Hyperlipidemia Related to the Use of HIV-Protease Inhibitors: Natural History and Results of Treatment with Fenofibrate

Bruno Caramelli, Claudia Y.S.M. de Bernoche, Ana M. C. Sartori, Andrei C. Sposito, Raul D. Santos, Maristela C. Monachini, Tania Strabelli and Davi Uip

Heart Institute (InCor) and AIDS Clinic, University of São Paulo Medical School, São Paulo, Brazil

Hyperlipidemia has been frequently recorded as a side effect of treating HIV patients with protease inhibitors (PI). This study was initiated to analyze the modifications on blood lipids in HIV-patients receiving PI and the safety and efficacy of the treatment with fenofibrate. Total (TC) and HDL-cholesterol, triglycerides (TG), and CD_4^+ T-cell counts were measured in 30 HAART-naive patients (Group I) before and after PI introduction. In a second phase of the study, the effects of fenofibrate on lipids, CPK, CD_4^+, and viral load were determined in 13 patients (Group II) with elevated TC or TG. In Group I, 60% of the patients showed TC or TG elevations. Average increments of 31% and 146% in TC and TG respectively ($p<0.0006$ and $p<0.0001$) were observed. In Group II, fenofibrate treatment was associated with decrements of 6.6% (TC) and 45.7% (TG) ($p=0.07$ and 0.0002) and no modifications on CPK, CD_4^+, and viral load. In conclusion, hyperlipidemia is common during the treatment of HIV with protease inhibitors, and fenofibrate appears to be an effective and safe choice for its treatment.

Key Words: Cholesterol, lipids, HIV infection, triglycerides.

HIV-protease inhibitors (PI) are now widely used for the treatment of HIV-positive patients [1]. A reduction in morbidity as well as an increase in life expectancy was obtained in patients treated with the highly active antiretroviral therapy (HAART) [1]. However, side effects have been described, associated with the use of PI [1-9]. Hyperlipidemia and particularly severe hypertriglyceridemia have been observed in patients taking protease inhibitors although a causal relation between lipid abnormalities and PI has not been well established [2-9]. This is an important issue, considering that maintaining the levels of blood cholesterol and triglycerides bellow high-risk levels is an important goal for primary prevention of cardiovascular diseases [10-11].

The natural history of hyperlipidemia in HAART-naive patients has not been established in prospective clinical trials. The incidence, prevalence, time interval to develop lipid alterations after PI introduction, and the diagnosis of the type of hyperlipidemia remain undetermined. Moreover, once the diagnosis is established, pharmacological treatment for this condition is controversial due to the risk of skeletal muscle toxicity and potential efficacy reduction related to metabolic induction leading to lower plasmatic levels of PI and virologic failure [7-8].

The greater effect on elevated levels of LDL-cholesterol than older generation fibrates, associated to the reduction of plasma triglycerides makes fenofibrate a monotherapeutic option for HIV-protease inhibitors related hyperlipidemia.[12-13]. However, data on its efficacy and safety in this clinical setting are lacking.

Received on 3 October 2001; revised 18 December 2001.
Address for correspondence: Dr. Bruno Caramelli, Heart Institute (InCor), Unidade de Medicina Interdisciplinar em Cardiologia. Av. Dr. Enéas C. de Aguiar 44, São Paulo, Brazil, Zip Code: 05403-900. E-mail: bcaramel@usp.br. Fax: (5511) 3069-5535. Phone: (5511) 3069-5376.

The Brazilian Journal of Infectious Diseases 2001;5(6):332-338.

1413-8670

This study aimed to prospectively determine the modifications on blood lipids after the introduction of the therapy with HIV-protease inhibitors and to establish the efficacy and safety of the pharmacological treatment with fenofibrate in HIV-positive patients.

Population and study design

In an earlier study published elsewhere, we analyzed the lipid profile of HIV-positive patients and compared it to age and sex matched volunteers [14]. That study was initiated in 1994, before the introduction of HIV-protease inhibitors. After the introduction of these drugs, we observed patients with high levels of total cholesterol (TC) and triglycerides (TG) as well as other authors [3-6]. From this greater group, we retrospectively studied the prospective collected data regarding blood lipid levels, in order to compare data before and after PI introduction. This comprised the first phase of our study and the population is identified as Group I.

In the second phase of our study (Group II) 13 patients taking PI and showing levels of TC or TG greater than 200 mg/dL were followed during treatment with fenofibrate. This population was included in an open, non-random and non placebo controlled study of patients who received dietary treatment plus fenofibrate. Except for 3 patients, Groups I and II did not represent the same population.

Patients showing low compliance of medication, those who used or had previous use of any drug that decreases blood lipid levels, those with liver or renal disease, or diabetes mellitus were excluded after diagnosis by history, treatment, or laboratory analysis.

The study protocol was approved by the ethical committee on human experimentation from the Hospital das Clínicas da Universidade de São Paulo.

Methods

Blood lipids: Fasting blood samples were obtained from patients before (baseline sample) and at least one month after the start of HIV-protease inhibitor (PI sample) in Group I and before and after the introduction of fenofibrate treatment in Group II. The concentrations of TC, TG and HDL-cholesterol were obtained through enzymatic colorimetric methods.

Patients were classified as having one of the predefined types of hyperlipidemia: isolated hypercholesterolemia (TC > 200 mg/dL and TG < 200 mg/dL), isolated hypertriglyceridemia (TC > 200 mg/dL and TG < 200 mg/dL) or combined hyperlipidemia (TC and TG > 200 mg/dL).

Treatment for hyperlipidemia: For the purpose of this study, levels of TC and/or TG > 200 mg/dL associated to the use of HIV-protease inhibitors were considered elevated and patients (Group II) received dietary recommendations according to the National Cholesterol Education Program (NCEP) guidelines plus micronized fenofibrate 200 mg daily [11].

Efficacy and safety of pharmacological treatment: Efficacy of the treatment was determined by the capacity of diet plus fenofibrate to reduce lipids to desired levels (TC < 200 mg/dL and TG < 200 mg/dL). Safety was evaluated by the determination of creatine kinase (CK) levels (enzymatic ultraviolet method), CD_4^+ T-cell counts and plasma HIV-1 RNA load (viral load) during drug treatment for PI related hyperlipidemia. Liver enzymes levels were also obtained for all patients included in Group II, after treatment with fenofibrate.

Plasma HIV-1 RNA determination: Viral load was measured in all Group II patients by NASBA (Organon Technika). The number of HIV-1 RNA copies was calculated on the basis of the manufacturer's reference standards.

PI regimen: The HIV-protease inhibitor regimen considered was the one taken at least one month before the time of PI sample analysis for Group I and, for Group II, the drugs taken at the time of fenofibrate introduction.

Statistical analysis

Baseline values were compared to data obtained after treatment for Groups I and II. For weight, total and HDL-cholesterol comparisons, two-tailed student t-test was adopted. For triglycerides and CD_4^+ T-cell counts, two-tailed Wilcoxon signed-rank test was used. Logarithmic transformation was adopted for plasma HIV-1 RNA load values and summarized by use of means and standard deviations. Statistical analysis was then performed by two-tailed Wilcoxon signed-rank test.

When appropriate, the frequencies of each type of hyperlipidemia before and after treatment were compared by Fisher's exact test. The level of significance accepted for all tests was 0.05.

Results

Group I

Clinical Characteristics. Thirty patients (24 men and 6 women) were included in Group I. The mean age was 39.6 years old and ranged from 23 to 67 years old. Twenty-one patients (70%) were taking indinavir, 6 (20%) were on HCG-saquinavir, 2 (6.6%) on ritonavir, and 1 (3.3%) on ritonavir plus HCG-saquinavir as the HIV-protease inhibitor regimen considered. The average time interval between baseline and the sample obtained after PI introduction was 203 days.

No significant modifications were observed on weight after the introduction of PI (72.5 ± 16.9 to 72.3 ± 16.0 kg). Regarding CD_4^+ T-cell counts, however, there was an increment related to the introduction of PI (159 ± 155 to 345 ± 247 cells/μL, $p < 0.0001$) (Table 1).

Blood lipids. Treatment with PI was associated with average increments of 31% for TC and 146% for TG. No significant modifications were observed in HDL-cholesterol levels (Table 1).

At inclusion, there were 23% of the patients had increased levels of TC, 26% with increased TG, and 36% needed treatment for any lipid alteration. After the introduction of PI, 43% of the patients showed elevated TC, 53% elevated TG ($p=0.17$ and $p=0.06$ respectively), and 60% had to be treated for any lipid alteration ($p=0.12$).

Regarding the type of hyperlipidemia, before treatment, isolated hypercholesterolemia, hypertriglyceridemia and combined hyperlipidemia corresponded to 10%, 13%, and 13 % of the patients respectively. After treatment, prevalences were 6%, 16%, and 36%, respectively ($p= 0.07$ for combined hyperlipidemia).

Group II

Clinical characteristics. Thirteen patients (11 men and 2 women) were included in Group II. The mean age was 43 (31-66) years old. Five patients (38.4%) were taking ritonavir plus HCG-saquinavir, 4 (30.7%) were on indinavir, 2 (15.3%) on ritonavir, and 2 (15.3%) on nelfinavir as the HIV-protease inhibitor regimen considered.

No significant modifications were observed on weight, CD_4^+ T-cell counts, and viral load in Group II, after the introduction of fenofibrate (Table 2). For all patients, CK and liver enzymes levels remained within the normal range.

Blood lipids. After an average time of 36 days (11 to 103) of treatment with fenofibrate, we observed an average decrement of 6.66% for TC and 45.70% for TG. For HDL-cholesterol, an average increment of 21.30 % was observed (Table 3).

There was a non-significant decrease on the prevalence of elevated levels of TC (84% to 69%, $p=0.64$) and a significant decrement on the prevalence of elevated levels of TG (100% to 61%, $p=0.03$). After treatment, 76 % of the patients still did not show desired levels of TC and TG. The prevalences of the types of hyperlipidemia before fenofibrate treatment were 0%, 15%, and 84% for hypercholesterolemia, hypertriglyceridemia, and combined hyperlipidemia respectively. After treatment, prevalences were, respectively, 15%, 7%, and 53% (non significant changes).

Table 1. Group I. Weight, CD_4^+ T-cell counts, total cholesterol, triglycerides and HDL-cholesterol before and after treatment, and average increment related to the use of protease inhibitors

Variable	Baseline	PI	Average increment	P*
Weight (kg)	72.50 ± 16.92	72.37 ± 16.07	-	0.92
CD_4 (cells/μL)	159 ± 155	345 ± 247	116%	<0.0001
Total-Cholesterol	164 ± 43	204 ± 63	31%	<0.0006
Triglycerides	131 (61-650)	216 (80-1730)	146%	<0.0001
HDL-Cholesterol	33 ± 9	33 ± 8	-	0.95

CD_4 = CD_4^+ T-cell count. Data are expressed in mean values ± standard deviation (median and range for triglycerides). Baseline = values before treatment; PI = values after treatment with protease inhibitor. Total and HDL-cholesterol, triglycerides are expressed in mg/dL.

* Two tailed student t-test was adopted for total and HDL-cholesterol comparisons. For triglycerides, two-tailed Wilcoxon signed-rank test was used.

Table 2. Group II. Weight, CD_4^+ T-cell counts , and plasma viral load before and after fenofibrate treatment for hyperlipidemia related to the use of protease inhibitors

Variable	Before	After	P*
Weight (kg)	69.09 ± 9.95	68.10 ± 10.01	0.16
CD_4 (cells/μL)	389 ± 156	412 ± 186	0.49
Viral load	2.64 ± 1.01	2.84 ± 1.25	0.49

Data are expressed in mean values ± standard deviation (after logarithmic transformation for viral load). CD_4 = CD_4^+ T-cell count.

* Two tailed student t-test was adopted for weight comparison. For CD_4^+ T-cell counts and plasma HIV-1 RNA load values (after logarithmic transformation) , two-tailed Wilcoxon signed-rank test was used.

Table 3. Group II. Total cholesterol, triglycerides and HDL-cholesterol, and average modification before and after fenofibrate treatment for dyslipidemia related to the use of protease inhibitors

Variable	Before	After	Average modification	P*
Total-Cholesterol	246 ± 61	226 ± 53	- 6.66%	0.07
Triglycerides	486 (314-720)	274 (64-478)	- 45.7%	0.0002
HDL-Cholesterol	32 ± 10	38 ± 12	+ 21.3%	<0.0001

Data are expressed in mean values ± standard deviation (median and range for triglycerides) and in mg/dL.

* Two tailed student t-test was adopted for total and HDL-cholesterol comparisons. For triglycerides, two-tailed Wilcoxon signed-rank test was used.

Table 4. Group II. Age, total cholesterol, triglycerides, HDL-cholesterol, viral load, CD_4^+ T-cell count, and the duration of treatment before and after treatment with fenofibrate

Patient	Age	TC 1	TG1	HDL1	Load 1	$CD_4$1	Time	TC 2	TG 2	HDL 2	Load 2	$CD_4$2
1	66	320	720	39	2.66	183	11	323	444	42	2.46	328
2	38	135	516	26	1.90	447	28	146	175	30	5.30	308
3	45	350	454	56	1.90	657	41	294	193	68	1.90	812
4	39	236	599	20	4.43	357	25	301	478	36	1.90	312
5	48	177	333	30	1.90	546	32	156	133	35	1.90	738
6	35	203	328	26	3.27	218	103	191	157	30	3.49	171
7	33	204	598	26	1.90	248	28	185	331	34	1.90	503
8	56	293	484	31	1.90	633	20	235	346	29	3.14	319
9	45	226	424	37	1.90	343	28	234	245	39	2.11	410
10	45	226	386	24	1.90	228	35	205	255	42	1.90	308
11	31	241	314	50	4.11	332	27	210	64	59	4.00	262
12	38	306	609	34	4.30	484	38	238	340	36	5.11	520
13	42	287	564	26	2.30	392	64	231	409	24	1.90	366

TC = total cholesterol; TG = triglycerides; HDL= HDL-cholesterol; Load=Viral load; CD_4= CD_4^+ T-cell counts; 1=before treatment data; 2=after treatment data; Time=time interval between 1 and 2. Data are expressed in years (Age); mg/dL (CT; TG; HDL); copies/mL after logarithmic transformation (Viral load); cells/µL (CD_4^+ T-cell counts); days (Time).

Discussion

Reduced levels of TC, HDL-cholesterol and elevation of TG were observed in earlier studies analyzing HIV-infected patients before the introduction of HIV-protease inhibitors [14-17]. In a cohort of 120 HIV-infected patients compared to seronegative controls, we found lower TC (171 $\pm$ 3 mg/dL and 185 $\pm$ 6 mg/dL), higher TG (159 $\pm$ 7 and 114 $\pm$ 9 mg/dL) and lower HDL-cholesterol (34 $\pm$ 1 and 50 $\pm$ 2 mg/dL) [14].

Several non-exclusive mechanisms have been postulated to explain these findings including alterations in lipoprotein metabolism produced by cytokines involved on the immune response to infection. Hypertriglyceridemia could be a result of an increase in VLDL-cholesterol synthesis or a reduction on the hydrolysis of chylomicrons and VLDL by lipoprotein lipase. Heavy alcohol consumption, nutritional status, and diet modifications also figure as possible causes for these findings [14].

In the present study, we found a similar lipid profile in HAART-naive patients. The introduction of HIV-protease inhibitors was followed by an elevation is CD_4^+ T-cell counts, an average 31% elevation of total-cholesterol, and a marked 146% average increment on triglycerides in the absence of significant modifications on weight that could explain it. As previously observed, these findings are part of a highly prevalent syndrome that includes a peripheral fat wasting called lipodystrophy, central adiposity, breast hypertrophy in women and insulin resistance or type 2 diabetes [9].

The pathogenesis of this syndrome still remains unknown but some authors suggest that it could be related to the homology of the catalytic region of HIV-1 protease, the site to which PI bind, to regions within two proteins that are involved in lipid metabolism: cytoplasmic retinoic-acid binding protein type 1 (CRABP-1) and low density lipoprotein-receptor-related protein (LRP). According to this hypothesis, protease inhibitors will

inhibit CRABP-1 and bind to LRP leading to the metabolic and clinical findings in the syndrome, including hyperlipidemia [5].

Hyperlipidemia, which has been observed as early as a few weeks after the introduction of HIV-protease inhibitors, became a major concern because of its association with atherosclerosis [9]. In fact, several recent case-reports described cardiovascular events in young patients taking PI and showing high levels of TC and TG in the absence of other risk factors [2,4-5].

The finding that 60% of patients with elevated levels of TC and/or TG in our study and that combined hyperlipidemia is the most common type of alteration observed, addresses the question if nutritional and/or pharmacological treatment for this specific condition is effective and safe.

The treatment for HIV-protease inhibitors related hyperlipidemia has gained great interest in the literature and therapeutic approaches were proposed. Dietary modifications could be difficult to achieve in this population with frequent gastrointestinal symptoms and fear of weight loss. Great concern has been expressed regarding the pharmacological treatment in this situation: there is a potential risk of skeletal muscle toxicity due to increased plasma levels of statins caused by CYP3A4 inhibition by PI, and of lower serum levels of PI related to p450 induction by these drugs, leading to virologic failure [7-8].

A fibrate plus a statin could be the best option for treatment, as combined hyperlipidemia often requires the association of these drugs. However, only small and no prospective studies were reported [6]. Limited data suggest that atorvastatin and gemfibrozil are effective in lowering both cholesterol and triglycerides although these lipids remained frequently above recommended levels in spite of treatment [6].

Fenofibrate might be an alternative to the statin-fibrate association in the treatment of HIV-protease inhibitor induced hyperlipidemia. Actually, by reducing both triglycerides and LDL-cholesterol without statin administration, fenofibrate might have safety and cost advantages in this situation. A previous report described the use of fenofibrate in two HIV-positive patients, provoking a marked decrease on triglycerides but still above recommended levels [17].

Although in a small and non-randomized pilot study, we observed an increase in HDL-cholesterol levels and a decrease in TG and TC, after the introduction of fenofibrate. In spite of the still high prevalence of patients not showing desired levels of blood lipids (76%), treatment with fenofibrate was associated with a "less atherogenic" lipid profile. Moreover, the fact that no patient showed elevation of CK and no significant variations were observed on CD_4^+ T-cell counts and viral load suggests that this treatment is safe.

Finally, considering the 7 patients with moderate hypertriglyceridemia (TG between 200-500 mg/dL), all but one showed normal or near normal levels of triglycerides after treatment (Table 4).

Clinical implications

The increment in life expectancy after the introduction of HIV-protease inhibitors will certainly increase the period of time for vascular endothelium to be exposed to a high lipidic, atherogenic milieu associated with the use of these drugs. This phenomenon could lead to an elevation in the incidence of cardiovascular events like an acute myocardial infarction or unstable angina. The results of primary and secondary atherosclerosis prevention studies, demonstrating the benefit of statins or fibrates, allied to the increase in life expectancy observed after the use of PI in HIV-patients might be a reason for the treatment of hyperlipidemia in this population [7,8,10-11].

The simple change from one protease inhibitor to another does not seems to be the right choice, as conclusive data about lipid levels modifications associated with different PI regimens are lacking. Conversely, future drugs should be studied looking for different or no effects on lipoprotein metabolism.

Diet and, if necessary, pharmacological treatment has been recommended [7,8]. However, once treatment is started, we still need to know for how long and if recommended levels are the same for this younger and, theoretically, lower risk population. All of these issues must be addressed on future clinical trials involving a greater number of patients.

Fenofibrate seems to be an effective and safe choice for patients with moderate hyperlipidemia related to the use of HIV-protease inhibitors.

References

1. Carpenter C.C.J., Fischl M.A., Hammer S.M., et al. Antiretroviral therapy for HIV infection in 1998. JAMA **1998**;280:78-86.
2. SoRelle R. Vascular and lipid syndromes in selected HIV-infected patients. Circulation **1998**;98:829-30.
3. Lo J.C., Mulligan K., Tai V.W., et al. "Buffalo hump" in men with HIV-1 infection. Lancet **1998**;351:867-70.
4. Henry K., Melroe H., Huebsch J., et al. Severe premature coronary artery disease with protease inhibitors. Lancet **1998**;351:1328.
5. Carr A., Samaras K., Chisholm D.J., Cooper D.A. Pathogenesis of HIV-protease inhibitor-associated peripheral lipodystrophy, hyperlipidemia, and insulin resistance. Lancet **1998**;351:1881-3.
6. Henry K., Melroe H., Huebesch J., et al. Atorvastatin and gemfibrozil for protease-inhibitor-related lipid abnormalities. Lancet **1998**;352:1031-1032.
7. Penzak SR, Chuck SK. Hyperlipidemia associated with HIV protease inhibitor use: pathophysiology, prevalence, risk factors and treatment. Scand J Infect Dis **2000**;32:111-23.
8. Dubé M.P., Sprencher D., Henry W.K., et al. Preliminary guidelines for the evaluation and management of dyslipidemia in HIV-Infected adults receiving antiretroviral therapy. Recommendations of the Adult ACTG Cardiovascular disease Focus group. Clin Infect Dis **2000**;31(5):1216-24.
9. Carr A., Samaras K., Thorisdottir A., et al. Diagnosis, prediction, and natural course of HIV-1 protease-inhibitor-associated lipodystrophy, hyperlidemia, and diabetes mellitus: a cohort study. Lancet **1999**;353:2093-9.
10. Shepherd J., Cobbe S.M., Ford I., et al. Prevention of coronary heart disease with pravastatin in men with hypercholesterolemia. West of Scotland Coronary Prevention Study Group. N Eng J Med **1995**;333:1301-7.
11. Summary of the Second Report of the National Cholesterol Education Program (NCEP). Expert Panel on Detection, Evaluation and Treatment of high Cholesterol in Adults (Adult treatment Panel II). JAMA **1993**;269:3015-23.
12. Miller D.B., Spence J.D. Clinical Pharmacokinetics of fibric acid derivatives (fibrates). Clin Pharmacokinetic **1998**;34:155-62.
13. Packard C.J. Overview of fenofibrate. Eur Heart J **1998**;19 Suppl A:A62-5.
14. Sposito A.C., Caramelli B., Sartori A.M., Ramires J.A.F. The lipoprotein in HIV infected patients. The Brazilian Journal of Infectious Diseases **1997**;1:275-83.
15. Grunfeld C., Pang M., Doerrler W., et al. Lipids, lipoproteins, triglycerides clearance, and cytokines in human immunodeficiency syndrome. J Clin Endocrinol Metab **1992**;74:1045-52.
16. Constans J., Pellegrin J.L., Peuchant E., et al. Plasma lipids in HIV-infected patients: a prospective study in 95 patients. European Journal of Clinical Investigation **1994**;24:416-20.
17. Thomas J.C., Lopes-Virella M.F., Del Bene V.E., et al. Use of fenofibrate in the management of protease inhibitor-associated lipid abnormalities. Pharmacotherapy **2000**;20(6):727-34.