Original Article: Metabolism

Rosiglitazone decreases intra- to extramyocellular fat ratio in obese non-diabetic adults with metabolic syndrome

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Abstract

Background Insulin resistance is intrinsically related to intramyocellular (IMCL) rather than extramyocellular (EMCL) triglyceride content. Conflicting results have been reported on the ability of insulin sensitizer agents, such as thiazolidinediones, to modify muscle fat distribution. The aim of this study was to investigate the role of rosiglitazone on muscle fat compartment distribution in an adult population of obese non-diabetic metabolic syndrome patients.

Patients and methods Fifteen obese, non-diabetic, metabolic syndrome patients were studied by means of proton nuclear magnetic resonance (^{1}H-NMR) spectroscopy before and after treatment with rosiglitazone 8 mg/day for 6 months. Anthropometrical and metabolic variables were assessed.

Results After rosiglitazone, body weight and hip circumference increased [100.9 (91.12–138.7) vs. 107.0 (79.6–142.8) kg and 118 (107–126) vs. 122 (110–131) cm]; while waist–hip ratio (WHR) decreased from 0.93 (0.87–1.00) to 0.89 (0.82–0.97) ($P < 0.001$ for all). Additionally, fasting plasma glucose, insulin and homeostatis model assessment of insulin resistance (HOMA-IR) significantly decreased while adiponectin increased over threefold [9.7 (3.7–17.7) vs. 38.0 (19.3–42.4) μg/ml] without any changes in resistin. Finally, the IMCL did not change [267.54 (213.94–297.94) vs. 305.75 (230.80–424.75) arbitrary units (AU), $P = 0.15$] while the EMCL increased [275.53 (210.39–436.66) vs. 411.39 (279.92–556.59) AU; $P < 0.01$] therefore decreasing the IMCL-to-EMCL (IMCL/EMCL) ratio [1.07 (0.78–1.23) vs. 0.71 (0.53–0.96); $P < 0.01$].

Conclusion Rosiglitazone treatment increased body weight and hip circumference and decreased WHR. More importantly, it decreased the IMCL/EMCL ratio by increasing the EMCL without any significant change on the IMCL.

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Keywords drug treatment, insulin resistance, metabolism

Abbreviations ^{1}H-NMR, proton nuclear magnetic resonance; AMPK, monophosphate-activated protein kinase; AU, arbitrary units; BMI, body mass index; CRP, C-reactive protein; EMCL, extramyocellular trygliceride content; FPG, fasting plasma glucose; HDL, high-density lipoprotein; HOMA, homestasis model assessment; IECV, interassay coefficient of variation; IMCL, intramyocellular trygliceride content; IR, insulin resistance; LDL, low-density lipoprotein; MS, metabolic syndrome; PAI-1, plasminogen activator inhibitor 1; PPAR-γ, peroxisome proliferator-activated receptor- γ; QUICKI, quantitative insulin sensitivity check index; WHR, waist–hip ratio

Introduction

The metabolic syndrome (MS) is considered as a highly prevalent and important clinical entity related to Type 2 diabetes and cardiovascular risk [1–5]. Formerly, it was also known as the insulin resistance syndrome, because of the seminal importance of insulin resistance (IR) as its main pathophysiological basis. IR may, however, be a bystander of more complex mechanisms involving visceral obesity, ectopic fat deposition and dysfunctional adipose tissue [6]. Liver and muscle fat deposits are the main sites for IR-related pathophysiology [7].

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Excessive fatty acids could be deposited as triglycerides at the extramyocellular (EMCL) as well as the intramyocellular (IMCL) compartments. The IMCL is intrinsically related to IR. Indeed, several studies utilizing proton nuclear magnetic resonance (^{1}H-NMR) spectroscopy have confirmed that the size of the IMCL is the main predictor of IR [8,9].

Adipose tissue is now considered as a dynamic endocrine organ, producing several proteins (adipokines). Adiponectin, an adipokine almost exclusively produced by the adipose tissue, is directly related to insulin sensitivity [10–12] and to IMCL in obese adolescents [13]. We, as well as other authors, have recently demonstrated an inverse relationship between IMCL size, but not EMCL, and adiponectin in adults [14,15].

Thiazolidinediones, insulin sensitizer agents acting as peroxisome proliferator-activated receptor-γ (PPAR-γ) agonists, may decrease circulating fatty acids and ameliorate fat distribution, including liver fat reduction [16]. In culture of muscle cells from diabetic patients it has been demonstrated that troglitazone, a formerly marketed PPAR-γ agonist, increased fatty acid oxidation [17], potentially reducing intramyocellular triglycerides. Therefore, a plausible mechanism for its action as an insulin sensitizer may be through reduction in muscle fat deposition. Few studies in humans have looked at this subject and their results are conflicting, showing increases in EMCL after rosiglitazone [18], decreasing levels in IMCL after pioglitazone and metformin [19] and, finally, decrements in IMCL after pioglitazone, but not after metformin [20]. It is possible that some methodological differences may have contributed to reported results.

Therefore, our aim was to investigate if rosiglitazone, used for a longer period of time, in a group of non-diabetic, MS patients, not taking other drugs acting on carbohydrate metabolism, influences muscle fat distribution.

Patients and methods

Sixteen obese adults [body mass index (BMI) 38.0 ± 5.7 kg/m^2; aged 41.4 ± 8.7 years; 11 females], MS patients, defined by the National Cholesterol Education Treatment Program—Adult Treatment Panel III (NCEP-ATPIII) [21] were selected at the Cardiometabolic Clinic for outpatient care of the State University of Rio de Janeiro. Rosiglitazone in a dose of 8 mg daily was administered to all volunteers for a 6-month period. Initially, the sample size was inferred based on Mayerson's study [18], but we have almost doubled their sample size and extended the follow-up period by an additional 3 months. In order to avoid changes in fat deposition, patients were advised to maintain their usual diet and lifestyle throughout the study. Exclusion criteria included diabetes, smoking and previous cardiovascular, kidney or liver diseases. Patients taken drugs known to affect glucose or lipid metabolism were excluded. All subjects gave written informed consent and the local Ethical Committee approved the protocol.

Anthropometric measurements and blood pressure

Height, weight, waist and hip circumferences, as well as blood pressure were collected by the same trained examiner as previously reported [22]. BMI was defined as the ratio between weight in kilograms and squared height in meters.

Waist circumference was obtained by measuring the narrowest point midway between the iliac crest and the lower costal margin. Hip circumference was measured at the largest diameter of the gluteal region. Waist–hip ratio (WHR) was determined by dividing the waist by the hip circumference. Supine blood pressure was measured twice after a 15-min rest using an automatic sphygmomanometer (multi-parameter patient monitor—LifeWindow LW6000; Digicare Biomedical Technology, West Palm Beach, WA, USA).

Assessment of insulin sensitivity and biochemical analysis

All patients underwent an oral glucose tolerance test using 75 g anhydrous glucose. Results were used to classify glucose tolerance state. As stated before, patients with Type 2 diabetes mellitus were excluded, but those with impaired glucose tolerance (IGT) were allowed to participate. Blood samples were collected after a 12-h overnight fast. All laboratory measurements were performed in duplicate using an automated method (Modular Analytics PP; Roche, Basel, Switzerland). Fasting plasma glucose (FPG), total cholesterol, triglycerides and high-density lipoprotein (HDL) cholesterol were measured respectively, by enzyme-colourimetric GOD-PAP [interassay coefficient of variation (IECV) = 1.09%], enzymatic GPO-PAP (IECV = 2.93%), enzymatic GPO-PAP (IECV = 1.29%) and enzyme colourimetric without pre-treatment (IECV = 3.23%). Plasma low-density lipoprotein (LDL) cholesterol was calculated according to Friedwald equation. Fasting plasma insulin was measured by automated chemoluminiscence (IECV = 4.68%). Fibrinogen (IECV = 4.33%) and high-sensitivity C-reactive protein (CRP) (IECV = 2.66%) were measured respectively by coagulometric and imunoturbidimetry methods on Modular Analytics P (Roche). Blood samples were centrifuged and stored at $-70°C$ for further analysis of adipokines. Adiponectin, resistin and plasminogen activator inhibitor 1 (PAI-1) were measured by human serum adipokine (panel A, Lincoplex kit CAT, HADK1, 61K-A; Linco Research, St Charles, MO, USA). Intra- and IECVs were 6.11 and 13.2%, 7.26 and 9.12%, 4.37 and 20.8%, respectively [22]. Homeostasis model assessment (HOMA) [23] and the quantitative insulin sensitivity check index (QUICKI) [24] were calculated to assess insulin resistance.

Assessment of muscle lipids

After an overnight fast, ^{1}H-NMR spectroscopy of the right soleus muscle was performed using a 1.5T MR Scanner (Magneton Vision; Siemens, Erlangen, Germany). Subjects were instructed to avoid strenuous physical exercise for at least 2 days before the examination. They were positioned

supine with their right lower leg at the centre of the coil. Thus, the tibia was oriented nearly parallel to the static magnetic field. The coil centre was approximately 10–15 cm below the knee joint. The volume of interest (13 × 13 × 30 mm^3) was centred within the soleus muscle and placed to avoid vascular structures and gross adipose tissue deposits. Spectra were acquired by point-resolved spectroscopy (PRESS) sequence with the following parameters: echo time 135 or 270 ms, repetition time 1600 ms and 128 or 256 scans with water suppression. IMCL was calculated from peak areas of IMCL methylene (CH$_2$) between 1.2 and 1.3 µg/g. EMCL was calculated from EMCL methylene between 1.4 and 1.5 µg/g. The creatine signal between 2.9 and 3.1 µg/g served as an internal reference for IMCL and EMCL quantification.

Statistical analysis

All group data are reported as median (first to third quartiles), unless otherwise stated, and were analysed by Prism 4.01 (Graphpad Inc., San Diego, CA, USA). The pooled group was divided into two subgroups, according to quartiles of weight gain. Comparison r within group at baseline and after treatment period was performed using Wilcoxon matched-pairs test. Significant differences were assumed to be present at $P < 0.05$.

Results

After 6 months, 15 patients ended the treatment period and had ^{1}H-NMR spectroscopy performed. Therefore, data included are for those who completed the whole study protocol.

Table 1 depicts data for all patients included in the final analysis. After 6 months of rosiglitazone treatment there was a significant increase in weight, BMI and hip circumference without change in waist circumference. Consequently, WHR decreased after treatment [0.93 (0.87–1.0) vs. 0.89 (0.82–0.97); $P < 0.001$].

Changing patterns of muscle triglyceride distribution after rosiglitazone was our main objective. IMCL did not change but EMCL significantly increased (Table 1). There was also an important increase in body weight and the pooled group was subsequently divided into higher (fourth quartile; i.e. ≥ 4.1 kg) and lower weight gainers during treatment. Once again, even in those who gained more than 4.0 kg there was no increase on IMCL (Fig. 1). In contrast, EMCL significantly increased in both groups. The IMCL to EMCL (IMCL/EMCL) ratio was then calculated in the pooled group at baseline and after treatment. This ratio significantly decreased after rosiglitazone [1.07 (0.78–1.23) vs. 0.71 (0.53–0.96); $P < 0.01$, Fig. 2]. Regarding glycaemic status, nine patients had impaired glucose tolerance. All studied variables related to carbohydrate metabolism showed a consistent modification, where FPG, insulin and HOMA-IR decreased and QUICKI increased. Adiponectin increased over threefold above basal level [9.7 (3.7–17.7) vs. 38.0 (19.3–42.4) µg/ml; $(P < 0.001)$], while resistin levels were unchanged (Table 1). There were small but significant increases in total

and LDL cholesterol, but HDL cholesterol and triglycerides did not change (Table 1). Finally, a significant $\sim 70\%$ reduction in CRP level was achieved [1.0 (0.5–2.3) to 0.3 (0.2–0.5) mg/l; $P < 0.0001$], while fibrinogen, but not PAI-1, decreased significantly (Table 1).

Rosiglitazone was well tolerated and only one patient developed minor pre-tibial oedema. Weight gain was the major side effect noticed.

Discussion

The main finding of this study was an increase in fat deposition in the extramyocellular muscle compartment after 6 months of treatment with an agonist of the PPAR- γ, rosiglitazone. However, this was not observed at the intramyocellular compartment, where triglyceride content was unchanged. Therefore, the IMCL/EMCL ratio significantly decreased (Fig. 2). It seems that there is a small increase in IMCL after rosiglitazone. Indeed, the median and third quartiles of IMCL increased by 14 and 42%, respectively. However, this was far from being statistically significant. Possibly a larger sample size in future studies may clarify this point.

Although augmentation of fatty acid disposal in skeletal muscle from diabetic patients has been demonstrated *in vitro* [17], potentially reducing IMCL, clinical studies in humans have shown contradictory results. Indeed, Mayerson and co-workers [18] studying diabetic patients taking rosiglitazone for only 3 months could not demonstrate any improvement in IMCL. Instead, a 39% increase in EMCL was achieved. Their results are in accordance with our findings, i.e. a 49.3% increase in EMCL. In contrast to the present study, there was no weight gain in Mayerson's group. Because of the massive weight gain achieved by our patients, they were divided into those who gained 4.1 kg or more (fourth quartile of weight gain) and those who did not. It then became apparent that IMCL did not change substantially in either group, whilst EMCL increased, even in those who gained < 4.1 kg. Thus, there was a significant decrease in the IMCL/EMCL ratio (Fig. 2). One study in Zucker fatty rats also showed similar results, i.e. a decrease in the IMCL/EMCL ratio with rosiglitazone [25].

Teranishi and co-workers [19] succeeded in demonstrating decreased IMCL after pioglitazone or metformin in a group of Type 2 diabetic patients. In contrast to our study, however, weight did not increase with pioglitazone and decreased slightly in the metformin group. This may suggest that lifestyle modification, including exercises and dietary changes, implemented before and maintained during the intervention period, as stated by the authors, influenced their results. Rasouli and co-workers [20] used muscle biopsy rather than ^{1}H-NMR spectroscopy in a group of glucose intolerant otherwise healthy subjects treated with pioglitazone or metformin for 10 weeks. There was a 34% decrease in IMCL with pioglitazone but no change within the metformin group. A modest increase in body weight (2.63 ± 0.65 kg) was accompanied by a decrease in

Table 1 Clinical and laboratory parameters of investigated subjects

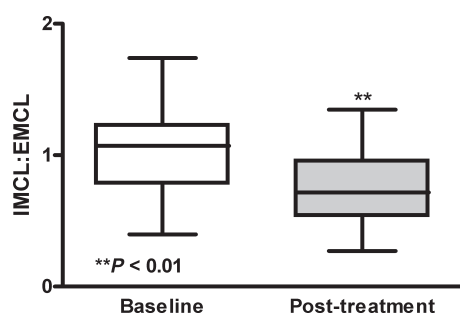|	Baseline	After rosiglitazone treatment
Weight (kg)	100.9 (91.12–138.7)	107.0 (79.6–142.8)‡
BMI (kg/m^2)	38.1 (32.8–44.3)	38.4 (34.6–48.4)‡
Waist (cm)	106.0 (98.0–112.0)	109.0 (101.0–114.0)
Hip (cm)	118 (107–126)	122 (110–131)‡
WHR	0.93 (0.87–1.00)	0.89 (0.82–0.97)‡
FPG (mmol/l)	5.7 (5.3–6.1)	5.2 (4.7–5.6)*
Insulin (pmol/l)	88 (64–122)	54 (43–77)‡
HOMA-IR	3.30 (2.47–4.37)	2.00 (1.61–3.16)‡
QUICKI	0.320 (0.307–0.333)	0.343 (0.321–0.355)‡
Systolic BP (mmHg)	141 (127.5–170)	134 (128.5–157.5)
Diastolic BP (mmHg)	85 (80–95)	83 (79.5–90)
Total cholesterol (mmol/l)	4.9 (4.45–6.03)	5.2 (4.91–6.85)†
LDL cholesterol (mmol/l)	3.0 (2.46–4.22)	3.5 (3.15–4.73)†
HDL cholesterol (mmol/l)	1.1 (0.96–1.19)	1.1 (0.91–1.25)
Triglycerides (mmol/l)	2.0 (1.11–2.38)	1.7 (1.34–2.38)
C-reactive protein (mg/l)	1.0 (0.5–2.3)	0.3 (0.2–0.5)‡
Fibrinogen (mg/dl)	304 (254.2–364.0)	265 (209.7–296.5)*
Adiponectin (µg/ml)	9.7 (3.7–17.7)	38.0 (19.3–42.4)‡
PAI-1 (ng/ml)	23.8 (21.2–28.1)	22.5 (20.6–24.7)
Resistin (ng/ml)	17.3 (17.2–18.2)	17.1 (16.9–17.8)
EMCL (AU)	276 (210–437)	411 (280–557)†
IMCL (AU)	268 (214–298)	306 (231–425)

*$P < 0.05$, †$P < 0.01$, ‡$P < 0.001$.

AU, arbitrary units; BMI, body mass index; BP, blood pressure; EMCL, extramyocellular trygliceride content; FPG, fasting plasma glucose; HDL, high-density lipoprotein; HOMA-IR, homeostasis model assessment of insulin resistance; IMCL, intramyocellular trygliceride content; LDL, low-density lipoprotein; PAI-1, plasminogen activator inhibitor 1; QUICKI, quantitative insulin sensitivity check index; WHR, wasit–hip ratio.
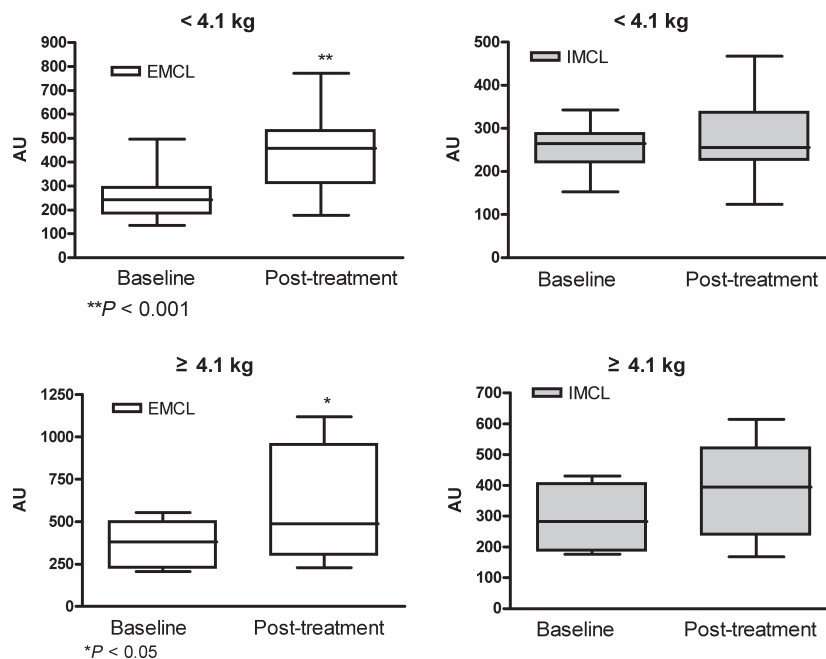
FIGURE 1 Intra- (IMCL) and extramyocellular (EMCL) lipid content at baseline and after 6 months of rosiglitazone treatment (post-treatment) divided according to weight gain during follow-up. AU, arbitrary units.

FIGURE 2 Intra- (IMCL) to extramyocellular (EMCL) ratio decrement after rosiglitazone treatment in non-diabetic obese adults with metabolic syndrome. $P < 0.01$.

visceral-to-subcutaneous fat ratio. Therefore, differences in methodology (muscle biopsy), in weight changes, periods of treatment or even in drug characteristics may explain our contradictory results.

A few possible interpretations can be raised from the present findings: (i) the excess fat was shunted to the extramyocellular compartment, which may behave as peripheral adipocytes accumulating triglycerides under the influence of thiazolidinediones [18]; (ii) the excess fat eventually driven into the intramyocellular compartment has been oxidized by PPAR-γ activation [20] or (iii) fat diversion from ectopic lipid depots into the subcutaneous depot [18,20,26,27]. Moreover, rosiglitazone therapy increases adipocyte sensitivity to insulin, thus decreasing peripheral fat lipolysis [18]. In accordance, in our group of patients, the massive increase in body weight was accompanied by modification of indirect measures of body fat distribution, namely an increase in hip circumference and a decrease in WHR.

In animals, adiponectin increases expression of genes involved in fatty acid transportation and oxidation, such as acyl-CoA oxidase, CD36 and uncoupled protein [28]. More importantly, administration of adiponectin (ADP) increased fat oxidation in muscle, decreased muscle triglycerides and ameliorated insulin sensitivity [28]. Thiazolidinediones activate adenosine monophosphate-activated protein kinase (AMPK) and increase serum concentration of adiponectin, which also activates AMPK in both muscle and liver [28,29]. The adiponectin level significantly increased during our study and could be responsible for increased muscle fat oxidation. However, as the IMCL did not decrease, such a relationship could not be demonstrated. Moreover, there was no correlation between the IMCL/EMCL ratio decrement and adiponectin increment (data not shown).

Regarding the modification of the entire profile of metabolic variables, such as FPG, insulin, HOMA-IR, QUICKI and fibrinogen, as well as of an inflammatory marker (PCR) and the lipid profile, this study is entirely in accordance with the spectrum of action of thiazolidinediones [16]. It is noteworthy that even in an experimental free living situation, where patients gained a substantial amount of weight, treatment with rosiglitazone modified all those markers of insulin sensitivity

towards normal, while ameliorating inflammatory markers. This may be as a result of favourable fat deposition, as suggested by the observed increase in body weight and hip circumference accompanied by decreasing WHR. Accordingly, in Type 2 diabetic patients, rosiglitazone decreases insulin resistance while increasing leptin, a marker of total body fat. Interestingly, an increase in maximum subcutaneous fat thickness after rosiglitazone, measured by sonography, correlated positively with leptin augmentation and negatively with HOMA-IR [30]. Taken together, amelioration of metabolic parameters may be a result of decreased IR secondary to peripheral fat deposition.

This study has a number of limitations: ideally, when a drug intervention is tested, a control, placebo-treated group should be included. However, in such a group of MS patients this could not be performed. However, the robust amelioration observed in the majority of metabolic parameters, adiponectin and CRP, as well as a remarkable (235%) improvement in endothelial function previously published in the same group [22], support our conclusions. Additionally, a before-and-after approach means that the treated group served as their own control. Finally, our data should not be generalized beyond the population studied because of the small sample size, tested by non-parametric methods. Heterogenity of our sample should be considered only on glycaemic status. However, our aim was not to investigate differences according to such subgroups as this would need a bigger sample size. We had not formally checked for adherence to our instructions regarding diet or physical activity. However, our patients gained a huge amount of weight, beyond what could be expected by rosiglitazone treatment, implying that they did not diet or exercise significantly. Such observations suggest that an isocaloric diet could have minimized the weight gain.

This study has also a number of advantages. This is the longest (6 months) study of those looking at thiazolidinedione intervention and muscle fat. We investigated a homogeneous population of obese MS adults, although with differences in glycaemic status as discussed above. A population of patients not using drugs that could interfere with glucose or fat metabolism is an additional advantage.

In conclusion, in a group of obese non-diabetic metabolic syndrome patients, treatment with rosiglitazone for 6 months increased body weight but improved several metabolic and inflammatory markers. The IMCL/EMCL ratio decreased, as extramyocellular fat increased without an affect on the intramyocellular fat compartment.

Competing interests

AFG-M, LRB, MT and BG are members of the Advisory Board for GSK in Brazil and have received fees for giving talks. For all the others authors, there is nothing to declare.

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References

1 Ford ES, Giles WH, Dietz WH. Prevalence of the metabolic syndrome among US adults: findings from the third National Health and Nutrition Examination Survey. *J Am Med Assoc* 2002; **287**: 356–359.

2 Ford ES, Giles WH. A comparison of the prevalence of the metabolic syndrome using two proposed definitions. *Diabetes Care* 2003; **26**: 575–581.

3 Lakka HM, Laaksonen DE, Lakka TA, Niskanen LK, Kumpusalo E, Tuomilehto J *et al*. The metabolic syndrome and total and cardiovascular disease mortality in middle-aged men. *J Am Med Assoc* 2002; **288**: 2709–2716.

4 Cornier MA, Dabelea D, Hernandez TL, Lindstrom RC, Steig AJ, Stob NR *et al*. The metabolic syndrome. *Endocr Rev* 2008; **29**: 777–822.

5 Lorenzo C, Williams K, Hunt KJ, Haffner SM. The National Cholesterol Education Program—Adult Treatment Panel III, International Diabetes Federation, and World Health Organization definitions of the metabolic syndrome as predictors of incident cardiovascular disease and diabetes. *Diabetes Care* 2007; **30**: 8–13.

6 Despres JP, Brewer HB. Metabolic syndrome: the dysmetabolic state of dysfunctional adipose tissue and insulin resistance. *Eur Heart J* 2008; **10**: B1–B3.

7 Bays H, Mandarino L, DeFronzo RA. Role of the adipocyte, free fatty acids, and ectopic fat in pathogenesis of type 2 diabetes mellitus: peroxisomal proliferator-activated receptor agonists provide a rational therapeutic approach. *J Clin Endocrinol Metab* 2004; **89**: 463–478.

8 Jacob S, Machann J, Rett K, Brechtel K, Volk A, Renn W *et al*. Association of increased intramyocellular lipid content with insulin resistance in lean nondiabetic offspring of type 2 diabetic subjects. *Diabetes* 1999; **48**: 1113–1119.

9 Virkamaki A, Korsheninnikova E, Seppala-Lindroos A, Vehkavaara S, Goto T, Halavaara J *et al*. Intramyocellular lipid is associated with resistance to *in vivo* insulin actions on glucose uptake, anti-lipolysis, and early insulin signaling pathways in human skeletal muscle. *Diabetes* 2001; **50**: 2337–2343.

10 Chandran M, Phillips SA, Ciaraldi T, Henry RR. Adiponectin: more than just another fat cell hormone? *Diabetes Care* 2003; **26**: 2442–2450.

11 Weyer C, Funahashi T, Tanaka S, Hotta K, Matsuzawa Y, Pratley RE *et al*. Hypoadiponectinemia in obesity and type 2 diabetes: close association with insulin resistance and hyperinsulinemia. *J Clin Endocrinol Metab* 2001; **86**: 1930–1935.

12 Sheng T, Yang K. Adiponectin and its association with insulin resistance and type 2 diabetes. *J Genet Genomics* 2008; **35**: 321–326.

13 Weiss R, Dufour S, Groszmann A, Petersen K, Dziura J, Taksali SE *et al*. Low adiponectin levels in adolescent obesity: a marker of increased intramyocellular lipid accumulation. *J Clin Endocrinol Metab* 2003; **88**: 2014–2018.

14 Thamer C, Machann J, Tschritter O, Haap M, Wietek B, Dahl D *et al*. Relationship between serum adiponectin concentration and intramyocellular lipid stores in humans. *Horm Metab Res* 2002; **34**: 646–649.

15 Godoy-Matos AF, Bahia LR, Domingues RC, Sicuro FL, Tambascia M, Geloneze B *et al*. Adiponectin is realted to intramyocellular lipid content in non-diabetic adults. *J Endocrinol Invest* 2009; **July 28** [Epub ahead of print].

16 Yki-Jarvinen H. Thiazolidinediones. *N Engl J Med* 2004; **351**: 1106–1118.

17 Cha BS, Ciaraldi TP, Park KS, Carter L, Mudaliar SR, Henry RR. Impaired fatty acid metabolism in type 2 diabetic skeletal muscle cells is reversed by PPARγ agonists. *Am J Physiol Endocrinol Metab* 2005; **289**: E151–E159.

18 Mayerson AB, Hundal RS, Dufour S, Lebon V, Befroy D, Cline GW *et al*. The effects of rosiglitazone on insulin sensitivity, lipolysis, and hepatic and skeletal muscle triglyceride content in patients with type 2 diabetes. *Diabetes* 2002; **51**: 797–802.

19 Teranishi T, Ohara T, Maeda K, Zenibayashi M, Kouyama K, Hirota Y *et al*. Effects of pioglitazone and metformin on intracellular lipid content in liver and skeletal muscle of individuals with type 2 diabetes mellitus. *Metabolism* 2007; **56**: 1418–1424.

20 Rasouli N, Raue U, Miles LM, Lu T, Di Gregorio GB, Elbein SC *et al*. Pioglitazone improves insulin sensitivity through reduction in muscle lipid and redistribution of lipid into adipose tissue. *Am J Physiol Endocrinol Metab* 2005; **288**: E930–E934.

21 Third Report of the National Cholesterol Education Program (NCEP). Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III) final report. *Circulation* 2002; **106**: 3143–3421.

22 Bahia L, Aguiar LG, Villela N, Bottino D, Godoy-Matos AE, Geloneze B *et al*. Adiponectin is associated with improvement of endothelial function after rosiglitazone treatment in non-diabetic individuals with metabolic syndrome. *Atherosclerosis* 2007; **195**: 138–146.

23 Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF, Turner RC. Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. *Diabetologia* 1985; **28**: 412–419.

24 Katz A, Nambi SS, Mather K, Baron AD, Follmann DA, Sullivan G *et al*. Quantitative insulin sensitivity check index: a simple, accurate method for assessing insulin sensitivity in humans. *J Clin Endocrinol Metab* 2000; **85**: 2402–2410.

25 Jucker BM, Yang D, Casey WM, Olzinski AR, Williams C, Lenhard SC *et al*. Selective PPARδ agonist treatment increases skeletal muscle lipid metabolism without altering mitochondrial energy coupling: an *in vivo* magnetic resonance spectroscopy study. *Am J Physiol Endocrinol Metab* 2007; **293**: E1256–E1264.

26 Kim JK, Fillmore JJ, Gavrilova O, Chao L, Higashimori T, Choi H *et al*. Differential effects of rosiglitazone on skeletal muscle and liver insulin resistance in A-ZIP/F-1 fatless mice. *Diabetes* 2003; **52**: 1311–1318.

27 Kelly IE, Han TS, Walsh K, Lean ME. Effects of a thiazolidinedione compound on body fat and fat distribution of patients with type 2 diabetes. *Diabetes Care* 1999; **22**: 288–293.

28 Yamauchi T, Kamon J, Minokoshi Y, Ito Y, Waki H, Uchida S *et al*. Adiponectin stimulates glucose utilization and fatty-acid oxidation by activating AMP-activated protein kinase. *Nat Med* 2002; **8**: 1288–1295.

29 Fryer LG, Parbu-Patel A, Carling D. The anti-diabetic drugs rosiglitazone and metformin stimulate AMP-activated protein kinase through distinct signaling pathways. *J Biol Chem* 2002; **277**: 25226–25232.

30 Kim HJ, Kim SK, Shim WS, Lee JH, Hur KY, Kang ES *et al*. Rosiglitazone improves insulin sensitivity with increased serum leptin levels in patients with type 2 diabetes mellitus. *Diabetes Res Clin Pract* 2008; **81**: 42–49.

Supporting Information

Additional Supporting Information may be found in the online version of this article:

Table S1. Differences observed after RSG treatment in IGT patients.

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