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Short-term metabolic and haemodynamic effects of GR79236 in normal and fructose-fed rats

Xianqin Qu [a], Gregory Cooney [a,b], Richard Donnelly [a,c,*]

[a] *Department of Pharmacology, University of Sydney, Sydney, Australia*
[b] *Department of Endocrinology, Royal Prince Alfred Hospital, Sydney, Australia*
[c] *School of Medical and Surgical Sciences, University of Nottingham, Nottingham, UK*

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Abstract

The adenosine (A1) receptor agonist, GR79236 (*N*-[(1*S*,*trans*)-2-hydroxycyclopentyl]adenosine), inhibits catecholamine-induced lipolysis in vitro, but the short-term metabolic and haemodynamic effects have not been previously reported in the fructose fed model of insulin resistance, dyslipidaemia and hypertension. This study reports the effects of GR79236 (1 mg/kg/day for 8 days) on nonesterified free fatty acid and triglyceride metabolism, oral and i.v. glucose tolerance, blood pressure and heart rate, and insulin sensitivity, in normal rats and rats fed a fructose-enriched diet. In normal rats, GR79236 significantly reduced fasting glucose (25%), free fatty acid (50%) and triglyceride (55%) concentrations, and improved glucose tolerance (AUC[glu] 21.2 ± 1.3 vs. 16.5 ± 1.1 mmol h/l, $p < 0.05$). Fructose feeding induced a state of insulin resistance and dyslipidaemia, as shown by an increase in steady-state plasma glucose levels (7.1 vs. 6.1 mmol/l), impaired i.v. glucose tolerance and a 3-fold rise in fasting triglyceride levels; fructose-fed rats also developed a significant increase in blood pressure. GR79236 ameliorated the effects of fructose feeding on fatty acid and triglyceride levels, and blood pressure, and improved i.v. glucose tolerance in fructose-fed rats. The hypotriglyceridaemic effect was due to a reduction in triglyceride secretion rate (17.3 ± 1.7 vs. 30.2 ± 1.1). Thus, in normal rats and in a dietary-induced rodent model of insulin resistance, dyslipidaemia and hypertension, GR79236 has lipid-lowering and glucose-lowering activity, as well as haemodynamic effects, which are potentially useful for treating both the metabolic and haemodynamic features of insulin resistance and NIDDM in humans.

Keywords: GR79236; Insulin resistance; Lipolysis; Fructose; Triglyceride; Non-insulin-dependent diabetes (NIDDM)

1. Introduction

The metabolic features of non-insulin-dependent diabetes mellitus (NIDDM) include hyperglycaemia, dyslipidaemia and impaired pancreatic insulin secretion. As part of the pathogenesis of NIDDM, skeletal muscle, liver and adipose tissues become resistant to the hormonal effects of insulin, which in turn leads to decreased insulin-mediated glucose disposal, hepatic glucose overproduction and a marked increase in lipolysis (DeFronzo et al., 1992). The increased rate of production of non-esterified free fatty acids enhances very-low-density-lipoprotein triglyceride synthesis in the liver, but according to Randle's hypothesis of the glucose–fatty acid cycle (Randle et al., 1963), increased concentrations of free fatty acids also exacerbate the hyperglycaemia of NIDDM via two biochemical mechanisms: (1) enhanced fatty acid availability increases lipid oxidation, which in turn inhibits both oxidative and non-oxidative glucose utilization (Lillioja et al., 1985; Ebeling and Koivisto, 1994); and (2) high fatty acid concentrations increases hepatic glucose overproduction (Saloranta et al., 1993; Boden et al., 1994). Evidence in support of the glucose–fatty acid cycle comes from several studies, both in vitro and in vivo, showing that acute reductions in free fatty acid levels improves insulin-mediated glucose disposal (Reaven et al., 1988a,b; Lee et al., 1996), but the extent to which fatty acid-lowering drugs produce hypoglycaemic effects in NIDDM remains uncertain, in part because traditional antilipolytic agents such as nicotinic acid and acipimox achieve only modest reductions in plasma free fatty acid concentrations and are poorly tolerated.

Adenosine has important metabolic and cardiovascular

* Corresponding author. Present address: University Department of Medicine, Division of Vascular Medicine, Derbyshire Royal Infirmary NHS Trust, Derby DE1 2QY, UK. Fax: (44-1332) 254-938; e-mail: richard.donnelly@btinternet.com

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effects in rats and humans. Extracellular purinoceptors with high affinity for adenosine have been classified into at least 4 subtypes: adenosine-1 (A_1), adenosine-2 (A_{2A} and A_{2B}) and adenosine-3 (A_3) receptors (Fredholm et al., 1994). The antilipolytic effects of adenosine are mediated by adenosine A_1-receptors on the adipocyte plasma membrane, while the cardiovascular effects (bradycardia and hypotension) are mediated by cardiac (A_1) and vascular (A_2) receptors (Olsson and Pearson, 1990). In adipose tissue and skeletal muscle, adenosine A_1 receptors predominate over A_2 (Challis et al., 1992) and there is evidence of defective adenosine A_1-receptor function in genetic models of obesity and insulin resistance (LaNoue and Martin, 1994).

GR79236 (*N*-[(1*S*,*trans*)-2-hydroxycyclopentyl]adenosine) is a new, orally-active adenosine A_1-receptor agonist which has been shown to inhibit catecholamine-induced lipolysis in isolated adipocytes from humans, rats and dogs (Strong et al., 1993; Merkel et al., 1995). The single-dose effects of GR79236 have been reported in normal rats (Gardner et al., 1994), pithed rats and dogs (Strong et al., 1993), and in a rodent model of diabetic ketoacidosis (Thompson et al., 1994). The purpose of this study was to evaluate the short-term metabolic and cardiovascular effects of GR79236 in normal rats and rats made insulin resistant by feeding a fructose-enriched diet. The fructose-fed rat is a well-established non-obese model of dietary-induced insulin resistance characterized by decreased insulin-mediated glucose uptake, hypertriglyceridaemia and hypertension (Tobey et al., 1982; Hwang et al., 1987).

2. Materials and methods

2.1. Materials

Male Sprague–Dawley rats were obtained from The Combined University Laboratory Animal Supply (NSW, Australia). The following reagents were purchased from Sigma, St. Louis, MO, USA: Triton WR1339, GPO-Trinder kits for measurement of serum glucose and triglycerides. The primary anti-rat insulin antibody was purchased from Linco Research (St. Louis, MO, USA) and plasma NEFAs were measured using a commercial kit (Wako, Osaka, Japan). The fructose diet (TD78463) was purchased from Teklad (Madison, WI, USA).

2.2. General

A detailed protocol of all experimental procedures was approved by the Animal Care Ethics Committee of the University of Sydney. Male Sprague–Dawley rats, initially weighing 180–200 g, were used for all experiments; they were maintained on a 12 h light/dark cycle (lights on 06.00–18.00 h) under constant temperature (21°C) and with ad libitum access to food and water. Normal rats were fed standard laboratory chow throughout the study, while animals in the fructose-fed group were switched to a pelleted, high-fructose diet containing 66% fructose, 22% protein and 12% fat. Fructose-fed rats were established on the diet for 10 days before starting treatment with GR79236 or vehicle, and fructose feeding was continued for the duration of drug therapy. In a series of studies, animals were randomly divided into two groups to receive GR79236 1 mg/kg body weight (b.wt) per day ($n = 56$) or vehicle (water) ($n = 56$) by oral gavage, given at 08.00 h each day for 8 days. All experimental procedures were performed on the eighth day, 2 h after the final dose of GR79236 (or vehicle). Tail-vein blood samples were collected from fasting rats for measurement of serum glucose, insulin, free fatty acid and triglyceride concentrations.

2.3. Oral glucose tolerance

Oral glucose tolerance tests (3 g/kg b.wt) were performed in conscious, chow-fed rats before and after 8 days treatment with GR79236 (or vehicle). Following an overnight fast, tail-vein blood samples were collected at baseline, and at 30, 60, 90 and 120 min after administration of glucose by oral gavage.

2.4. Intravenous glucose tolerance

After 6 days treatment with GR79236 or vehicle, fructose-fed rats were anaesthetised (ketamine 100 mg/kg b.wt and xylazine 10 mg/kg b.wt) prior to insertion of an indwelling cannula in the right jugular vein, which was tunneled and exteriorized posteriorly and regularly flushed with 0.2 ml heparin saline (50 U/ml). Animals were allowed to recover for 3 days (meanwhile the fructose diet and GR79236 were continued) before an i.v. glucose tolerance test was performed. Fasted rats were given 0.5 g/kg b.wt of glucose i.v., followed by saline to flush the cannula, and blood samples were drawn at −10, 0, 2, 5, 10, 15 and 30 min after glucose administration.

2.5. Total triglyceride secretion rate

Total triglyceride secretion rate was measured according to the method of Otway and Robinson (1967a,b), as described in detail previously by our group (Donnelly et al., 1994). In brief, triglyceride secretion rate is proportional to the rate of increase in plasma triglyceride concentrations following injection of Triton WR1339, which inhibits lipoprotein lipase-mediated triglyceride clearance. Rats were wrapped in a towel and a 0.5 ml blood sample was obtained from an incision at the tip of the tail. The cut was sealed with laboratory tape, and then Triton (300 mg/ml) injected into the proximal tail vein in a dose of 800 mg/kg b.wt. Animals were returned to their cages,

and subsequent blood samples of 0.2 ml were obtained at 60 and 120 min for determination of serum triglyceride concentration. Rats were then anaesthetized for determination of plasma volume by central injection of ^{125}I-labeled albumin, as described previously (Otway and Robinson, 1967b). The triglyceride secretion rate is calculated as follows:

$$\text{Triglyceride Secretion Rate} = \frac{(TG_{120\,min} - TG_{0\,min}) \times PV}{2\ h}$$

where TG_0 and TG_{120min} represent triglyceride levels at baseline and 2 h, and PV is plasma volume.

2.6. Insulin suppression test

In vivo insulin sensitivity in GR79236-treated versus control rats was evaluated using a constant-rate infusion of insulin (4 mU/kg b.wt per min), glucose (11 mg/kg per min) and somatostatin (300 pmol/kg per min) in 1% bovine serum albumin, 140 mM NaCl and 5 mM KCl, as described previously (Mondon and Reaven, 1988). Animals were fasted overnight and anaesthetised prior to insertion of a cannula in the right jugular vein. The infusion was administered at a rate of 1 ml/h for 180 min using a Harvard infusion pump, and blood samples were collected from the tip of the tail at 0, 160, 170 and 180 min to calculate steady-state plasma glucose and steady-state plasma insulin concentrations (Mondon and Reaven, 1988).

2.7. Blood pressure and heart rate

The following procedure was used for measurement of blood pressure and heart rate, as described previously by our group (Donnelly et al., 1995). All animals were familiarised with the procedure on 2 consecutive training days prior to actual measurements being recorded (and averaged) on each of the following 2 days. The 4-day protocol for blood pressure and heart rate measurement was performed at baseline and after drug treatment. On each day, rats were removed from the animal house and taken to the laboratory at 9 a.m. where they were allowed free access to diet and water and kept in a quiet area prior to blood pressure measurement at 1 p.m. In brief, the tail-cuff method, without external preheating, was used to measure the systolic blood pressure (Recorder 8006 Ugo Basile, Milan, Italy), while the ambient temperature was maintained at 30°C (Hwang et al., 1987; Donnelly et al., 1995). Systolic blood pressure and heart rate were measured in the conscious state, and previous work has shown that blood pressure measurements obtained with this technique correlate well with those obtained by direct arterial cannulation (Buñag, 1973). The mean of 5 consecutive readings was used as the measurement of systolic blood pressure on each of the 2 days. Blood pressure and heart rate recordings over the 2 days before and after treatment were averaged and used for statistical comparisons.

2.8. In vivo lipid synthesis

The rate of lipogenesis was determined in GR79236-treated and control rats by measuring the in vivo incorporation of ^{3}H into fatty acids, as described previously (Stansbie et al., 1976). In brief, ^3H_2O (20 μCi/g b.wt) was injected i.p. and rats were exsanguinated 1 h later. Blood was collected from the heart (to measure plasma-specific radioactivity), and liver, epididymal fat pad, white adipose tissue and interscapular brown adipose tissue samples were collected and snap-frozen in liquid N_2 prior to assay for ^{3}H-fatty acids. Tissue samples (200–500 mg) were weighed and saponified with 3 ml of 1 M NaOH at 70°C for 60 min, followed by addition of 3 ml of 95% ethanol and heating for another 60 min before cooling to room temperature. The fatty acids were then precipitated from the saponified tissue mixture by adding 0.5 ml of 9 M H_2SO_4, and extracted from the aqueous phase by shaking in 3×5 ml volumes of petroleum spirit and washing with 5 ml of dH_2O. The organic extract was evaporated in a fume cupboard and the residue dissolved in 5 ml of scintillation fluid and counted. The rate of fatty acid synthesis was calculated in μg atoms of ^{3}H incorporated per hour per gram of tissue (^{3}H/h per g).

Table 1
Effects of GR79236 on body weight, haemodynamics and fasting metabolic parameters in normal rats

	GR79236 ($n = 12$)		Vehicle ($n = 12$)	
	baseline	day 8	baseline	day 8
Body weight (g)	193 ± 4	232 ± 6	196 ± 9	232 ± 10
Serum glucose (mmol/l)	6.1 ± 0.2	4.6 ± 0.3[a]	6.2 ± 0.2	6.3 ± 0.4
Serum nonesterified free fatty acids (mmol/l)	1.4 ± 0.1	0.7 ± 0.1[b]	1.3 ± 0.1	1.3 ± 0.1
Serum triglycerides (mmol/l)	1.0 ± 0.1	0.5 ± 0.03[b]	0.8 ± 0.04	0.8 ± 0.03
Serum insulin (pmol/l)	96 ± 22	83 ± 18	89 ± 12	116 ± 24
Systolic blood pressure (mmHg)	134 ± 3	112 ± 4[c]	124 ± 3	115 ± 7
Heart rate (bpm)	457 ± 6	443 ± 9	440 ± 9	427 ± 7

[a] $p < 0.01$; [b] $p < 0.001$; [c] $p < 0.05$.

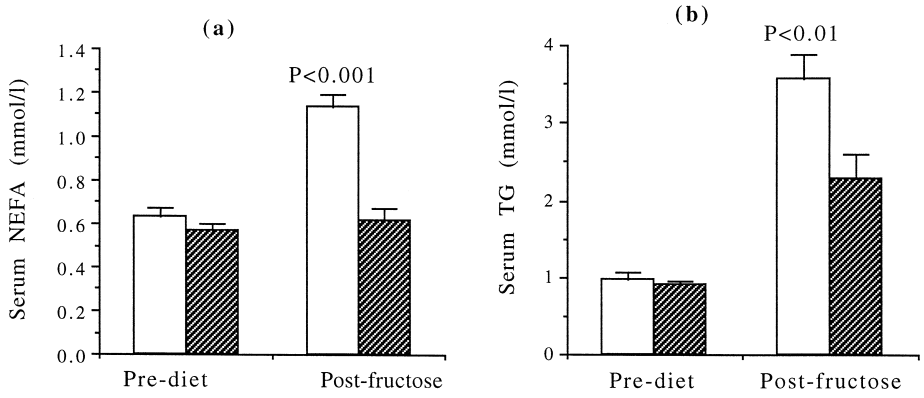

Fig. 1. Fasting serum nonesterified free fatty acid (NEFA) (a) and triglyceride (TG) (b) concentrations before and after fructose feeding in rats treated with GR79236 (striped column, $n = 11$) or vehicle (empty column, $n = 12$). The high-fructose diet was given for 10 days prior to commencing GR79236 (or vehicle) 1 mg/kg per day for 8 days.

2.9. Laboratory assays

Serum glucose and triglyceride levels were measured using enzymatic colorimetric methods (Trinder Glucose kit, GPO-Trinder Tryglyceride kit, Sigma). Serum insulin concentrations were measured by double-antibody radioimmunoassay using rat insulin standards and an anti-rat insulin primary antibody. Serum nonesterified free fatty acid concentrations were measured using a commercial enzymatic colorimetric kit.

2.10. Statistical analysis

Serum glucose and insulin concentration–time profiles for oral and intravenous glucose tolerance tests were compared by ANOVA. Measurements of BP, fasting plasma glucose, insulin, triglyceride and free fatty acid concentrations were analyzed by one factor ANOVA. All measurements are expressed as mean ± standard error (S.E.M.), and statistical significance was accepted at the 5% level.

3. Results

Treatment with GR79236 for 8 days was generally well tolerated, and weight gain was similar in drug-treated compared with vehicle-treated animals (Table 1).

3.1. Effects on fatty acid and triglyceride metabolism

In normal, chow-fed rats, GR79236 produced significant reductions in fasting serum free fatty acid (50%) and triglyceride (55%) concentrations ($p < 0.001$, Table 1). Furthermore, in rats maintained on a high fructose diet prior to (and during) drug treatment, GR79236 attenuated the 2–3 fold rise in serum fatty acid and triglyceride concentrations associated with fructose feeding (Fig. 1).

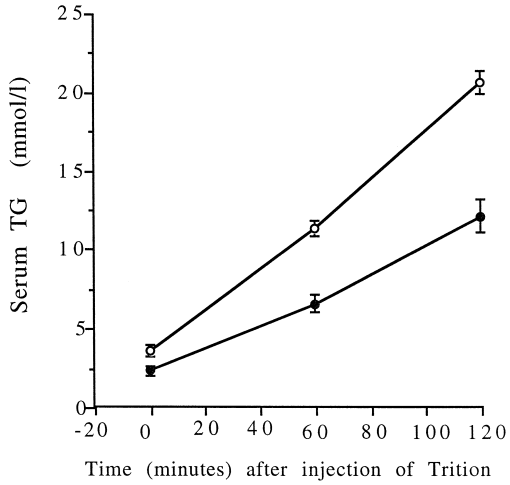

Fig. 2. Serum triglyceride (TG) concentrations before and after tail-vein injection of Triton in groups of fructose-fed rats treated with GR79236 (●, $n = 12$) or vehicle (○, $n = 12$).

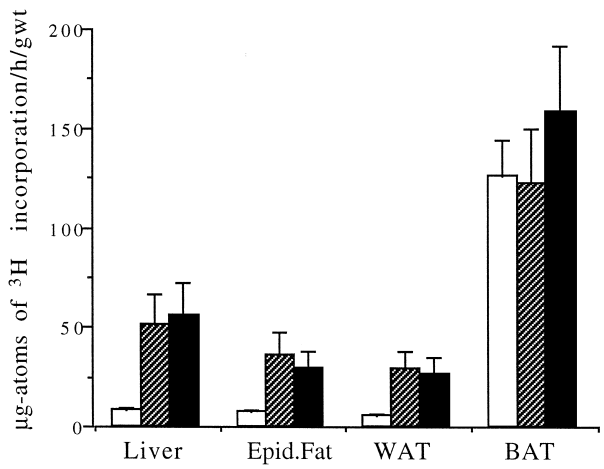

Fig. 3. In vivo lipid synthesis (^{3}H incorporation into fatty acids) in groups of chow-fed controls (empty column, $n = 6$), fructose + GR79236 (striped column, $n = 9$), and fructose + vehicle (filled column, $n = 9$).

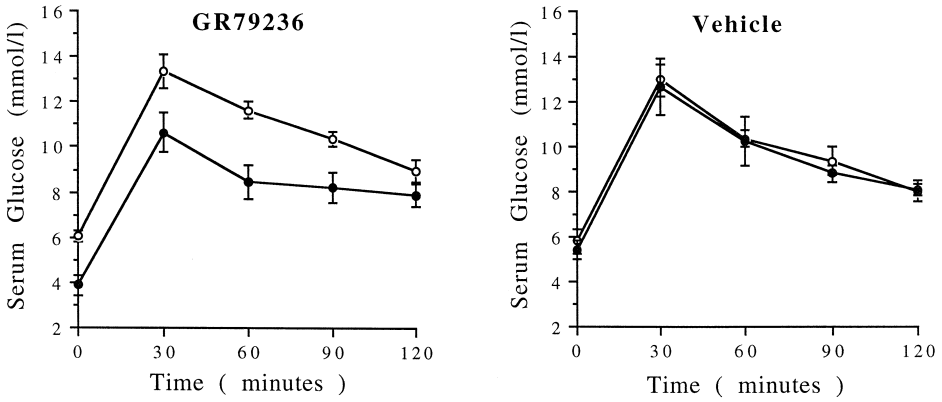

Fig. 4. Oral glucose tolerance tests performed in fasted normal rats before (○) and after (●) 8 days treatment with GR79236 ($n = 8$, left panel) or vehicle ($n = 8$, right panel).

To explore the mechanism of the hypotriglyceridaemic effect, measurements of total triglyceride secretion rate were performed in fructose-fed rats. The rise in serum triglyceride concentrations in the two groups following injection of triton is shown in Fig. 2. There was no difference in plasma volume between the two groups (3.8 ± 0.1 vs. 4.0 ± 0.1 ml/100 g b.wt), but treatment with GR79236 was associated with a significant reduction in total triglyceride secretion rate: 17.3 ± 1.7 compared with 30.2 ± 1.1 for vehicle-treated animals ($p < 0.001$, Fig. 2).

Fructose feeding was associated with a marked increase in fatty acid synthesis in liver and white adipose tissue, but GR79236 had no significant effect on fructose-induced lipogenesis (Fig. 3).

3.2. Effects on glucose tolerance and insulin sensitivity

In normal rats, GR79236 produced a significant reduction in fasting serum glucose concentrations (Table 1), and, following an oral glucose load, there was a significant reduction in area-under-the-curve (AUC) of the serum glucose concentration–time profile: mean AUC[glu] was 21.2 ± 1.3 at baseline versus 16.5 ± 1.1 mmol h/l after 8 days treatment with GR79236 ($n = 8$, $p < 0.02$), compared with corresponding values of 19.9 ± 0.6 and 19.3 ± 1.3 mmol h/l for vehicle-treated rats (Fig. 4).

Intravenous glucose tolerance tests were performed in groups of fructose-fed rats treated with GR79236 or vehicle, and in untreated chow-fed controls. Fructose-fed animals were generally less glucose tolerant relative to chow-fed rats, and the fructose-fed group treated with GR79236 had significantly lower serum glucose concentrations at all timepoints compared with rats treated with fructose + vehicle (Fig. 5).

GR79236 had no significant effect on serum insulin concentrations, either in the fasted state or after a glucose challenge (Table 1). Fructose feeding produced a state of insulin resistance, as shown by higher steady-state plasma glucose values during the insulin suppression test (7.1 ± 0.6 versus 6.1 ± 0.4 mmol/l), but GR79236 had no significant effect on whole-body insulin sensitivity either in normal rats or those fed a high fructose diet (Table 2). Nor did drug therapy (or fructose feeding) affect the rate of insulin clearance; steady-state plasma insulin values were similar in both groups.

3.3. Haemodynamic effects

In normal, chow-fed rats, GR79236 produced a small but significant reduction in systolic blood pressure: 134 ± 3 mmHg at baseline to 112 ± 4 after 8 days, compared with corresponding mean values of 124 ± 3 mmHg and 115 ± 7 for vehicle-treated rats (Table 1). The fall in blood pressure was associated with no significant change in heart rate: 457 ± 5 vs. 443 ± 9 bpm (GR79236 group) and 440 ± 9 vs. 427 ± 7 bpm (vehicle group) at baseline and 8 days, respectively.

Fructose feeding was associated with a significant in-

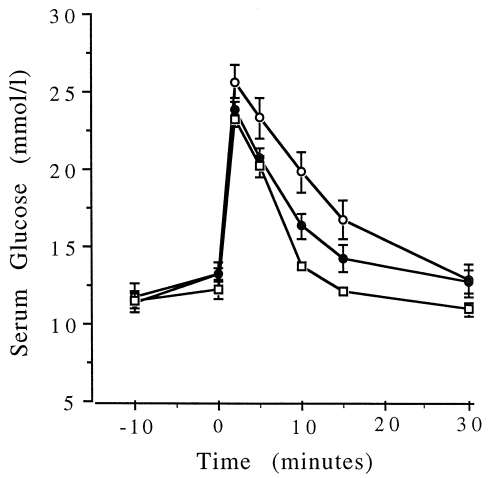

Fig. 5. Intravenous glucose tolerance tests in 3 groups of fasted rats: chow-fed controls (□, $n = 5$), fructose-fed + GR79236 (●, $n = 6$) and fructose-fed + vehicle (○, $n = 6$).

Table 2
Effects of GR79236 on steady-state plasma glucose (a marker of whole body insulin sensitivity) and steady-state plasma insulin concentrations during the insulin suppression test in groups of normal (chow fed) and fructose-fed Sprague–Dawley rats

Insulin suppression test	Normal rats		Fructose-fed rats	
	GR79236-treated ($n = 11$)	vehicle-treated ($n = 10$)	GR79236-treated ($n = 11$)	vehicle-treated ($n = 11$)
Steady-state plasma glucose (mmol/l)	5.4 ± 0.6	6.1 ± 0.4	6.5 ± 0.9	7.1 ± 0.7
Steady-state plasma insulin (pmol/l)	1114 ± 148	1293 ± 113	1254 ± 139	1567 ± 103

Table 3
Effects of GR79236 on systolic blood pressure and heart rate before and after fructose feeding

	GR79236 ($n = 12$)		Vehicle ($n = 12$)	
	pre-diet	fructose	pre-diet	fructose
Systolic blood pressure (mmHg)	125 ± 2	133 ± 5	126 ± 3	146 ± 3[a]
Heart rate (bpm)	446 ± 4	406 ± 11[a]	448 ± 6	434 ± 8

[a] $p < 0.05$ versus baseline (pre-diet).

crease in blood pressure, as shown in previous studies (Hwang et al., 1987; Donnelly et al., 1995), and GR79236 had a significant antihypertensive effect, associated with a modest reduction in heart rate, in rats with fructose-induced hypertension (Table 3).

4. Discussion

The major finding of this study is that short-term treatment with GR79236, in a dose of 1 mg/kg per day, is associated with metabolic and haemodynamic effects both in normal Sprague–Dawley rats and in rats with fructose-induced insulin resistance, hypertriglyceridaemia and hypertension. Previous studies with GR79236 have shown dose-dependent inhibition of catecholamine-induced lipolysis in vitro (Strong et al., 1993), and in this study fasting nonesterified free fatty acid concentrations were reduced by approximately 50%, even in normal rats. It is worthy of note that similar reductions in free fatty acid levels (63%, maximal 1 h after dosing) were reported in a single-dose study in normal rats (Gardner et al., 1994), which suggests that over an 8-day treatment period tolerance to the antilipolytic effect of GR79236 does not occur. This is in contrast to the down-regulation in response observed after chronic dosing with other inhibitors of lipolysis (Myles et al., 1985). There was no effect of the drug on rates of fatty acid synthesis in white and brown adipose tissues, but plasma triglyceride levels were reduced, on average, by 55% as a result of decreased total triglyceride secretion rate. It seems likely that the decrease in triglyceride secretion reflects decreased substrate (fatty acid) availability, especially in the liver. The mechanism of the hypertriglyceridaemia in fructose-fed rats is still unclear, but it is of clinical significance that GR79236 ameliorated a common form of dyslipidaemia which is characteristic of dietary-induced insulin resistance both in rodents and humans.

Unfortunately, this study provides no information about the duration of the antilipolytic effect of GR79236, since each experiment was performed 2 h after administration of the final dose (day 8). It would be particularly important to assess whether the kinetic profile of this drug offers potential advantages over traditional short-acting compounds such as nicotinic acid.

High circulating fatty acid concentrations have been identified as a risk factor for NIDDM (Paolisso et al., 1995), and diabetes is associated with resistance to adenosine and prostaglandin mediated inhibition of lipolysis (Green and Johnson, 1991). Randle et al. (1963) were the first to propose that high circulating fatty acid levels, via the glucose-fatty acid cycle, might exacerbate the defects in insulin-mediated glucose uptake and thereby worsen hyperglycaemia in patients with diabetes. Since the rate of adipose tissue lipolysis is the primary determinant of plasma free fatty acid concentrations, there has been clinical interest in the effects of antilipolytic drugs on glucose metabolism. Phenylisopropyladenosine has been shown to have glucose-lowering activity (Reaven et al., 1988a,b), but in general the metabolic responses to antilipolytic drugs have been inconsistent (Axelrod et al., 1979a,b). The principal mechanism by which adenosine inhibits lipolysis is via G-protein-linked inhibition of adenylate cyclase and reduced formation of cAMP (LaNoue and Martin, 1994). However, it has been shown that adenosine (via the adenosine A_1-receptor) also lowers the concentration of insulin required to produce half-maximal effects on lipolysis and glucose transport by mechanisms that are independent of cAMP (Londos et al., 1985; Green, 1987), e.g., via a direct effect on insulin-stimulated translocation of glucose transporter proteins in the adipocyte plasma membrane (Kuroda et al., 1987).

In the present study, there was clear evidence that GR79236 reduced fasting serum glucose concentrations in normal rats and reduced the AUC[glu] following an oral glucose load. Furthermore, the effects of fructose feeding on intravenous glucose tolerance were ameliorated with drug therapy. Serum insulin concentrations in rats are widely variable, therefore in relatively small groups of animals it is often difficult to show significant improvements in fasting insulin levels even with well-known antidiabetic drugs. It is perhaps surprising that, despite obvious improvements in glucose tolerance, GR79236 ap-

peared to have no significant effect on whole-body insulin sensitivity, as reflected by similar steady-state plasma glucose values during the insulin suppression tests, but this might be due to differential effects of the drug on insulin sensitivity in different tissues. For example, adenosine (via the adenosine A_1-receptor) and GR79236 increase insulin-mediated glucose uptake in isolated adipocytes (Heseltine et al., 1995), but adenosine-A_1 agonists seem to have the opposite effect in skeletal muscle (Challis et al., 1992). Thus, interpretation of the insulin sensitivity experiments is difficult when adenosine A_1-receptor-mediated effects on glucose transport and metabolism may be quite different in various tissues, and when fructose feeding may have tissue-specific effects on adenosine A_1-receptor mediated responses. High circulating free fatty acid concentrations worsen hepatic glucose overproduction, and another antilipolytic drug, acipimox, lowers plasma glucose levels primarily due to its effects on the liver (Lee et al., 1996). Since fructose feeding increases both glucose and triglyceride output from the liver, it seems likely that the metabolic effects of GR79236 observed in this study are due, at least in part, to changes in hepatic metabolism.

The cardiovascular effects of adenosine include A_1-receptor-mediated slowing of atrioventricular node conduction and decreased contractility in the heart, and adenosine A_2-receptor-mediated vasodilation of peripheral arterioles (Olsson and Pearson, 1990). Adenosine-A_1 agonists have recently been used in clinical studies of heart failure (Bertolet et al., 1996), but in experimental models of hypertension it has been suggested that tolerance develops to adenosine A_1-mediated (but not A_2-mediated) antihypertensive effects (Casati et al., 1994). In this study, the effect of GR79236 on blood pressure in normal rats was of similar magnitude to that reported in earlier single-dose studies (Gardner et al., 1994; Merkel et al., 1995), but the antihypertensive response in fructose-fed animals is perhaps of greater clinical significance. The mechanism of fructose-induced hypertension has not been clearly established, but the rise in blood pressure is not Na^+-dependent (Donnelly et al., 1995) and manoeuvres that improve insulin sensitivity, e.g., exercise, also reduce blood pressure (Reaven et al., 1988a,b). Since fatty acid levels correlate with blood pressure in humans (Simon et al., 1996), and in view of the pathophysiological association between insulin resistance and hypertension, it is possible that the antihypertensive effect of GR79236 is in part secondary to the effects on NEFA and glucose metabolism.

Previous single-dose studies with GR79236 have reported 10–12% reductions in heart rate (Gardner et al., 1994; Merkel et al., 1995), but in this study the fall in blood pressure occurred without significant bradycardia in normal rats. While this may be due to the limitations of tail-cuff plethysmography, it has been shown that adenosine A_1-receptors in the atria are especially prone to desensitization (Lee et al., 1993), in which case the antihypertensive response during chronic dosing (in contrast to acute administration) may be independent of cardiac effects. Alternative mechanisms that might account for the blood pressure-lowering effect of an adenosine A_1 agonist during repeated dosing include inhibition of renin release (Churchill and Churchill, 1985) and increased production of atrial natriuretic factor (Massi et al., 1992).

Thus, short-term treatment with this novel antilipolytic drug improves lipid and glucose metabolism, and lowers blood pressure, in normal rats and in a dietary-induced model of insulin resistance, hypertension and dyslipidaemia that resembles many of the characteristic features of Syndrome X (the so-called insulin resistance syndrome) in humans.

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