The Effect of Rosiglitazone on Insulin Sensitivity, Beta Cell Function, Bone Mineral Density, and Body Composition in HIV-positive Patients on Highly-active Antiretroviral Therapy (HAART)

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Key words
- lipodystrophy
- adiponectin
- leptin

Abstract

Highly active antiretroviral therapy (HAART) leads to lipodystrophy and is associated with detrimental changes in glucose and lipid metabolism. This study investigated the impact of rosiglitazone on insulin sensitivity, beta cell function, bone mineral density, and body composition in HIV+ nondiabetic subjects under HAART. In this randomized, double blind, placebo controlled parallel group study, 40 HIV+ subjects were treated with rosiglitazone 4 mg/day (R, n = 23) or placebo (P, n = 17) for 6 months. Glucose, insulin and C peptide concentrations were analyzed for assessing insulin sensitivity and secretion. Adiponectin and leptin were evaluated. Body fluid compartments were measured with bioelectrical impedance spectroscopy, and bone mineral density and body composition with Dual X Ray absorptiometry. Rosiglitazone improved peripheral insulin sensitivity (+36.7 ± 15.7 ml/min/m^2, p = 0.03, means ± SEM), while no change was observed in P (+4.5 ± 19.5 ml/min/m^2, p = 0.55). Liver insulin resistance, beta cell activity, and hepatic insulin clearance did not change. Plasma adiponectin increased (R: +2.47 ± 0.86 µg/ml, p = 0.01 vs. P: +0.45 ± 0.60, p = 0.28). Rosiglitazone had no influence on body composition, fat distribution and bone mineral density but expanded extra-cellular fluid volume in HIV infected persons (R: +0.50 ± 0.21 l, p = 0.02 vs. P: 0.10 ± 0.25 l, p = 0.32). Lipid metabolism in P remained unchanged, in R total cholesterol and LDL cholesterol levels increased significantly (p < 0.05). Rosiglitazone treatment resulted in improved peripheral insulin sensitivity with increased circulating adiponectin in HIV patients under HAART. No effect was seen on body fat distribution, bone mineral density, and weight. These side effects and their potential for cardiac risk must be weighed against the beneficial effects on glucose metabolism.

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Introduction

The introduction of antiretroviral combination regimens has resulted in a dramatic decrease of morbidity and mortality and an improved quality of life among patients with AIDS over the last years. Limiting factors for the long-term success of highly active antiretroviral therapy (HAART) are the detrimental side effects such as metabolic alterations, like insulin resistance and lipodystrophy. Metabolic abnormalities during HAART comprise unfavorable alterations in lipid and glucose metabolism, which might potentially result in the development of type 2 diabetes mellitus (type 2 DM) [1]. In this regard, impaired glucose tolerance has been reported in 16 to 35% [1,2] and type 2 DM in about 7% of the treated subjects [2].

To date, pathogenic mechanisms of lipodystrophy are poorly understood. Protease inhibitors (PI) and nucleoside-analogue reverse transcriptase inhibitors (NRTI) are supposed to act synergistically in a cascadic process. In general, lipodystrophy is seen as a consequence of the fact that lipid and adipocyte regulatory proteins are partial homologues to the catalytic site of HIV-1 protease, to which PIs bind. In fact, indinavir and other PIs directly affect the intrinsic transport activity of glucose transporter-4 (Glut-4), which is a key element in the development of type 2 DM [3,4]. On the other hand, NRTI cause increased rates of lipolysis [5], which has been described as a potential mechanism for inducing insulin resistance through inhibition of insulin-stimulated glucose uptake and increased insulin-suppressed hepatic glucose output [6].

Thiazolidinediones (glitazones, TZDs), which are peroxisome proliferator-activated receptor-γ (PPAR-γ) agonists, improve insulin sensitivity in liver and peripheral skeletal muscle [7] and have emerged as an effective treatment for type 2 DM. TZDs promote fatty acid uptake and storage in adipose tissue and thereby result in weight gain and increase of subcutaneous adipose-tissue mass [8,9]. Therefore, TZDs may represent a therapeutic option to improve metabolic alterations and lipoatrophy in HIV-infected subjects under HAART. Clinical trials using rosiglitazone in HIV-infected patients, however, demonstrated varying results regarding lipid and glucose metabolism [10–13]. In addition, recent publications suggest that glitazones might negatively impact bone metabolism of diabetic patients [14–16] beyond the known adverse effects like weight gain, edema, and increased low density lipoprotein cholesterol levels.

We therefore performed the present study to determine the impact of rosiglitazone on glucose metabolism, lipodystrophy, and bone mineral density in nondiabetic HIV-infected subjects under HAART therapy. Adiponectin and leptin were also determined to further elucidate the possible role of adipocytokines on potential underlying mechanisms.

Participants and Methods

A total of 69 nondiabetic HIV-infected patients with lipodystrophy were screened to determine the eligibility for a 6 months randomized, double-blind, placebo-controlled trial and an additional 6 months open label period of rosiglitazone (4 mg once daily). Patients were recruited through the HIV-outpatient clinic at the Medical University Vienna, Department of Dermatology. Nineteen individuals did not agree to participate in the study because of lack of time, in three individuals fat redistribution was not confirmed by an investigator, in one patient liver aminotransferase exceeded the normal level more than 2.5-fold, and one patient had type 2 DM. Forty-four patients were considered eligible for the present study and were randomized either to rosiglitazone or placebo. The study was approved by the Ethic Committee on Human Studies of the Medical University of Vienna. All patients gave written informed consent before study entry.

Patient characteristics are shown in **Table 1**. Exclusion criteria were diabetes mellitus, history of myocardial infarction, malignancy, chronic kidney or liver disease, seizure, and psychiatric disorders. In no patient osteoporosis or reduced bone mineral density was diagnosed and none of them was treated for this condition. Eligibility criteria included stable HAART for more than three months. At the start of the study no uniformly objective criteria for diagnosing lipodystrophy were available. Therefore, patients were diagnosed by self-reported evidence of fat redistribution including lipoatrophy and/or lipoaccumulation confirmed on physical examination by an investigator as well as clinically significant changes of lipid and/or glucose metabolism.

Blood samples

Blood samples, except those for glucose concentrations, were immediately centrifuged and stored at −80 °C.

Plasma HIV-RNA quantification

Plasma HIV-RNA levels were determined using an in vitro nucleic acid amplification test (lower limit of detection: 50 copies/ml; Roche Ultrasensitive Assay; Roche Diagnostic Systems).

CD4 cell counts

Peripheral blood CD4 lymphocyte subset counts were determined by FACScan flowcytometer using monoclonal antibodies, and the Cellquest software (Becton-Dickinson, Mountain View, CA, USA) [17].

Laboratory measurements

Fasting cholesterol, HDL and LDL cholesterol (LDL-C), triglycerides (TG), hemoglobin (HGB), and liver function (Alanine aminotransferase ALAT, Aspartate transaminase AST, Gamma-glutamyl transferase GGT) were determined in the routine biochemical laboratory. Adiponectin and leptin were determined in duplicate by commercially available radioimmunoassay (RIA; Human Leptin and Human Adiponectin RIA, Linco Research, Inc., St. Charles, Mo. USA) with an intra- and inter-assay coefficient of variation of < 6 % for all assays.

Oral glucose tolerance test

Each patient underwent an standardized oral glucose tolerance test (OGTT, 75 mg, as described before [18]) in randomized order before and six months after study entry. The OGTT was carried out to obtain additional information on dynamic insulin sensitivity and stimulated pancreatic secretory capacity. Plasma insulin and C-peptide concentrations were analyzed by commercially available radioimmunoassay from Linco. Inter- and

Table 1 Baseline characteristics (mean ± SEM) of study population

Variable	Both groups (n = 40)	Placebo (n = 17)	Rosiglitazone (n = 23)
Age (years)	46.0 ± 1.5	46.7 ± 2.1	45.4 ± 2.1
Sex, n (male/female)	37/3	16/1	21/2
Duration of HIV infection, years	9.3 ± 0.7	8.6 ± 1.1	9.9 ± 1.0
Duration of antiretroviral therapy, years	7.04 ± 0.6	6.7 ± 0.7	7.3 ± 0.9
CD4 count [cells/mm^3]	527.3 ± 41.5	523.1 ± 65.9	530.4 ± 54.5
Undetectable HIV RNA level (%)*	90	89	91
Antiretroviral use, n (%)	100	17	23
Nucleoside reverse transcriptase inhibitor, n (%)	40	17	23
Non-nucleoside reverse transcriptase inhibitor, n (%)	29	11	16
Thymidinanalogue, n (%)	28	11	17
Protease inhibitors	10	2	8
Other medication in use, n (%)			
Lipid lowering agent, n (%)	15	5	9
Antihypertensive therapy, n (%)	6	1	5

*Less than 50 copies/ml HIV RNA

Table 2 Anthropometric and body composition variables (mean ± SEM) at baseline and after 6 months' treatment with rosiglitazone

Variable		Placebo		Rosiglitazone		Change from baseline		Sig. Between groups
						Placebo	Rosiglitazone	
		Baseline	6 months	Baseline	6 months	Δ Mean ± SEM	Δ Mean ± SEM	
Weight	kg	69.1 ± 1.9	69.0 ± 2.0	73.2 ± 2.7	73.8 ± 2.6	−0.1 ± 0.5	0.6 ± 0.5	n.s.
BMI	kg/m^2	22.3 ± 0.4	22.2 ± 0.4	23.8 ± 0.7	24.0 ± 0.6	−0.05 ± 0.2	0.2 ± 0.2	n.s.
Skinfold								
Upper arm	mm	5.7 ± 0.4	6.1 ± 0.4	5.9 ± 0.5	5.8 ± 0.6	0.4 ± 0.3	0.10 ± 0.22	n.s.
Thigh	mm	5.9 ± 0.4	5.9 ± 0.3	5.8 ± 0.6	6.4 ± 0.8	−0.1 ± 0.3	0.7 ± 0.3*	n.s.
Waist/Hip		0.95 ± 0.01	0.97 ± 0.01	1.0 ± 0.01	0.98 ± 0.01	0.0 ± 0.01	0.0 ± 0.0	n.s.
Resistance$_{BIA}$	ohm	469.2 ± 12.8	478.6 ± 16.1	460.6 ± 12.3	441.5 ± 10.6	9.4 ± 6.7	−19.1 ± 5.4*	p = 0.002
Reactance$_{BIA}$	ohm	52.3 ± 2.2	53.7 ± 1.9	53.3 ± 2.1	50.7 ± 1.8	1.4 ± 1.7	−2.5 ± 1.1*	p = 0.03
Extracellular fluid$_{BIS}$	l	18.2 ± 0.5	18.1 ± 0.7	18.3 ± 0.6	19.1 ± 0.6	−0.1 ± 0.2	0.50 ± 0.2*	p = 0.03
Intracellular fluid$_{BIS}$	l	24.3 ± 1.0	24.7 ± 1.1	25.7 ± 1.2	26.4 ± 1.4	0.1 ± 0.3	0.5 ± 0.5	n.s.
DXA Measurements								
Fat mass $_{upper\ extremities}$	kg	1.3 ± 0.1	1.3 ± 0.1	1.7 ± 0.2	1.7 ± 0.2	−0.0 ± 0.1	−0.00 ± 0.06	n.s.
Fat mass $_{limb}$	kg	1.9 ± 0.1	1.9 ± 0.5	2.6 ± 0.3	2.6 ± 0.3	−0.0 ± 0.1	−0.0 ± 0.1	n.s.
Fat mass $_{trunk}$	kg	5.6 ± 0.6	5.8 ± 0.6	6.9 ± 0.7	6.8 ± 0.6	0.2 ± 0.2	−0.1 ± 0.2	n.s.
Fat mass $_{total}$	%	14.1 ± 1.1	14.2 ± 1.1	16.6 ± 1.2	15.4 ± 1.2	0.22 ± 0.2	−0.1 ± 0.3	n.s.
Lean body mass $_{total}$	kg	58.8 ± 1.6	59.6 ± 1.8	60.0 ± 2.0	61.7 ± 2.1	0.9 ± 0.3*	1.8 ± 0.5*	n.s.
Bone Mineral Density								
Lumbar spine	g/cm^2	1.0 ± 0.02	1.0 ± 0.02	1.0 ± 0.03	0.9 ± 0.05	0.0 ± 0.0	0.0 ± 0.0	n.s.
Femoral neck	g/cm^2	0.8 ± 0.02	0.8 ± 0.02	0.8 ± 0.02	0.8 ± 0.02	0.0 ± 0.0	0.0 ± 0.0	n.s.
Hip total	g/cm^2	0.9 ± 0.02	0.9 ± 0.02	0.9 ± 0.03	0.9 ± 0.02	0.0 ± 0.0	0.0 ± 0.0	n.s.

*$p < 0.05$ vs. baseline. BIS: Bioelectrical Impedance Spectroscopy; BIA: Bioelectrical Impedance

intra-assay coefficients of variation of both assays were 5 and 8 %, respectively.

Anthropometric and body composition measures

Measurement of body height and weight and bioelectrical impedance was performed as described by Pernerstorfer-Schoen et al. [19]. Fat-free mass (FFM) was calculated by Kyle's sex-specific predictive equation [20]. Fluid compartments (extracellular fluid ECF, intracellular fluid ICF) were measured by bioimpedance spectroscopy (Hydra ECF/ICF, 4200; Xitron Technol., San Diego, CA, USA) using the manufacturer's software.

Measurements of total body and segmental fat mass were performed with the DXA scanning technique using a Hologic QDR-4500 Delphi A instrument (Hologic, Waltham, Ma, USA) and Enhanced Whole Body 11.2 software. Circumferences of waist and skinfold thickness (SF) of the upper arm and thigh were quantified following standardized procedures. SF was measured using a Holtain caliper (Crymych, U.K.) with a CV for the day-to-day variability of 1.16.

Data analysis

Glucose, insulin, and C-peptide concentration data were analyzed by mathematical models for assessing insulin secretion, beta cell function, and insulin sensitivity [21, 22]. Insulin sensitivity from OGTT was calculated as the OGIS index that represents glucose clearance after oral administration [23]. OGIS has been extensively validated against the corresponding value obtained during the gold standard glucose clamp [23] and already successfully used in several other studies [18, 24]. Insulin sensitivity in basal conditions has been assessed by the QUICKI index [25]. The insulin secretion model reconstructs the patterns per unit volume of C-peptide beta cell release and post-hepatic insulin appearance into peripheral circulation, providing the basal fasting prehepatic insulin secretion rate per unit volume (BSR), the stimulated insulin secretion per unit volume (TIS), that is, the total amount of insulin released by the beta cell during the OGTT, and the hepatic insulin extraction (HIE), as percent of the secreted hormone during the whole test (not just the first pass) [22]. Finally, the beta cell ability to compensate for changes in insulin resistance was determined by the adaptation index, defined as OGIS × TIS [26].

Statistical analysis

Data and results are presented as means and standard error of the means (SEM). To analyze differences within groups t-test for normally distributed and Wilcoxon test for not normally distributed data were used. After logarithmic transformation differences between groups, controlled differences in baseline values, were tested with analysis of covariance (ANCOVA).

Correlations in dependency of the distribution of the data were analyzed with Pearson's and Spearman's correlation coefficient, respectively. Equality of distributions was tested with Chi-square test and Fisher's exact test. Statistical analyses were performed by using SPSS V.12.0 (SPSS, Inc., Chicago, IL, USA).

Results

Forty individuals completed the six months follow-up. Two patients withdrew their informed consent after randomization within the first week after study entry; two patients were lost for follow-up. At baseline, patients under placebo and rosiglitazone treatment were comparable regarding age, sex, and duration of HAART (**Table 1**). Metabolic variables and body composition at baseline were similar between treatment groups (**Table 2**).

Glucose metabolism

Table 3 shows measurements and parameters obtained from OGTT data. In patients treated with rosiglitazone post load plasma concentrations of insulin decreased by 21.5 % ($p = 0.01$; between groups $p = 0.005$), C-peptide by 11.7 % (between groups

Table 3 Metabolic control markers (mean ± SEM) at baseline and after 6 months treatment with rosiglitazone

Variable		Placebo		Rosiglitazone		Change from baseline		Sig. between groups
						Placebo	Rosiglitazone	
		Baseline	6 months	Baseline	6 months	Δ Mean ± SEM	Δ Mean ± SEM	
$Glucose_{basal}$	mg/dl	93.9 ± 2.9	96.1 ± 3.4	103.9 ± 5.4	93.6 ± 2.0	2.2 ± 1.9	−10.26 ± 5.94	n.s.
$Insulin_{basal}$	µU/ml	15.4 ± 1.9	21.6 ± 4.8	27.7 ± 9.5	17.3 ± 2.8	6.2 ± 3.8	−1.4 ± 3.0	n.s.
$C\text{-}Peptide_{basal}$	ng/ml	4.2 ± 1.5	3.3 ± 4.0	3.3 ± 0.4	2.9 ± 0.3	−0.9 ± 1.5	−0.04 ± 0.28	n.s.
$Glucose_{120\,min}$	mg/dl	123.5 ± 5.5	128.8 ± 7.0	136.5 ± 11.8	115.8 ± 5.3	5.4 ± 6.0	−20.70 ± 11.74	n.s.
$Insulin_{120\,min}$	µU/ml	93.2 ± 18.5	110.3 ± 15.3	99.7 ± 15.1	67.3 ± 9.8	17.2 ± 16.7	−21.5 ± 1.0*	p = 0.005
$C\text{-}Peptide_{120\,min}$	ng/ml	9.5 ± 0.8	10.3 ± 10.8	10.0 ± 0.5	8.7 ± 0.6	0.8 ± 0.8	−1.2 ± 0.7	p = 0.04
BSR	pmol/l/min	56.4 ± 7.8	66.7 ± 8.1	66.9 ± 8.9	63.0 ± 5.8	12.0 ± 6.5	−3.9 ± 6.8	n.s.
TIS	nmol/l 3h	33.1 ± 2.4	35.8 ± 2.3	33.5 ± 1.7	31.7 ± 1.7	2.9 ± 2.3	−1.8 ± 1.4	n.s.
HIE	%	60.9 ± 2.7	62.7 ± 3.5	63.3 ± 3.5	64.0 ± 2.7	0.9 ± 2.9	0.7 ± 2.4	n.s.
QUICKI		0.3 ± 0.01	0.3 ± 0.01	0.3 ± 0.01	0.3 ± 0.01	−0.0 ± 0.0	0.0 ± 0.0	n.s.
OGIS	$ml/min/m^2$	382 ± 20	387 ± 21	384 ± 18	421 ± 13	4.5 ± 19.5	36.7 ± 15.7*	n.s.
Adaptation Index	$pmol/m^2$	70 ± 16	77 ± 17	73 ± 13	74 ± 11	0.9 ± 0.7	−0.9 ± 0.7	n.s.
Total cholesterol	mg/dl	248.9 ± 11.7	251.8 ± 6.6	226.0 ± 9.5	256.0 ± 12.7	2.9 ± 10.3	25.5 ± 9.6*	n.s.
LDL cholesterol	mg/dl	141 ± 9.0	150.4 ± 6.1	125.0 ± 9.7	145.4 ± 12.3	9.4 ± 8.7	18.5 ± 9.0*	n.s.
HDL cholesterol	mg/dl	42.5 ± 2.4	43.9 ± 2.4	42.1 ± 2.6	43.0 ± 3.9	2.1 ± 1.2	1.4 ± 2.7	n.s.
Triglycerides	mg/dl	365.0 ± 51.7	297.0 ± 26.7	319.2 ± 43.8	363.6 ± 57.3	−68.0 ± 55.2	44.4 ± 51.4	n.s.
Adiponectin	µg/ml	2.6 ± 0.6	3.0 ± 0.9	2.8 ± 0.4	5.3 ± 1.1	0.5 ± 0.60	2.5 ± 0.9**	p = 0.007
Leptin	ng/ml	2.9 ± 0.5	3.6 ± 0.8	4.4 ± 0.9	4.0 ± 0.8	0.7 ± 0.4	−0.4 ± 0.5	n.s.

*p < 0.05 and **p < 0.01 vs. baseline

p = 0.04), and glucose by 15.2 % (between groups p = 0.08). In the placebo-treated subjects no changes were observed. TZD treatment thus resulted in an improved oral glucose insulin sensitivity (OGIS) after the physiologic oral glucose load (p = 0.03, **Table 3**). No change was observed in the fasting insulin sensitivity (QUICKI). The measurement of C-peptide enabled a broader analysis of the effects of rosiglitazone on the glucose-stimulated insulin response. Indeed, post load C-peptide slightly decreased, indicating that beta cell function was only marginally affected, as also demonstrated by the quantification of the basal (BSR) and dynamic (TIS) beta cell (pre-hepatic) activity, which were nearly unchanged in both groups. The almost unchanged beta cell function was also confirmed by the similar adaptation index (**Table 3**). Liver insulin degradation also remained unchanged as shown by the quantification of the hepatic insulin extraction (HIE, **Table 3**). In the placebo group, neither insulin secretion nor insulin sensitivity nor insulin clearance changed when compared with baseline values.

Adiponectin and leptin

Adiponectin concentrations almost doubled after six month of rosiglitazone treatment, whilst leptin did not change significantly (**Table 3**). Change of adiponectin after 6 months correlated negatively with change of reactance (r = −0.41, p = 0.012).

Body fat mass and bone density

Six-month treatment with rosiglitazone had no effect, neither on segmental and total body fat mass nor on bone density. However, skinfold thickness of the thigh increased (p = 0.04 vs. baseline), but was not different between groups. The extracellular fluid measured with bioelectrical impedance spectroscopy (ECF_{BIS}) increased significantly (p = 0.02 vs. baseline; between groups p = 0.07). Mean body weight did not change significantly, weight gain, however, was observed in 60 % of all patients (placebo 59 %, min −3.3, max +2.2 kg; rosiglitazone 61 %, min −3.0, max +7.2 kg). Although weight and body mass index did not change significantly, a significant increase of lean body mass (LBM) was observed in both groups after 6 months (P: p = 0.023 and R: p = 0.002). The change of LBM_{DEXA} correlated with intracellular fluid (correlation coefficient r = 0.5 and p = 0.04 in both groups).

Lipid metabolism and safety variables

At baseline, in 9 patients (4 placebo, 5 rosiglitazone) plasma triglyceride levels exceeded 2.5-fold of normal values. Triglyceride levels higher than 885 mg/dl (> 10 mmol/l) were measured in two patients (one per group). After six months, lipid metabolism in the placebo group remained unchanged. In patients treated with rosiglitazone total cholesterol level increased by 25.5 ± 9.6 mg/dl (p < 0.01), LDL-C by 18.5 ± 9.0 mg/dl (p = 0.02) and triglycerides by 44.4 ± 51.4 mg/dl (p = 0.6). Prescription of lipid lowering agents increased from 38 % at baseline to 70 % at 6 months (placebo +35 %, rosiglitazone +31 %). Other safety variables are shown in **Table 4**. With regard to liver enzyme function: an increase of AST of more than 60 % was observed in both groups, however, this did not reach statistical significance (p = 0.06 for placebo, p = 0.07 for rosiglitazone). In both groups immunological and virological parameters remained unaffected during the course of the study. The increase of mean viral load in the rosiglitazone group is due to treatment failure in one patient, the median of viral load remained stable in both groups.

Influence of treatment agents

Twenty-eight patients were treated with thymidin-analogues (11 in the placebo and 17 in the rosiglitazone group). Two patients of the placebo group and 8 of the rosiglitazone group were treated with a protease inhibitor. Distributions within the two groups did not differ significantly (p = 0.15). There was no statistically significant influence of thymidin analogues and protease inhibitors on the change of the metabolic parameters in patients treated with rosiglitazone.

Discussion

Our study found improved parameters of glucose metabolism and increased adiponectin levels following treatment with

Table 4 Safety variables (mean ± SEM)

Variable		Placebo		Rosiglitazone		Change from baseline		Sig. between groups
						Placebo	Rosiglitazone	
		Baseline	6 months	Baseline	6 months	Δ Mean ± SEM	Δ Mean ± SEM	
GGT	U/l	38.3 ± 4.9	49.0 ± 8.4	63.3 ± 16.5	62.7 ± 19.3	9.53 ± 5.23	−0.65 ± 18.25	n.s.
AST	U/l	15.18 ± 1.3	26.1 ± 4.7	18.09 ± 1.7	29.1 ± 4.9	10.06 ± 4.41	11.00 ± 4.13	n.s
ALT	U/l	22.5 ± 3.0	28.0 ± 4.2	23.4 ± 2.3	33.2 ± 5.9	5.88 ± 3.89	9.74 ± 5.50	n.s.
HGB	g/dl	15.3 ± 0.3	15.1 ± 0.3	15.4 ± 0.3	14.9 ± 0.3	−0.32 ± 0.19	−0.54 ± 0.18*	n.s.
CD4 count	cells/mm^3	523.1 ± 65.9	536.5 ± 74.2	530.4 ± 54.5	551.3 ± 60.2	13.4 ± 27.2	21.0 ± 31.3	n.s.
Viral load	copies/ml	717.5 ± 640.9	564.7 ± 496.9	55.9 ± 4.4	1595.1 ± 1540.3	152 ± 144	1539.2 ± 1540.6	n.s.
Undetectable viral load#	number	15	15	21	20			

#Less than 50 copies/ml HIV RNA; *$p < 0.05$ vs. baseline

rosiglitazone in HIV-infected patients with lipodystrophy. This was accompanied by an expansion of extracellular fluid. However, no significant influence of rosiglitazone treatment on body composition, bone mineral density and fat distribution was observed. Lipodystrophy and associated metabolic alterations are major health problems in HIV-infected individuals treated with HAART. TZDs have been shown to improve insulin sensitivity and increase subcutaneous fat in diabetic individuals [8]. Our results demonstrated that 4 mg rosiglitazone daily over six months improved glucose metabolism in nondiabetic HIV-positive patients. Rosiglitazone treatment significantly decreased postload glucose and insulin levels. This effect was paralleled by a significant improvement of peripheral insulin sensitivity (OGIS), as also reported in other individuals treated with HAART over 3 months with 4 mg/d rosiglitazone [11]. Beta cell activity, both from insulin and C-peptide data, did not show any change, indicating that rosiglitazone treatment only acted on insulin action. To assess insulin action we have employed two methods, OGIS and QUICKI. It has been shown that OGIS correlates strongly with total glucose disposal, as assessed by the glucose clamp, while QUICKI correlates with the basal hepatic insulin resistance [24]. Thus, in our study the effect of rosiglitazone was peculiarly on muscle and adipose tissue, while liver insulin sensitivity does not appear to be affected, since QUICKI did not change. Hepatic insulin clearance as well was unchanged following treatment with rosiglitazone.

In HIV infected individuals with lipodystrophy plasma concentration of adiponectin has been shown to be decreased [27] or within the normal range [28]. It has been proposed that hypoadiponectinemia is linked to metabolic disorders induced by HAART [29]. PPAR-γ agonists, like rosiglitazone, increase the expression and plasma concentrations of adiponectin in diabetic individuals [30,31]. The increase of adiponectin in our patients is in line with what has been observed in diabetic patients. Other authors have reported similar findings in HIV infected individuals [12,13,32]. Leptin, which closely correlates with subcutaneous fat deposition, did not change following rosiglitazone treatment. This might reflect the unchanged fat mass and distribution as assessed by DXA measurements.

In our study population, however, treatment with rosiglitazone neither ameliorated body fat distribution nor improved segmental or total body fat mass as shown in patients with type 2 DM [8]. In contrast to previous results [10,11], lipdystrophy remained unaffected. In this regard, one could argue that the rosiglitazone dose of 4 mg/d was not high enough to influence body composition. The observed significant effects on glycemic control, adiponectin and the expansion of the extracellular fluid compartment, suggest that the dose chosen should be adequate to also affect fat distribution. Furthermore, our negative results on body composition measures are in accordance with two other trials which used rosiglitazone 8 mg/d [12,13]. Despite the absence of an effect on body fat, we observed an increase of ECF, without seeing any edema in the treatment group. Therefore, in case of weight gain, monitoring fluid compartments by BIS seems to be an appropriate opportunity to distinguish between increase of fat mass and expansion of the extracellular fluid compartment. It is unlikely that the significant increase of LBM is due to the treatment with rosiglitazone, since both groups experienced an increase in LBM. Therefore, this increase in LBM could be linked to lifestyle changes and optimized nutrition in association with a more intensive care during the study period.

Recent reports suggest a negative impact of rosiglitazone on bone mineral density. Grey et al. [14] reported bone loss in healthy women without diabetes or osteoporosis treated with TZD 8 mg/d over 14 weeks. Moreover, the Health ABC study observed a greater bone loss in older diabetic women, but not in men during an observation period of four years [33]. In contrast, a retrospective observational study suggests a possible negative influence of TZDs over four years on bone metabolism even in older men with diabetes [15]. In the present study, treatment with 4 mg/d rosiglitazone over a 24-week period did not result in bone loss in non-diabetic, albeit younger, HIV-infected patients. The relatively small sample size, the lower dose of rosiglitazone, the short treatment period and the presence of other confounders like smoking, exercise, and eating behavior, which we are unable to control, might limit the validity of this finding.

Concerns have been expressed regarding adverse effects of rosiglitazone on lipoprotein concentrations and particle size in diabetic patients [34] and HIV positive patients [12,13,35]. These findings were confirmed in our study. We observed a significant increase of total cholesterol and LDL-C concentrations, even though lipid lowering agents were more frequently prescribed in both groups. We did not investigate particle size. The increase of already elevated triglyceride levels following rosiglitazone treatment might explain the worsening of liver enzymes, which indicate fatty liver disease. In addition, the more frequent prescription of lipid lowering agents may have further counteracted the beneficial effect of rosiglitazone on liver function as seen in other studies [1,2]. The decrease of hemoglobin in our study is a common finding in patients treated with glitazones [11,13]. However, this decline cannot be explained by the increase of total body water and was also observed in diabetic

individuals treated with pioglitazone [36]. The underlying mechanism, however, is not clear yet.

In conclusion, treatment with rosiglitazone in HIV positive individuals leads to an improvement of peripheral insulin sensitivity with a concomitant increase in adiponectin, without altering liver insulin resistance and beta cell function. Bone mineral density remained unchanged. Neither body fat distribution nor segmental or total body fat mass were ameliorated, while extracellular fluid volume expanded and LDL-C levels increased. When TZD treatment is considered in HIV infected patients, these side effects and their potential for cardiac risk must be weighed against the beneficial effects on glucose metabolism.

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Conflict of Interest

B. Ludvik has been paid by Glaxo Smith Kline for lectures and has received fees for consulting. Role of the funding source: Glaxo Smith Kline supported the study by providing the medication. There was no influence on design, conduct, or reporting of the study, nor on the decision to submit the manuscript for publication from the side of Glaxo Smith Kline.

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