Virological and immunological response in HIV-1-infected patients with multiple treatment failures receiving raltegravir and optimized background therapy, ANRS CO3 Aquitaine Cohort.

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# The first two authors contributed equally to this study

Abstract

Background

The efficacy of raltegravir plus optimized background therapy (OBT) has been demonstrated for antiretroviral (ARV)-experienced HIV-1 infected patients in randomized clinical trials. We studied viro-immunological response, pharmacokinetic parameters, and genotypic test results in an observational cohort of multiple ARV class-experienced patients starting a raltegravir-based regimen.

Methods

Already enrolled ANRS CO3 Aquitaine Cohort patients with virologic failure were included in this study after starting a raltegravir-based regimen (400 mg twice a day, week 0). Virologic success was defined by plasma HIV-1 RNA level [viral load (VL)] <2.7 $\log_{10}$ copies/mL at week 12 and <1.7 $\log_{10}$ copies/mL at week 24. One patient was excluded from further analysis (no follow-up after week 4).

Results

Fifty-one patients (male/female = 43/8, median age = 48 [interquartile range = 43; 55] years) were included. At week 0, median CD4 count was 244 [110; 310]/mm^3 and median VL was 4.2 [3.6; 4.7] $\log_{10}$ copies/mL. At week 24, 39 (78%) patients experienced virologic success: 4 (44%), 14 (82%) and 21 (87%) of patients with a genotypic sensitivity score <1, [1–2[and ≥2 (P =0.02), respectively. Raltegravir-related mutations emerged in 9 of 11 failing patients (82%): Q148H/R (n=5), N155S/H (n=3) and S230N (n=1). Median CD4 rise from week 0 to week 4 and week 24 were 28 [−4; 85] and 57 [0; 156] cells/mm^3 , respectively. A poor immune response was independently associated with a lower VL decline (week 0 to week 12) [odds ratio (OR): 3.5 95% confidence interval (CI): 1.4; 8.4 for 1 $\log_{10}$ less] and CD4+% at baseline (OR: 2.6, 95% CI: 0.97; 8.3 for 10% lower).

Conclusions

Raltegravir plus OBT provided a good virologic success rate in highly pre-treated patients under clinical routine conditions.

MESH Keywords Adult ; Anti-HIV Agents ; pharmacokinetics ; therapeutic use ; Antiretroviral Therapy, Highly Active ; CD4 Lymphocyte Count ; Cohort Studies ; Drug Resistance, Viral ; Female ; HIV Infections ; drug therapy ; immunology ; virology ; HIV-1 ; classification ; genetics ; isolation & purification ; Humans ; Male ; Middle Aged ; Pyrrolidinones ; pharmacokinetics ; therapeutic use ; Treatment Outcome ; Viral Load

Author Keywords Raltegravir ; antiretroviral treatment experienced patients ; viro-immunological response ; integrase resistance mutations

Introduction

Raltegravir is a HIV-1 integrase inhibitor which has been successfully used in both treatment-naïve patients 1 and heavily treated patients 2 ,3 .

We studied the viro-immunological response of raltegravir with optimised background therapy (OBT) in an observational prospective cohort of multiple antiretroviral class experienced patients under clinical routine conditions. Furthermore, we evaluated pharmacokinetic (PK) parameters, genotypic test results of integrase, reverse transcriptase and protease genes at baseline and in failing patients.

Patients and Methods

Study population

The patients were selected from the ANRS Co3 Aquitaine Cohort, a prospective hospital-based cohort of HIV-1 infected patients in south-western France. Informed consent was obtained for all patients. The Aquitaine Cohort has an Institutional Review Board (IRB) approval from the Bordeaux University IRB.

Patients who experienced multiple virological failure on highly active antiretroviral therapy (HAART) {plasma HIV-1 RNA level viral load (VL) >1.7 $\log_{10}$ copies/ml} were consecutively enrolled in the present study between October 2006 and February 2008 after onset of raltegravir (400 mg twice a day)-based HAART (week 0). The patients were monitored at weeks 0, 4, 12 and 24. Clinical, biological and therapeutic data were collected prospectively at each visit.

Virological and immunological outcomes

Virological success was defined as plasma VL <2.7 $\log_{10}$ copies/mL at week 12 and <1.7 $\log_{10}$ copies/mL at week 24, quantified using the CobasTaqman HIV assay (Roche Diagnostics, Basel, Switzerland). Patients with missing VL values at week 24 were considered as virologic success if the VL was <2.7 $\log_{10}$ copies/mL at week 12. One patient had no follow up visit after week 4 and was excluded from further analysis. A poor immune response was defined as a gain of $CD4^+$ cells ≤50 cells/mm^3 from week 0 to week 24.

Genotype-resistance testing

Sequencing procedures used for reverse transcriptase, protease and integrase are available on the HIV French resistance website4 Complete integrase gene sequence was determined at baseline, and in patients with virologic failure. We calculated the genotypic sensitivity score (GSS) that represents the sum of genotypic sensitivities (according to the ANRS genotype-interpretation algorithm) to the drugs in the OBT.

Determination of plasma Raltegravir concentrations

Blood samples were drawn to determine plasma raltegravir concentrations at the PK steady-state 4 weeks after starting raltegravir as well as at week 12 and week 24. Minimum (C_{min}) and maximum (C_{max}) serum drug concentrations, corresponding to around 12 h and 3 h after raltegravir ingestion, respectively, were measured using a validated HPLC with mass spectrometry detection5 .

Statistical analyses

Analyses were performed using SAS 9.1 (SAS Institute, Inc., Cary, NC). Patients' characteristics were compared between groups using a Fisher's exact test for qualitative variables, and a Wilcoxon-Mann-Whithney test or a Kruskal-Wallis test for quantitative variables. Distributions are described as medians (25^{th} ; 75^{th} percentiles), unless stated otherwise. We tested the following variables for their association with virological failure: patients' characteristics, prior treatments, baseline viro-immunological parameters, number of PI, nucleoside reverse transcriptase inhibitor (NRTI), NNRTI resistance-related mutations, GSS, PK parameters and integrase polymorphisms (PMs) having a prevalence >10%. Factors associated with virological success and with a poor immune response were analysed using logistic regression.

Results

Baseline patient characteristics

The baseline characteristics of the 51 patients enrolled in this observational cohort study are reported in table 1 . Most of the patients (72%) received raltegravir as part of an expanded access program in France (Autorisation Temporaire d'Utilisation). ARV drugs frequently prescribed as OBT were ritonavir-boosted darunavir (n=36, 71%), etravirine (n=22, 43%) and enfurvitide (n=13, 28%). Raltegravir in combination with ritonavir-boosted darunavir and etravirine was prescribed in 15 patients (29%). The most frequently NRTIs co-prescribed with raltegravir were tenofovir (n=20, 39%), emtricitabine (n=18, 35%) or lamivudine (n=11, 22%).

Responses to the raltegravir containing therapy

Virological response

Virologic success was observed for 39 patients (78%) (Table 2). According to a GSS of <1, [1–2[and ≥2 virologic success occurred in 4 (44%), 14 (82%) and 21 (88%) patients (P =0.02) with a VL decline (from baseline to week 24) of −0.64 (−2.8; −0.3), −2.4 (−3.1; −1.1)

and −2.2 (−2.9; −1.6) $\log_{10}$ copies/mL (P =0.27), respectively. Among the 26 Pms having a prevalence >10% from the baseline genotype the PM T206S (n=8, 18%) was significantly associated with a lower response rate (P =0.02). Furthermore, the clinical AIDS stage [odds ratio (OR):0.2, 95% confidence interval (CI): 0.05; 0.80 C vs. A/B, P =0.02), the nadir of CD4+ cell count (OR: 1.6, 95% CI: 1.1; 2.1 for a difference of 10 cells/mm^3 , P =0.006), the absolute value of CD4+ cell count at baseline (OR: 5.9, 95% CI: 1.9; 18.1 for a difference of 100 cells/mm^3 , P =0.002) and the HIV-1 RNA level at baseline (OR: 0.1, 95% CI: 0.04; 0.5 for a difference of 1 $\log_{10}$ copies/mL, P = 0.002) were significantly associated with virologic response in univariable analysis. In the adjusted analysis initial VL (OR: 0.2, 95% CI: 0.06; 0.98 for 1 $\log_{10}$ copies/mL higher, P =0.046) and the nadir of $CD4^+$ cell count (OR: 1.6, 95% CI: 1.0; 2.7 for 10 cells/mm^3 higher, P = 0.049) were independently associated with virological response.

Immune response

A $CD4^+$ gain (baseline to week 24) ≤50 cells/mm^3 was found in 20 patients. In adjusted logistic regression, a poor immune response was independently associated with a lower VL decline (W0week 0–12) (OR: 3.5 95% CI: 1.4; 8.4 for 1 $\log_{10}$ less, P=0.009) and CD4+% at baseline (OR: 2.6 95% CI: 0.97; 8.3 for 10% lower, P=0.08), but did not explain the whole variability of $CD4^+$ response (R^2 =0.29). At week 24, 32 patients had a VL <1.7 $\log_{10}$ copies/mL (one patient with missing values for $CD4^+$ cells) and for 12 of them we observed a $CD4^+$ gain of ≤50 cells/mm^3 (38%, 95% CI: 22%, 56%). These patients had similar baseline CD4+ cell counts (249 versus 246/mm^3), almost similar nadir of $CD4^+$ cell count (146 versus 117/mm^3) and lower VL at week 0 (3.4 versus 4.3 $\log_{10}$ cp/mL) than patients with both virologic and immunological success (n=20).

Pharmacokinetic parameters

Raltegravir PK parameters were stable during follow-up. Median C_{min} was 250 (150; 350), 300 (200; 350) and 290 (150; 350) ng/mL at weeks 4, weeks 12 and weeks 24, respectively. Median C_{max} was 1000 (870; 1400) ng/mL at week 4, 990 (800; 1200) ng/mL at week 12 and 980 (780; 1200) ng/mL at week 24. The minimal observed value of C_{min} was 50 ng/mL (0.10 μM) for each follow-up visit exceeding the 95% inhibitory concentration (IC_{95}) of 0.033 μM. Patients with etravirine in the OBT had slightly lower PK parameters but there was no statistical significant interaction of etravirine on raltegravir PK parameters.

Emerging integrase mutations

Four different patterns of emerging mutations were observed: i) five patients presented the emergence of Q148H/R with secondary mutations (V72I, L74M, G140A/S, E138A, K156N, K160N, V201I and T206S), ii) the N155S/H mutation emerged in three patients and was replaced in the following three to five month by a pattern including the mutation Y143C/H/R and secondary mutations (L74M, T97A, G163R, V151I, S230R), iii) the S230R mutation was selected in one patient and iv) two patients had virologic failure without emerging mutations.

Discussion

We observed a potent antiretroviral effect in patients failing multiple previous antiretroviral regimens before. Our findings were comparable with virological success rates observed in similar populations with heavily treatment-experienced patients 2 ,3 on raltegravir-based HAART. Despite a rapid VL suppression, the overall median $CD4^+$ cell rise was 57 (0; 156)/mm^3 between baseline and week 24 comparable to that observed in the P005 and Benchmrk studies. A poor immune response was associated with VL decline and $CD4^+$ % at baseline, but explained only around 30% of the entire variability of the $CD4^+$ response. We found a discordant response (VL <1.7 $\log_{10}$ copies/mL and $CD4^+$ gain ≤50 cells/mm^3) in 38% of patients at week 24. Further investigation is needed to evaluate other hypotheses such as permanent immune activation, host factors or thymus exhaustion for a poor immune response despite virological success.

Factors associated with virological response at week 24 were the VL at baseline and the nadir of $CD4^+$ cell counts. These findings are in agreement with the fact that most of the patients were already in an advanced disease stage.

The high proportion of integrase resistance mutations that developed in patients who failed therapy (9/11, 82%) in this study was consistent with findings in Benchmrk studies (68%)3 , 6 and the protocol 005 study (92%)7 . Our findings confirm the low genetic barrier of raltegravir. The low genetic barrier may have an influence on future drug options especially in comparable patients, as cross-resistance to elvitegravir and other integrase inhibitors under investigation have already been reported 8 .

PK parameters did not provide a statistically meaningful predictive value for virological success, probably due to the fact that observed C_{min} values were quite homogeneous and exceeded the IC_{95} of raltegravir in all patients. Etravirine co-prescription did not influence raltegravir PK parameters, confirming the negligible PK interaction between etravirine and raltegravir observed in healthy subjects10 .

Conclusion

Raltegravir plus OBT provided a good virological success rate in HIV-1-infected heavily pre-treated patients with multiple treatment failures under clinical routine conditions comparable to that reported in randomized clinical trials.

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Appendix

The Groupe d'Epidemiologie Clinique du Sida en Aquitaine (GECSA) steering the ANRS CO3 Aquitaine Cohort is organized as follows:

Scientific committee: F. Dabis (Chair and Principal Investigator), M. Dupon, M. Longy-Boursier, P. Morlat, JL. Pellegrin, and JM. Ragnaud.

Epidemiology, Methodology: M. Bruyand, G. Chêne, F. Dabis, S. Lawson-Ayayi, R. Thiébaut.

Infectious diseases, Internal Medicine: M. Bonarek, F. Bonnal, F. Bonnet, N. Bernard, O. Caubet, L. Caunègre, C. Cazanave, J. Ceccaldi, FA Dauchy, C. De La Taille, S. De Witte, M. Dupon, P. Duffau, H. Dutronc, S. Farbos, MC Gemain, C. Greib, D. Lacoste, S. Lafarie-Castet, P. Loste, D. Malvy, P. Mercié, P. Morlat, D. Neau, A. Ochoa, JL. Pellegrin, JM. Ragnaud, S. Tchamgoué, JF. Viallard.

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Virology: H. Fleury, ME. Lafon, B. Masquelier.

Pharmacology: D. Breilh.

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Data collection: MJ. Blaizeau, M. Decoin, S. Delveaux, S. Gillet, C. Hannapier, O. Leleux, B. Uwamaliya-Nziyumvira.

Data management: S. Geffard, G. Palmer, D. Touchard.

Footnotes:

‡ ANRS CO3 Aquitaine Cohort (see Appendix).

Transparency declaration None to declare.

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Table 1

Baseline characteristics of 51 patients receiving raltegravir-based HAART. ANRS CO3 Aquitaine Cohort, 2007–2008.

Characteristic	Value
Sex (M/F), n (%)	43/8 (84/16)
Age, years	48 (43; 55)
Clinical CDC stage (A/B/C), n (%)	10/24/17 (20/47/33)
Follow up time, month	8(6; 13)
Drugs in the OBT	3 (2; 4)
HIV-1 infection duration, years since diagnosis	17 (14; 20)
HIV-1 RNA zenith, $\log_{10}$ copies/mL	5.4 (4.8; 5.8)
$CD4^+$ nadir, cells/mm^3	90 (33; 175)
Baseline HIV-1 RNA level, $\log_{10}$ copies/mL	4.2 (3.6; 4.7)
Baseline $CD4^+$ count, cells/mm^3	244 (110; 310)
Previous antiretroviral therapy	
Duration of exposure, years	10 (6; 12)
Previous NRTI, nb	6 (5; 7)
Previous PI, nb	5 (4; 6)
Previous NNRTI, nb	2(1; 2)
Raltegravir co-prescribed antiretrovirals, n (%)	
+ 2 NRTIs	18 (35)
+ 1 PI	40 (80)
+ 1 NNRTI	22 (43)
Genotype characteristics at baseline	
NRTI resistance-related mutations	5 (4; 6)
PI resistance-related mutations	
Minor[a]	9 (8; 11)
Major[a]	4 (3; 5)
Total (minor + major)	13 (11; 15)
NNRTI resistance-related mutations	2(1; 3)
GSS ANRS (<1/≥1&<2/≥2)	9/17/25(18/33/49)

Baseline: Initiation of raltegravir-based HAART. Values are medians (IQR) unless stated otherwise. NRTI: nucleoside reverse-transcriptase inhibitor; NNRTI: non-nucleoside reverse-transcriptase inhibitor; PI: protease inhibitor; nb: numbers; OBT: Optimised background therapy;

a Major and minor protease mutations were defined according to the IAS–USA panel[9] ;

b GSS: Genotypic sensitivity score of optimized background therapy according to the ANRS algorithms.

Table 2

Viro-immunological response in patients receiving raltegravir-based HAART, ANRS CO3 Aquitaine Cohort, 2007–2008.

	Follow-up		
	W4 (n=43)	**W12 (n=50)**	**W24 (n=45)**
<1.7 log_{10} copies/mL			
n	26	27	33
%	60	54	73
95% CI	44; 75	39; 68	58; 85
<2.6 log_{10} copies/mL			
n	36	42	35
%	84	84	78
95% CI	69; 93	71; 93	63; 89
Delta HIV-1 RNA log_{10} copies/mL			
median	−2.1	−2.1	−2.1
IQR	−2.6; −2.1	−2.9; −1.2	−2.9; −1.1
Virological success *			
n			39
%			78
95% CI			64; 88
CD4+ cells/mm^3			
median	255	298	286
IQR	165; 376	176; 396	164; 400
Delta $CD4^+$ cells/mm^3			
median	28	40	57
IQR	−4; 85	2; 95	0; 156

CI: confidence interval; IQR: Interquartil range; W4: week 4; W12: week 12; W24: week 24; Delta CD4+: difference of CD4+ cell counts between W4, W12, W24 and baseline, respectively; Delta HIV-1 RNA: difference of HIV-1 RNA between baseline and W4, W12 and W24, respectively.

* HIV-1 RNA level less than 2.7 log_{10} copies/mL at W12 and HIV-1 RNA level less than 1.7 log_{10} copies/mL at W24, patients with missing data at W24 and HIV-1 RNA level less than 2.7 log_{10} copies/mL at W12 were considered with virologic success; One patient had neither a HIV-1 RNA value at W12 nor at W24.