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Effects of ritonavir and amprenavir on insulin sensitivity in healthy volunteers

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Abstract

Background—Some HIV protease inhibitors (PIs) have been shown to induce insulin resistance *in vitro* but the degree to which specific PIs affect insulin sensitivity in humans is less well understood.

Methods—In two separate double-blind, randomized, cross-over studies, we assessed the effects of a single dose of ritonavir (800 mg) and amprenavir (1200 mg) on insulin sensitivity (euglycemic hyperglycemic clamp) in healthy normal volunteers.

Results—Ritonavir decreased insulin sensitivity (−15%; $P=0.008$ versus placebo) and non-oxidative glucose disposal (−30%; $P=0.0004$), whereas neither were affected by amprenavir administration.

Conclusion—Compared to previously performed studies of identical design using single doses of indinavir and lopinavir/ritonavir, a hierarchy of insulin resistance was observed with the greatest effect seen with indinavir followed by ritonavir and lopinavir/ritonavir, with little effect of amprenavir.

Keywords

amprenavir; diabetes; HIV protease inhibitors; indinavir; insulin resistance; lopinavir/ritonavir; ritonavir

Introduction

Shortly after the introduction of highly effective antiretroviral therapy with HIV protease inhibitors (PI), a number of disturbances in glucose metabolism including fasting hyperglycemia, impaired glucose tolerance, and frank diabetes were reported [1,2]. Previous studies from our laboratory have established that both single and repeated doses of the PI indinavir decreased insulin-mediated glucose disposal in healthy volunteers [3,4]. The

effects of other PIs on insulin sensitivity, however, remain unclear. From a clinical standpoint, understanding the degree of induction of insulin resistance of a given PI relative to other PIs is advantageous in weighing the benefits of efficacy versus the adverse effects on glucose metabolism. There are few prospective studies of PIs and insulin sensitivity, and differences in design and methodology make direct comparison of single PIs among such studies difficult.

The design of the clinical trial is an important consideration in the comparative assessment of the effects of PIs on insulin sensitivity. In patients with HIV infection, it is rare to initiate single-agent PI therapy without other antiretrovirals, and thus the effects seen in such individuals may be mediated in part by concomitant antiretroviral medications. Furthermore, HIV infection itself, therapy-induced restoration to health, immunoreconstitution, and changes in body composition may contribute to alterations in insulin sensitivity. The use of healthy normal volunteers in studies allows for the isolation of the direct effects of PIs. Moreover, a placebo/drug cross-over design permits the further reduction in variance between study participants.

To compare the effects of different PIs on insulin sensitivity, we performed two, separate, randomized, double-blind, placebo-controlled trials of ritonavir and amprenavir using the euglycemic hyperinsulinemic clamp technique in healthy normal volunteers. Because *in vitro* studies have shown that PIs inhibit insulin-mediated glucose disposal through an acute and direct blockade of the peripheral glucose transporter GLUT4 [5], a single dose of PI was administered prior to the start of the euglycemic hyperinsulinemic clamp study. We have previously shown that a single dose of indinavir or lopinavir/ritonavir acutely inhibited insulin-mediated glucose disposal in healthy subjects [4,6], providing support for the *in vitro* model. Here, we take advantage of this single-dose study design in order to rapidly and effectively replicate nearly identical study conditions between two studies of the drugs ritonavir and amprenavir. By using the same design as the previously reported studies of the effects of a single dose of anti-retroviral drugs on insulin sensitivity, we are better able to compare the effects of ritonavir and amprenavir to those previously reported for indinavir and lopinavir/ritonavir.

Methods

Eight healthy men on no medications were recruited from the community for the ritonavir study, and six of those men subsequently returned for the amprenavir study. The participants had no history of medical illnesses and no abnormalities on screening physical examination or routine hematology and chemistry tests. HIV-1 antibody test was negative prior to the study. The study protocol was approved by the Committee on Human Research of UCSF, and informed consent was obtained from each individual.

Exclusion criteria included body mass index $>27 \text{ kg/m}^2$, serum total cholesterol $> 6.2 \text{ mmol/l}$, triglycerides $> 3.8 \text{ mmol/l}$, fasting glucose $> 7.0 \text{ mmol/l}$, serum aspartate or alanine aminotransferases $> 50 \text{ U/l}$ and creatinine $> 124 \text{ } \mu\text{mol/l}$.

Study design

Ritonavir and amprenavir were each studied in randomized, double-blind, placebo-controlled trials. The participants were instructed to eat a diet containing at least 150 g of carbohydrate for 3 days prior to each study and they kept a diet journal 3 days prior to each study, which was reviewed by a dietitian to assess dietary adherence. The participants were admitted to the General Clinical Research Center (GCRC) at the San Francisco General Hospital (SFGH) the morning prior to the day of the study and began a 24-h urine collection.

After an overnight (10 h) fast, blood was drawn for baseline studies prior to the administration of the study medication or placebo.

In the ritonavir study, participants randomly received a single dose of either ritonavir (Abbott Laboratories Abbott Park, Illinois USA) 800 mg soft gel caps or placebo. Ritonavir or placebo was given 2 h before the start of the clamp based on the drug pharmacokinetic profile with the intent of reaching peak levels at the start of the clamp. In the amprenavir study, a single dose of amprenavir (GlaxoSmithKline Research Triangle Park, North Carolina, USA) 1200 mg soft gel caps or placebo was given to subjects 1 h before the start of the clamp. A euglycemic hyperinsulinemic clamp was performed from 0900–1200 h by an investigator blinded to the study medication. The participants were re-admitted to the GCRC within 7–28 days for alternative treatment (active drug or placebo), and the above studies were repeated.

We chose to study full-dose ritonavir, as insulin resistance and changes in glucose metabolism were reported early in the use of PIs, when full, rather than boosting, doses were used. We chose an 800 mg dose of ritonavir because only a single dose was given without the food normally used to enhance absorption. We expected that the standard dose of ritonavir (600 mg) might not achieve and maintain plasma concentrations during the euglycemic hyperinsulinemic clamp. Furthermore, plasma concentrations of ritonavir are highly variable. The time to peak concentration is 2 h, and the half-life is approximately 3 to 5 h for the 600 mg dose in the fasting state [7]. The pharmacokinetic profile of ritonavir is based on oral solution studies. Amprenavir is more bio-available (90% plasma protein binding) than ritonavir (98–99% plasma protein binding), and food is not required to enhance the absorption of amprenavir [8]. Therefore we chose the standard dose of 1200 mg of amprenavir to reach $C_{\max}$ in 1 h. The time to peak concentration of amprenavir is approximately 1–2 h, and the half-life of amprenavir is approximately 7–10 h.

Euglycemic hyperinsulinemic clamp

The clamp was performed as described by DeFronzo *et al.* [9]. Participants fasted overnight prior to the procedure. Cannulae were placed into antecubital veins bilaterally and a vein in the dorsum of the hand which was kept in a heated box at 50–55°C, for arterialized venous blood sampling. At $t=0$, insulin (Humulin R; Eli Lilly, Indianapolis, Indiana, USA) was administered as a primed continuous intravenous infusion for 10 min, followed by a constant infusion at the rate of 40 mU/m² per min until $t=180$ min. Whole blood glucose concentration was measured every 5 min after the start of the insulin infusion. 20% dextrose was infused at a variable rate to maintain the plasma glucose concentration at 4.5 mmol/l with a coefficient of variation <5% based on the negative feedback principle. Blood samples were also collected for determination of plasma glucose and serum insulin concentrations. Insulin-mediated glucose disposal was calculated using data from the last hour ($t=120$ to 180 min) of the euglycemic hyperinsulinemic clamp. Insulin-mediated glucose disposal was obtained from all eight participants from the ritonavir study and all six participants from the amprenavir study.

Resting energy expenditure

The O₂ consumption and CO₂ production were measured by indirect calorimetry (DeltaTrac metabolic monitor; Yorba Linda, California, USA) at 0730 h prior to and during the last hour of the clamp study. Non-protein respiratory quotient and substrate oxidation rates were calculated after correction for protein oxidation. Protein oxidation was estimated by urea nitrogen excretion measured in the 24-h urine collection [10]. The rate of non-oxidative glucose metabolism was calculated by subtracting the rate of carbohydrate oxidation from the rate of dextrose infusion during the clamp. We were unable to obtain resting energy

expenditure in one individual in the ritonavir study and one in the amprenavir study. We were also unable to obtain non-oxidative and oxidative glucose metabolism in one individual in the ritonavir study.

Measurements

Whole blood and plasma glucose and lactate were measured using a glucose analyzer (YSI 2300 STAT-Plus Glucose and Lactate Analyzer; YSI Inc., Yellow Springs, Ohio, USA). Serum insulin levels were determined by radioimmunoassay (Linco Research, Inc., St. Charles, Missouri, USA) with a 3.2% intra-assay coefficient of variation, a lower detection limit of 14.3 pmol/l, and 0.2% cross-reactivity with proinsulin. Homeostasis model assessment insulin resistance index (HOMA-IR) was calculated from baseline fasting plasma glucose and fasting serum insulin which were measured before administration of the PI or placebo and again at the start of the clamp (2 h after administration of ritonavir or 1 h after administration of amprenavir or their respective placebos) [11]. Baseline fasting lipids and free fatty acids (prior to administration of medication) were measured at the start of the clamp by enzymatic colorimetric methods (Sigma Diagnostics, St. Louis, Missouri, USA and Wako Chemicals, Richmond, Virginia, USA).

Ritonavir and amprenavir levels were measured by liquid chromatography, tandem mass spectrometry within the Drug Research Unit, San Francisco General Hospital. For ritonavir, the method had a lower detection limit of 25 ng/ml, with an inter- and intra-assay coefficient of variation ranging from 5.6 to 10.6% and 3.1 to 6.4%, respectively. For amprenavir, the method had a lower detection limit of 44 ng/ml, with inter- and intra-assay coefficients of variation ranging from 5.2 to 9.6% and 5.2 to 10.2%, respectively. The area under the concentration time curve (AUC) during the study (10–180 min of the clamp) was estimated using the linear-linear trapezoidal rule. By a predefined criterion, we excluded data from subjects whose PI levels did not reach within one standard deviation of average peak values as reported in the literature.

Statistical analysis

Data were analyzed using Sigma Stat v. 3.0 (Systat Software, Inc., San Jose, California, USA). Paired *t* tests were used. Data are presented as mean±SEM. *P* values are two-tailed.

Results

The participants ranged in age from 25 to 69 years. Mean ages for the ritonavir and amprenavir studies were 49±5 and 49±8 years, respectively. Body weight, resting energy expenditure, and fasting serum insulin, plasma glucose, lactate, and lipids did not differ when measured prior to administration of active drug or placebo in each study (Table 1a).

In the ritonavir study, all but one subject achieved therapeutic drug levels during the clamp (Fig. 1a and b). The plasma level of ritonavir reached a concentration of 9.7±1.3 mg/l and remained > 8.7 mg/l, which is 2.2-fold over $C_{\min}$ (4 mg/l) throughout the clamp study (Fig. 1a). The 3-h AUC for ritonavir was 27.4±3.2 mg/l per h. The one participant who did not achieve therapeutic drug levels was excluded from the final analysis. The inclusion of his data did not, however, affect the significance of any change in the outcome measures, including fasting plasma glucose or serum insulin, insulin-mediated glucose disposal, oxidized glucose disposal, or non-oxidized glucose disposal. The plasma level of amprenavir reached 6.6±0.8 mg/l at the start of the clamp; 1 h later, the plasma levels declined to 2.9±0.4 mg/l and remained > eight-fold over $C_{\min}$ (0.33 mg/l) until the end of the study (Fig. 1b). The 3-h AUC for amprenavir was 9.5±1.4 mg/l per h.

In addition to measuring fasting insulin and glucose levels prior to receiving medication or placebo, fasting insulin and glucose levels were measured prior to the start of the clamp to assess the effects of the drugs on HOMA-IR. Two hours after administration of ritonavir, both fasting insulin levels and HOMA-IR were significantly higher with ritonavir administration in comparison with placebo (Table 1b). In contrast, at peak levels of amprenavir (1 h after administration of drug), fasting insulin levels and HOMA-IR were not changed significantly. Neither ritonavir nor amprenavir altered fasting glucose levels at time 0 of the clamp.

During the euglycemic hyperinsulinemic clamp, steady state glucose levels ($t120-180$ min) were nearly identical between both studies (Fig. 2a and b). In the ritonavir study, glucose levels were 4.4 ± 0.02 versus 4.4 ± 0.03 mmol/l ($P=0.39$) for ritonavir and placebo, respectively (Fig. 2a). Insulin levels were 621 ± 35 and 590 ± 41 pmol/l ($P=0.11$) for ritonavir and placebo, respectively. In the amprenavir study, steady-state glucose levels were 4.4 ± 0.02 and 4.4 ± 0.03 mmol/l ($P=0.9$) for amprenavir and placebo, respectively (Fig. 2b). Steady-state insulin levels of 621 ± 30 versus 610 ± 36 pmol/l; $P=0.46$ (amprenavir versus placebo) were achieved. Fasting free fatty acid (FFA) levels were suppressed comparably with insulin administration in both studies. FFA AUC levels were 0.250 ± 0.029 and 0.230 ± 0.008 mmol/l ($P=0.43$) for placebo and ritonavir, respectively. FFA AUC levels were 0.297 ± 0.015 and 0.329 ± 0.023 mmol/l ($P=0.15$) for placebo and amprenavir, respectively.

Despite similar study conditions, the two PIs exerted different effects on insulin-mediated glucose disposal (Fig. 3a and b) and on the rate of glucose infusion required to maintain euglycemia (Fig. 2a and b). Insulin-mediated glucose disposal rate per unit of insulin (M/I) decreased from 9.4 ± 0.7 to 8.0 ± 0.7 mg/kg per min per μ U/ml insulin with placebo and ritonavir (Fig. 3a), respectively ($P=0.008$). The difference in glucose infusion can be seen as early as the 20 min time point. Mean M/I declined in the ritonavir study by an average of 16% with ritonavir in the eight study participants. The non-oxidative component of total glucose disposal decreased by 30% (from 6.1 ± 0.5 to 4.3 ± 0.5 mg/kg per min, $P=0.0004$). The oxidative component of glucose disposal did not, however, change (2.4 ± 0.3 versus 2.4 ± 0.2 mg/kg per min; $P=1.0$). All of the subjects in the amprenavir study had participated in the ritonavir study; in these six study participants, insulin-mediated glucose disposal decreased by 18% with ritonavir administration (9.3 ± 1.0 to 7.6 ± 0.9 mg/kg per min; $P=0.01$). Non-oxidized glucose disposal decreased by 30% (6.1 ± 0.5 to 4.3 ± 0.5 mg/kg per min; $P=0.0003$).

In contrast, amprenavir administration did not alter insulin-mediated glucose disposal. M/I was 8.5 ± 1.2 versus 8.4 ± 1.0 mg/kg per min per μ U/ml insulin with placebo and amprenavir, respectively ($P=0.8$) (Fig. 3b). Likewise, the non-oxidative component of total glucose disposal did not change with amprenavir administration (4.9 ± 0.9 versus 4.9 ± 0.7 mg/kg per min; $P=0.9$). The oxidative component of glucose disposal also did not change (2.2 ± 0.3 versus 2.0 ± 0.3 mg/kg per min; $P=0.5$).

Discussion

We found that single doses of the PIs ritonavir and amprenavir had different effects on insulin sensitivity as measured by insulin-mediated glucose disposal, although a similar reduction of FFA levels by insulin were observed on both drugs. Full-dose ritonavir caused a 15% reduction in insulin-mediated glucose disposal whereas amprenavir had little effect. Previously we have reported that a single dose of indinavir in therapeutic range decreased insulin-mediated glucose disposal by 34%. We have also reported that a single dose of lopinavir/ritonavir decreased insulin-mediated glucose disposal by 13% [6]. All four studies were performed under identical study conditions with participants of similar ages and ethnic

composition. While direct comparison of four drugs was not possible in single study, the identical study design in similar study populations permits a relative comparison of the effects of each PI versus placebo. Taken together, we found that indinavir had the greatest effect on insulin sensitivity as measured by insulin-mediated glucose disposal. Full-dose ritonavir and lopinavir/ritonavir had similar effects on insulin-mediated glucose disposal. In contrast, amprenavir did not alter insulin-mediated glucose disposal.

The single dose of ritonavir was given only 2 h before the study, suggesting that ritonavir acutely induced insulin resistance. These findings are consistent with an acute and immediate blockade of the peripheral glucose transporter, GLUT4, as has been demonstrated previously [5]. This decrease in insulin-mediated glucose disposal was mostly due to a defect in peripheral glucose storage, as reflected in the decline in the rate of non-oxidative glucose disposal. Both indinavir and lopinavir/ritonavir also acutely decreased non-oxidative, but not oxidative glucose disposal [4,6].

Free fatty acids were suppressed to comparable levels. The ability of insulin to inhibit lipolysis during the euglycemic hyperinsulinemic clamp with ritonavir administration suggests that insulin signaling was not globally impaired, a finding similar to that with indinavir. The increase in fasting insulin 2 h after administration of ritonavir raises the possibility that hepatic insulin sensitivity is affected. We previously have shown that 4 weeks of indinavir increases endogenous glucose production in the fasting state in addition to decreasing insulin sensitivity during the clamp [12].

In both studies, plasma levels of ritonavir and amprenavir closely resembled those observed in pharmacokinetic studies of healthy normal volunteers. During the last hour of the clamp, ritonavir drug levels were 8.7 ± 0.9 to 9.0 ± 1.0 mg/l, a range that is similar to full-dose ritonavir drug levels achieved in pharmacokinetic studies [7] and is 2.2-fold higher than the $C_{\min}$ for ritonavir. Amprenavir levels peaked at the start of the clamp followed by a decline to levels of greater than 2.0 ± 0.8 mg/l in a pattern typical of amprenavir kinetics in healthy normal volunteers and HIV-infected patients [8,13] and is more than eight-fold higher than the $C_{\min}$ for amprenavir. The single 1200 mg dose of amprenavir is known to have a biphasic pattern by which drug levels remain at less than 2 mg/l during 18 h of the once-daily dosing. At such levels, we found that insulin-mediated glucose disposal did not change. A very small reduction in insulin sensitivity may not have been detected in the amprenavir study; however, such a small reduction in insulin sensitivity may not be clinically significant. Due to the transient nature of the peak levels of amprenavir, it was not possible to measure insulin-mediated glucose disposal at peak amprenavir levels; however, HOMA-IR at peak amprenavir levels at the start of the clamp study also did not change (Table 1b). In contrast, the single dose of ritonavir increased HOMA-IR at the start of the clamp study and decreased insulin-mediated glucose disposal. These findings suggest that amprenavir has little effect on altering insulin sensitivity at either peak or average therapeutic levels. These results are for single dose amprenavir, and it is possible that amprenavir given twice daily may still have an effect on insulin resistance.

Differences in the standard therapeutic drug levels and in the solubility of these drugs in aqueous solution may partly explain the disparities in results between human, animal, and *in vitro* studies. In a comparative study of the effects of 50 $\mu\text{mol/l}$ indinavir, ritonavir, and amprenavir on glucose uptake in 3T3L-1 adipocytes, ritonavir caused a striking inhibition of 2-deoxy glucose uptake, followed by a more modest inhibition by amprenavir and indinavir [14]. These levels used *in vitro* in an assay without binding proteins, however, exceed peak plasma levels and do not take into account free drug levels. In a follow-up study, rats were infused intravenous amprenavir, ritonavir, or lopinavir/ritonavir to achieve drug levels of 8–12 $\mu\text{mol/l}$, and euglycemic hyperinsulinemic clamps were performed [15]. Interestingly, a

similar rank order of insulin sensitivity was observed in rats (ritonavir followed by lopinavir/ritonavir and amprenavir) as in our study. At 11.3 $\mu\text{mol/l}$ of amprenavir, there was an 18.4% decrease in insulin-mediated glucose disposal in rats, but the drug levels achieved in rats were much higher than those observed with typical pharmacokinetics in humans. As free drug levels may differ between rats and humans, it is difficult to extrapolate the direct effects on humans in the absence of clinical studies. In our study, drug levels were within therapeutic range for amprenavir. This suggests that the biphasic pharmacokinetics of amprenavir in humans may be advantageous in maintaining drug concentrations at levels that do not induce insulin resistance.

Careful consideration should be given to interpreting these results in the current clinical setting due to changes in dosing practices and formulations over time. In these studies which began 4 years ago, we used full dose ritonavir, which is not currently used today. We chose to study full dose ritonavir because insulin resistance and changes in glucose metabolism were reported early in the use of PIs, when full, rather than boosting, doses were used. Ritonavir is more commonly used to boost other PIs, and it is currently not known if boosting doses of ritonavir alter insulin sensitivity. The effects of boosting doses of ritonavir are further complicated by the concomitant effects of the boosted PIs on insulin sensitivity. In the case of lopinavir/ritonavir, we previously found that a single dose decreased insulin sensitivity by 13%, but it is unclear if lopinavir, ritonavir, or both PIs are responsible for the induction of insulin resistance. Amprenavir 1200 mg soft gel capsules are no longer used and have largely been replaced by fosamprenavir, which has yet to be studied with regard to insulin sensitivity.

In summary, we report that a single dose of ritonavir decreased insulin-mediated glucose disposal in healthy normal volunteers, whereas amprenavir has little effect on insulin-mediated glucose disposal under nearly identical study conditions. These studies are placed in context of the known effects of a single dose of indinavir and lopinavir/ritonavir in identical study designs. A single dose of indinavir induced the largest decrease in insulin-mediated glucose disposal. Full-dose ritonavir and lopinavir/ritonavir had lesser effects on insulin-mediated glucose disposal whereas amprenavir had no significant effect on insulin-mediated glucose disposal. These were four separate studies in identical protocols and similar, but not identical, study populations. As commonly used dosages and formulations of PIs have evolved over time and differ from those used in this study, careful consideration should be given to application of these findings to clinical practice.

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Fig. 1.

(a) Drug levels of ritonavir during the euglycemic hyperinsulinemic clamp. (b) Drug levels of amprenavir during the euglycemic hyperinsulinemic clamp.

Fig. 2.

(a) Whole blood glucose levels and rate of glucose infusion during the euglycemic hyperinsulinemic clamp after administration of placebo and ritonavir. (b) Whole blood glucose levels and rate of glucose infusion during the euglycemic hyperinsulinemic clamp after administration of placebo and amprenavir.

Fig. 3.
 (a) Insulin-mediated glucose disposal with placebo and ritonavir administration. (b) Insulin-mediated glucose disposal with placebo and amprenavir administration.

Table 1
Metabolic parameters at baseline and at the start of the euglycemic hyperinsulinemic clamp

	Placebo	Ritonavir	P value	Placebo	Ampronavir	P value
a Baseline parameters (prior to administration of medication)						
<i>n</i>	8			6		
Age	49±5.2			49±7.6		
Weight (kg)	76.3±3.0	76.4±2.8	NS	77.5±2.8	77.5±3.1	NS
Body mass index (kg/m ²)	23.9±0.7	23.9±0.6	NS	24.7±1.0	24.7±1.0	NS
Fasting plasma glucose (mmol/l)	5.1±0.1	5.3±0.1	NS	5.2±0.1	5.2±0.2	NS
Fasting serum insulin (pmol/l)	75.9±8.3	84.4±7.1	NS	60.7±5.1	69.0±4.8	NS
HOMA insulin resistance index	2.4±0.3	2.8±0.2	NS	2.0±0.2	2.2±0.1	NS
Fasting lactate (mmol/l)	0.81±0.05	0.91±0.09	NS	0.84±0.11	0.89±0.11	NS
Total cholesterol (mmol/l)	4.2±0.3	4.5±0.2	NS	4.3±0.2	4.4±0.2	NS
LDL cholesterol (mmol/l)	2.7±0.3	3.0±0.2	NS	2.6±0.2	2.8±0.2	NS
HDL cholesterol (mmol/l)	1.3±0.1	1.3±0.09	NS	1.3±0.1	1.3±0.1	NS
Triglycerides (mmol/l)	0.6±0.1	0.6±0.1	NS	0.7±0.2	0.6±0.1	NS
Free fatty acids (mmol/l)	0.232±0.059	0.208±0.042	NS	0.209±0.021	0.252±0.031	NS
Resting energy expenditure (kcal/day)	1867±117	1881±94	NS	1787±60	1884±94	NS
b Glucose metabolism at the start of the euglycemic hyperinsulinemic clamp (2 h after administration of ritonavir, 1 h after administration of ampronavir)						
Plasma glucose (mmol/l)	5.1±0.1	5.3±0.1	NS	5.4±0.1	5.3±0.2	NS
Serum insulin (pmol/l)	71.3±8.6	103.6±7.1	0.047	84.5±7.2	89.3±9.0	NS
HOMA insulin resistance index	2.3±0.3	3.5±0.2	0.002	2.8±0.2	3.0±0.4	NS

HDL, high-density lipoprotein; HOMA, homeostasis model assessment; LDL, low-density lipoprotein; NS, not significant.