrence of diarrhoea was found to be associated with the exposure to

nelfinavir and not with the exposure to saquinavir (Fig. 3a), even if the diarrhoea was not corrected by the use of loperamide (data not shown). For the other gastrointestinal complaints, there was evidence of a time-dependent relationship with the plasma drug concentration ratios (Figs 3b–d). Both abdominal distension and nausea became associated with a low exposure to nelfinavir and saquinavir after the first weeks of therapy and for nausea, this association seemed to increase over time ( $P < 0.001$ ). The association between abdominal pain and a high drug exposure remained present during the entire treatment period ( $P < 0.001$ ). For all other complaints, no evidence was found for a relationship with plasma drug exposure (data not shown).

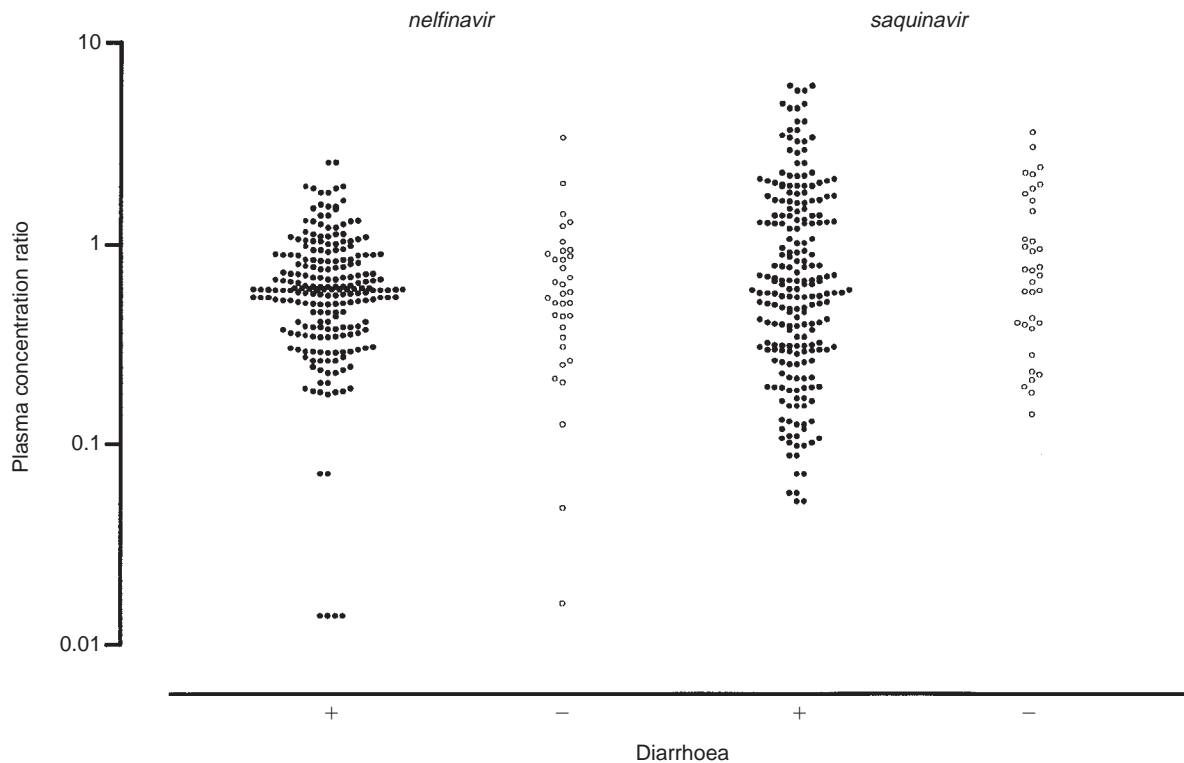
## Discussion

The quadruple induction therapy in the ADAM study was quite well tolerated. Seven out of 65 patients had to discontinue their study medication within the first 26 weeks of therapy. The incidence and severity of the observed side effects were comparable with those reported in other studies [15,16,27,28]. The most frequently reported side effect, diarrhoea, led to a discontinuation of the antiretroviral drug combination in only one patient. Although tolerable with the use of loperamide, diarrhoea was persistent in almost all patients. Furthermore, the occurrence of diarrhoea was not consistently associated with the exposure to nelfinavir or saquinavir, in contrast to other gastrointestinal complaints.

**Table 3.** Toxicity and plasma concentration ratios.

Side effect	Yes/No	n	Median plasma concentration ratio	
			NFV median (IQR)	SQV median (IQR)
Diarrhoea	Yes	41	0.59 (0.44–0.83)	0.61 (0.34–1.02)*
	No	15	0.62 (0.51–1.05)	1.19 (0.65–1.66)*
Nausea	Yes	11	0.59 (0.41–0.77)	0.78 (0.27–1.52)
	No	45	0.60 (0.45–0.87)	0.70 (0.43–1.36)
Abdominal distension	Yes	8	0.57 (0.43–0.81)	0.38 (0.29–0.85)
	No	48	0.60 (0.44–0.88)	0.78 (0.45–1.57)
Abdominal pain	Yes	8	0.95 (0.61–1.23)**	1.73 (0.83–2.70)**
	No	48	0.59 (0.44–0.83)**	0.68 (0.42–1.10)**
Headache	Yes	10	0.59 (0.44–0.90)	0.70 (0.45–1.36)
	No	46	0.60 (0.45–0.85)	0.75 (0.42–1.39)
Fatigue	Yes	17	0.58 (0.51–0.81)	0.78 (0.43–1.52)
	No	39	0.61 (0.41–0.89)	0.72 (0.89–1.36)
Elevated liver enzymes	Yes	9	0.60 (0.44–0.87)	0.78 (0.45–1.34)
	No	47	0.60 (0.45–0.85)	0.72 (0.42–1.39)
Elevated cholesterol	Yes	19	0.61 (0.41–0.95)	0.67 (0.28–1.39)
	No	36	0.60 (0.46–0.83)	0.75 (0.44–1.23)
Elevated triglycerides	Yes	9	0.94 (0.51–1.01)	0.67 (0.28–1.82)
	No	46	0.60 (0.44–0.83)	0.75 (0.42–1.34)

IQR, inter quartile range. \*  $P = 0.04$  (Wilcoxon test); \*\*  $P = 0.02$  for nelfinavir and  $P = 0.05$  for saquinavir (Wilcoxon test).



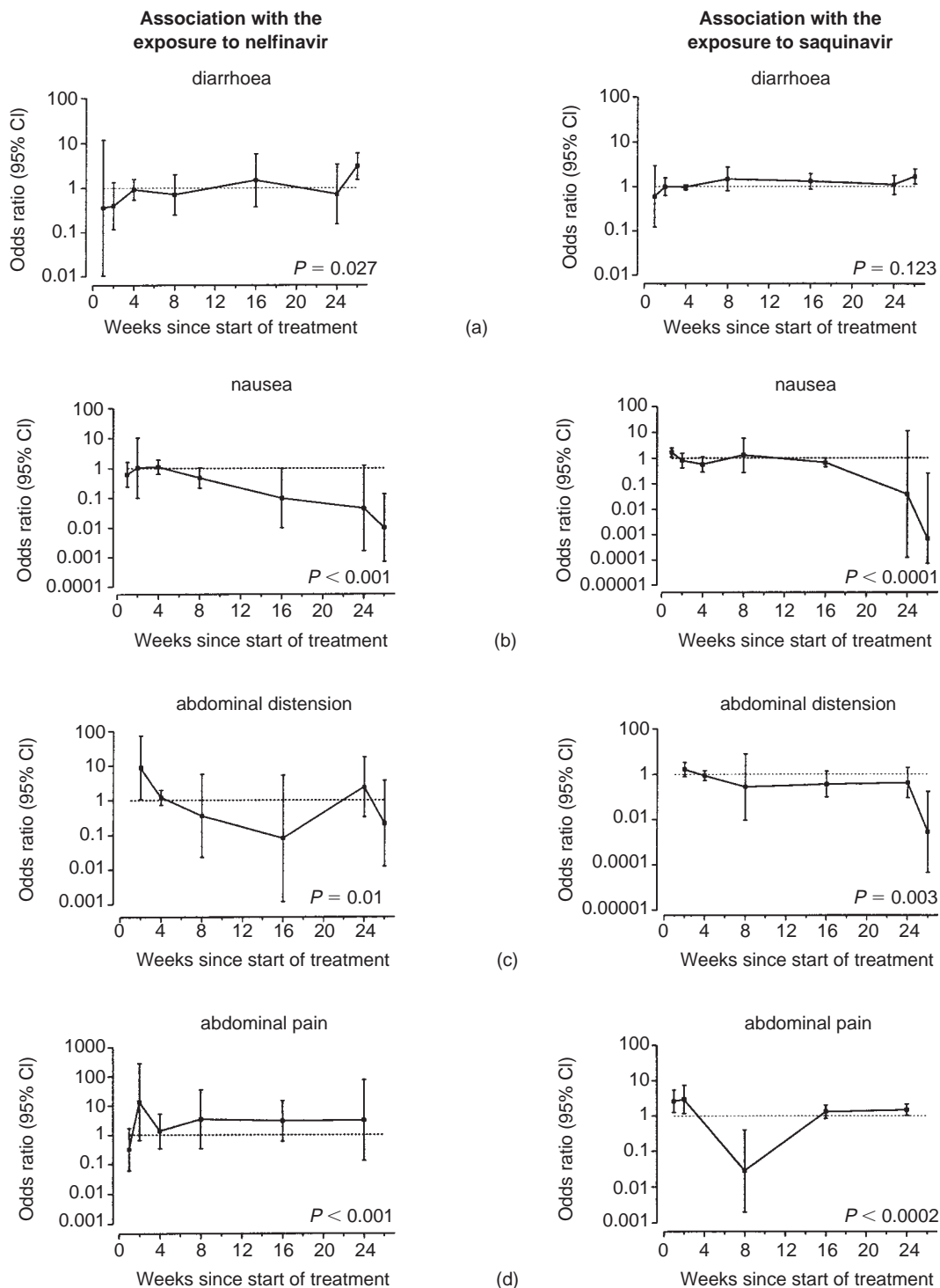
**Fig. 2.** Plasma concentration ratios of nelfinavir and saquinavir in patients with and without diarrhoea. Left side, nelfinavir; right side, saquinavir. Dots, patients with diarrhoea; circles, patients without diarrhoea.

Recently, many studies have focused on the association between protease inhibitor use and the occurrence of a new onset of diabetes mellitus, lipodystrophy or hyperlipidaemia [14]. The median time to the development of peripheral lipodystrophy was reported to be 10 months which might explain why this side effect was not observed within our population [14]. Hyperlipidaemia was present within the observed treatment period, although the elevations of triglycerides and cholesterol were usually mild and infrequent in comparison with those reported in studies using a combination of zidovudine and zalcitabine [17–19].

Surprisingly, the plasma concentrations of nelfinavir and saquinavir obtained were relatively low in comparison with the expected levels obtained under standardized conditions. These expected plasma drug concentrations, obtained in a clinical setting and after the use of a standard meal, probably resulted in higher plasma drug levels than those obtained in daily practice. This was found even though all the patients in this study were instructed to take their medication with food. Despite the relatively low exposure, virologic efficacy of the quadruple regimen was found to be satisfactory in the first 26 weeks. Only one patient failed to attain a plasma HIV-1 RNA concentration below 400 copies/ml. However, it remains to be seen whether these plasma drug concentrations are sufficient for durable suppression of the viral replication.

In contrast to the findings of Khaliq *et al.* the plasma concentration ratios of the protease inhibitors in the antiretroviral combination used in the ADAM study did not decline during the first 26 weeks [29]. However, by replacing saquinavir HGC with saquinavir SGC, the exposure to saquinavir within this population may have been maintained over time [30,31].

Figure 1 illustrates the variation of plasma concentration ratios of nelfinavir and saquinavir in patients over time. Considering this wide variation, the odds ratio for the occurrence of a specific side effect at a specific time point as a result of drug exposure was estimated. Most gastrointestinal complaints were found to have a time-dependent relationship with drug exposure. For both nausea and abdominal distension, the association with drug exposure varied within the first weeks of treatment. This change of association within the first weeks of treatment could indicate that the pharmacokinetics of the drugs and the tolerance of the patient had not yet reached an equilibrium. In addition, the use of stavudine and lamivudine might have concealed a possible relationship of nausea with the exposure to protease inhibitors. After the first weeks of therapy, however, abdominal distension and nausea were both associated with a low exposure to the protease inhibitors used. For nausea this association became even stronger over time. This could indicate a reduced uptake of the protease inhibitors in these patients, but



**Fig. 3.** The association between a certain side effect and exposure to nelfinavir (left column of figures) or saquinavir (right column of figures). In each figure, the dotted line represents an odds ratio of one, e.g. no association, whereas the solid line represents the odds ratio between a certain side effect and exposure to nelfinavir or saquinavir over time. Bars represent the 95% confidence interval. (a), diarrhoea; (b), nausea; (c), abdominal distension; (d), abdominal pain.

could also illustrate that patients with nausea may well have developed a less compliant behaviour, resulting in low plasma drug concentrations. Plasma drug monitoring in these patients could be useful when virologic efficacy appears insufficient.

The patients with abdominal pain were found to have both a high drug exposure as well as a greater decrease in HIV-1 RNA within the first 26 weeks. Hoetelmans *et al.* found that a higher clearance rate of HIV-1 RNA was found in patients with a higher exposure to

nelfinavir and saquinavir [9]. It may therefore be concluded that the elevation of drug exposure to nelfinavir and saquinavir in order to increase virologic effectiveness might be hampered by the occurrence of abdominal pain.

In dose-finding studies, diarrhoea appeared to be the dose-limiting side effect of nelfinavir [32,33], while a higher bio-availability of saquinavir was more likely to cause diarrhoea, suggesting a relationship between the exposure to a PI and the occurrence of this side effect [31,34]. The patients with diarrhoea in our study were found to have lower levels of saquinavir than patients without diarrhoea (Wilcoxon test). This was however not confirmed using the GEE method. In fact, using this more sensitive method, the occurrence of diarrhoea was not related to the exposure to saquinavir, but had an inconsistent association with the exposure to nelfinavir. It is possible that the occurrence of diarrhoea has been both a cause and an outcome of drug exposure, which might have concealed a significant association.

None of the other clinical or laboratory abnormalities could be related to the observed drug exposure. The occurrence of fatigue and headache might be explained by the (continuous) use of nucleoside reverse transcriptase inhibitors as well. In addition, fatigue is a side effect that is more likely to occur in patients with a more advanced disease stage. This was confirmed in our study since patients experiencing fatigue had a significantly lower CD4+ cell count and a higher HIV-1 RNA concentration in plasma at baseline than patients without this complaint.

In conclusion, the quadruple induction therapy was quite well tolerated. Diarrhoea was the most frequently reported side effect. During the first 26 weeks of therapy, the elevations of plasma triglycerides and cholesterol were mild and infrequent in comparison with those observed in other PI combinations. Clear and time-dependent relationships with the amount of drug exposure were only found for nausea, abdominal distension and abdominal pain. The exposure to the protease inhibitors in this population was lower than expected, suggesting that the compliance to timing, dosing and food intake required may not be achieved in daily practice. Nevertheless, the virologic efficacy of the quadruple drug regimen was quite satisfactory.

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## Appendix 1

Side effect	Grade I	Grade II	Grade III	Grade IV
Nausea	Mild or transient; reasonable intake maintained.	Moderate discomfort or intake decreased for < 3 days.	Severe discomfort or minimal intake for $\geq$ 3 days.	Hospitalization required.
Abdominal pain	Transient or mild discomfort; no limitation in activity; no medical intervention/therapy required.	Mild to moderate limitation in activity; some assistance may be needed: no or minimal medical intervention/therapy required.	Marked limitation in activity; some assistance usually required: medical intervention/therapy required, hospitalizations possible.	Extreme limitation in activity; significant assistance required; significant medical intervention/therapy required, hospitalization or hospice care probable.
Abdominal distension	Transient or mild discomfort no limitation in activity; no medical intervention/therapy required.	Mild to moderate limitation in activity; some assistance may be needed: no or minimal medical intervention/therapy required.	Marked limitation in activity; some assistance usually required: medical intervention/therapy required, hospitalizations possible.	Extreme limitation in activity; significant assistance required; significant medical intervention/therapy required, hospitalization or hospice care probable.
Diarrhoea	Mild or transient: two to three episodes/day or mild diarrhoea lasting < 1 week.	Moderate or persistent: five to seven loose stools/day or diarrhoea lasting $\geq$ 1 week.	Bloody diarrhoea or orthostatic hypotension or > 7 loose stools/day or IV therapy required.	Hypotensive shock or hospitalization required.
Fatigue	Normal activity reduced < 25%.	Normal activity reduced 25–50%.	Normal activity reduced > 50% cannot work.	Unable to care for himself/herself.
Headache	Mild: no medication required.	Moderate: non-narcotic analgesia therapy required.	Severe: responds to initial narcotic therapy.	Intractable: requiring repeated narcotic therapy.
Elevated liver enzymes	1.25 to 2.5 $\times$ ULN of AST, ALT, AF or $\gamma$ -GT.	> 2.5 to 5.0 $\times$ ULN of AST, ALT, AF or $\gamma$ -GT.	> 5.0 to 10.0 $\times$ ULN of AST, ALT, AF or $\gamma$ -GT.	> 10.0 $\times$ ULN of AST, ALT, AF or $\gamma$ -GT.
Elevated triglycerides	–	4.5–8.4 mmol/l or 400–750 mg/dl.	8.5–13.5 mmol/l or 751–1200 mg/dl.	> 13.5 mmol/l or > 1200 mg/dl.
Elevated cholesterol <sup>a</sup>	–	6.2–7.7 mmol/l or 240–300 mg/dl.	7.8–10.3 mmol/l or 301–400 mg/dl.	> 10.3 mmol/l or > 400 mg/dl.

<sup>a</sup>ACTG, guidelines for toxicity 1997. ULN, upper limit of normal.