Comparison of the Effects of Pioglitazone and Metformin On Hepatic and Extra-Hepatic Insulin Action in People with Type 2 Diabetes

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Abstract

Objective: To determine mechanisms by which pioglitazone and metformin effect hepatic and extra-hepatic insulin action.

Design: 31 subjects with type 2 diabetes were randomly assigned to pioglitazone (45 mg) or metformin (2000 mg) for four months.

Results: Glucose was clamped before and after therapy at ~5 mmol/L, insulin raised to ~180 pmol/l, C-peptide suppressed with somatostatin, glucagon replaced at ~75 pg/ml and glycerol maintained at ~200 mmol/l to ensure comparable and equal portal concentrations on all occasions. Insulin induced stimulation of glucose disappearance did not differ before and after treatment with either pioglitazone (23 ± 3 vs. 24 ± 2 µmol/kg/min) or metformin (22 ± 2 vs. 24 ± 3 µmol/kg/min). In contrast, pioglitazone enhanced (p<0.01) insulin induced suppression of both glucose production (6.0 ± 1.0 vs. 0.2 ± 1.6 µmol/kg/min) and gluconeogenesis (n=11: 4.5 ± 0.9 vs.0.8 ± 1.2 µmol/kg/min). Metformin did not alter either suppression of glucose production (5.8 ± 1.0 vs. 5.0 ± 0.8 µmol/kg/min) or gluconeogenesis (n=9: 3.7 ± 0.8 vs. 2.6 ± 0.7 µmol/kg/min). Insulin induced suppression of free fatty acids was greater (p<0.05) following treatment with pioglitazone (0.14 ± 0.03 vs. 0.06 ± 0.01 mmol/L) but unchanged with metformin (0.12 ± 0.03 vs. 0.15 ± 0.07 mmol/L).

Conclusions: Thus relative to metformin, pioglitazone improves hepatic insulin action in people with type 2 diabetes, partly by enhancing insulin induced suppression of gluconeogenesis. On the other hand, both drugs have comparable effects on insulin induced stimulation of glucose uptake.

Key words: gluconeogenesis; glycogenolysis; insulin resistance; free fatty acids
Thiazolidinediones and metformin are extensively used to treat people with type 2 diabetes. Both are considered to be insulin “sensitizers” (1-5). However, the mechanism of action of these drugs remains an area of active investigation. Although it is commonly stated that thiazolidinediones lower glucose concentration primarily by increasing glucose uptake and metformin by decreasing glucose production (1-5), the data supporting these statements are scarce and often contradictory. *In vitro* and animal studies have identified multiple potential targets for these drugs (6-12). Both increase insulin signaling and muscle as well as adipocyte glucose uptake (6-8,11). Both have been shown to modulate the activity of hepatic enzymes particularly those involved in the gluconeogenic pathway (9,10,12-16). In contrast, the results have not always been consistent in humans (for review see (17)). The effects of metformin have been particularly variable with some (18-21) but not all (22-25), showing an improvement in insulin action. However, since any therapy that lowers glucose concentration (e.g. treatment with insulin or sulfonylurea) improves insulin action (26-28), it has been difficult to distinguish in placebo controlled trials between a specific effect of the thiazolidinedione or metformin from that due to he associated reduction in glucotoxicity.

The relative ability of these agents to modulate hepatic and extra-hepatic insulin action is uncertain since most studies have compared the effects of a thiazolidinedione or metformin to that observed during treatment with placebo (14,19,21,26-31) or prior to treatment (13,15,16,18), rather than to one another. To our knowledge, the exception is the recent study by Tikkainen et al (32). In this carefully designed clinical trial, previously untreated subjects with type 2 diabetes mellitus were randomly assigned to receive either rosiglitazone or metformin for 16 weeks. Hepatic insulin action, when estimated by multiplying fasting insulin concentration times fasting endogenous glucose production, comparably improved in both groups following treatment. On the other hand, when hepatic insulin action was assessed during a hyperinsulinemic euglycemic clamp, suppression of glucose production was greater following treatment with metformin than treatment with rosiglitazone. However, the insulin concentrations were higher during the clamp with metformin than with rosiglitazone confounding interpretation of the data.

In an effort to gain greater insight regarding the mechanism of action of these commonly used therapeutic agents, hepatic and extra-hepatic insulin action were measured in people with type 2 diabetes using a hyperinsulinemic euglycemic clamp prior to and following four months of treatment with either pioglitazone or metformin. We specifically sought to test the hypothesis that in the presence of physiologic insulin concentrations, suppression of endogenous glucose production is greater following treatment with a thiazolidinediones than following treatment with metformin. In order to accurately assess hepatic insulin action, insulin was raised to concentrations that in the absence of treatment with a sensitizer are known to result in sub-maximal suppression of endogenous glucose production (32). C-peptide was suppressed with somatostatin, glucagon replaced at ~75 pg/ml and glycerol maintained at ~200 mmol/l to insure comparable and equal portal concentrations on all occasions. Gluconeogenesis and glycogenolysis were measured using the deuterated water method. We report, that whereas pioglitazone and metformin have comparable effects on insulin induced stimulation of glucose uptake, pioglitazone improved hepatic insulin action at least in part by enhancing insulin induced suppression of gluconeogenesis.
Research Design and Methods

Subjects

After approval from the Mayo Institutional Review Board, 31 subjects with type 2 diabetes mellitus gave informed written consent to participate in the study. All subjects were in good health and at a stable weight. None regularly engaged in vigorous physical exercise. Following completion of the pre-treatment study, subjects were randomly assigned (double-blind double-placebo controlled) to receive either pioglitazone (45 mg daily) or metformin (1000 mg twice daily) for four months. Of the sixteen subjects in the pioglitazone group, 12 had been previously treated with metformin alone, 1 sulfonylurea alone, 2 with combination of metformin and sulfonylurea, and 1 with diet alone. Of the fifteen subjects in the metformin group, 12 had been previously treated with either metformin alone, 1 sulfonylurea alone and, 2 with combination of metformin and sulfonylurea. Oral hypoglycemic medications were discontinued at least ten days prior to the pre-treatment study visit. Following randomization, pioglitazone was started at 30 mg daily for the first week and then increased to 45 mg dose at the end of the first week. Metformin was started at 500 mg once daily and then increased in increments of 500 mg/week to a maximum dose of 2000 mg within four weeks. Subjects continued to receive the pioglitazone or metformin during the post-treatment study, i.e. study drugs were not discontinued during the evening prior and morning of study and each subject took their regular and study medications as prescribed. Subjects on stable doses of thyroxine, estrogen replacement therapy, 3-hydroxy-3-methylglutaryl coenzyme A reductase inhibitors and metabolically neutral antihypertensive medication (low dose thiazide, calcium channel blockers or losartan) continued these medicines during the study.

Experimental Design

All subjects were instructed to follow a weight maintenance diet containing 55% carbohydrate, 30% fat and 15% protein for at least three days prior to the day of study. Subjects were admitted to the Mayo Clinic Clinical Research Unit at 1700 on the evening before the study. A standard 10 cal/kg meal (55% carbohydrate, 30% fat and 15% protein) was eaten between 1730 and 1800 hours. After sampling blood for baseline enrichment, 1.67 $\text{H}_2\text{O}$/kg weight of body water was given in three divided doses at 1800, 2000 and 2200 hours. Small sips of water (containing $\text{H}_2\text{O}$) were permitted upon request. After the meal, an 18-gauge catheter was inserted into a forearm vein and an infusion of insulin was started (100 U regular human insulin in 1 L of 0.9% saline containing five ml of 25% human albumin). The insulin infusion rate was adjusted to maintain glucose concentrations at ~5 mmol/L during the night (33).

An infusion of glycerol was started at 0600 in amounts equal to that given as part of a separate protocol in which subjects also were infused with Intralipid. A primed (fasting glucose in mmol/L divided by 5.5 mmol/L times 12 $\mu$Ci ) continuous (0.12 $\mu$Ci/min) infusion of [3-$\text{H}$] glucose (Perkin Elmer, Boston, MA) at 0700; and infusions of insulin (0.6 mU/kg/min), somatostatin (60 ng/kg/min), growth hormone (3 ng/kg/min) and glucagon (0.65 ng/kg/min) were started at 1000 (time 0 min). An infusion of [9,11,12,12-$\text{H}_4$] cortisol also was started at 0600 as part of a separate protocol. Beginning at 0930, glucose was infused in amounts sufficient to maintain plasma glucose concentrations at ~5 mmol/L. All infused glucose contained [3-$\text{H}$] glucose in order to minimize the change in plasma glucose specific activity. In addition, the rate of the “basal” [3-$\text{H}$] glucose infusion was varied to mimic the anticipated changes in endogenous glucose production.
Analytical Techniques

Blood samples were collected in prechilled syringes and dispensed into prechilled tubes. Samples for free fatty acids were collected in tubes containing 50 µL of Paraaxon® (diethyl-p-nitrophenyl-phosphate) (Sigma Chemicals, St Louis, MO) diluted to 0.04 % in diethyl ether to prevent ex-vivo lipolysis. Samples were placed in ice, centrifuged at 4°C, and separated. All other samples were stored at -20°C until analysis. Plasma glucose was measured by a glucose oxidase method using a YSI glucose analyser (Yellow Springs, OH). Plasma insulin was measured using a chemiluminescence method with the Access® Ultrasensitive Immunoenzymatic assay system (Beckman, Chaska, MN). C-peptide and glucagon concentrations were assayed by radioimmunoassay (RIA, Linco Research Inc., St. Louis, MO). [3-3H] glucose specific activity was measured by liquid scintillation counting as previously described. Plasma glycerol and free fatty acid concentrations were measured by a modified microfluorometric enzymatic method using the Cobas, MIRA analyzer (Roche). Enrichment of deuterium on the 2nd and 5th carbons of plasma glucose was measured as previously described. (35). Body composition (including fat free mass, total fat mass, and visceral fat mass) was measured using dual energy X-ray absorptiometry (DPX scanner, Lunar Corp, Madison, WI) combined with a computerized tomograph scan at the level of L2 /L3 as previously described (36). Velocity sedimentation/gel filtration chromatography was used for separation of adiponectin complexes using a human adiponectin radioimmunoassay kit(LINCO,St Louis,MO) in 8 subjects in the pioglitazone group and 8 subjects in the metformin group).

Calculations

Rates are expressed in the figures and text as µmol per kg lean body mass per minute. Basal and clamp responses were assessed by taking the mean of the values present from -30 to 0 minutes and from 270 to 300 minutes respectively. Glucose appearance and disappearance were calculated using the steady state equations of Steele et al. (37). Endogenous glucose production during the clamp was calculated by subtracting the exogenous glucose infusion rate from the total glucose appearance rate. The rate of gluconeogenesis was calculated by multiplying the plasma ratio of C5 and C2 glucose enrichments times endogenous glucose production (38). Glycogenolysis was calculated by subtracting the rate of gluconeogenesis from endogenous glucose production. Rates of gluconeogenesis and glycogenolysis could be measured in twenty of the thirty-one subjects (n=11 in the pioglitazone group and n = 9 metformin group). Rates could not be measured in the other subjects due to high baseline C5 enrichment.

Statistical Analysis

Data in the text and figures are expressed as mean ± SEM. Student’s paired t test was used to determine whether values were different before and after treatment. A p value of less than 0.05 was considered as statistically significant.

Results

Patient characteristics (table 1)

Patient characteristics are provided in table 1. Age, gender, body mass index, lean body mass, percent body fat, and visceral fat did not differ between groups before treatment. Body weight and percent body fat were slightly higher (p<0.05) with pioglitazone but remained unchanged with metformin. HbA1c did not differ prior to therapy and did not change during treatment with pioglitazone. However, HbA1c increased (p<0.05) slightly during treatment with metformin.
Glucose, insulin, C-peptide and glucagon concentrations (figure 1)

Glucose concentrations before and during the final thirty minutes of the clamp did not differ prior to or during treatment in either the pioglitazone or metformin groups.

The insulin infusion rate required to maintain euglycemia during the final hour before the clamp were slightly but not significantly lower following treatment with either metformin (1.7 ± 0.3 vs. 1.2 ± 0.3 U/hr) or pioglitazone (1.9 ± 0.3 vs. 1.2 ± 0.3 U/hr). While fasting insulin concentrations following the overnight exogenous insulin infusion also tended to decrease after treatment with either pioglitazone (143 ± 27 vs. 94 ± 21 pmol/L) or metformin (136 ± 22 vs. 97 ± 18 pmol/L), neither change was statistically significant. Plasma insulin concentrations during the final thirty minutes of the clamp did not differ prior to and during treatment with either pioglitazone (185 ± 12 vs. 181 ± 9 pmol/L) or metformin (185 ± 10 vs. 178 ± 8 pmol/L).

C-peptide concentrations following the overnight exogenous insulin infusion did not differ during treatment with either pioglitazone (0.25 ± 0.03 vs. 0.29 ± 0.03 nmol/L) or metformin (0.18 ± 0.03 vs. 0.21 ± 0.03 nmol/L). Somatostatin resulted in comparable and near complete suppression of C-peptide secretion on all occasions.

Plasma glucagon concentrations following the overnight exogenous insulin infusion did not differ during treatment with either pioglitazone (80 ± 4 vs. 86 ± 6 pg/ml) but were higher (p<0.01) during treatment with metformin (63 ± 4 vs. 95 ± 9 pg/ml). On the other hand, plasma glucagon concentrations during the final thirty minutes of the clamp did not differ prior to and during treatment with either pioglitazone (79 ± 5 vs. 74 ± 3 pg/ml) or metformin (68 ± 3 vs. 76 ± 4 pg/ml).

Glucose infusion rate required to maintain euglycemia, glucose specific activity (figure 2)

Compared to pretreatment, the glucose infusion rate required to maintain euglycemia increased (p<0.01) during treatment with pioglitazone (16.7 ± 3.2 vs. 24.1 ± 3.0 µmol/kg/min) but did not differ during treatment with metformin (16.4 ± 3.2 vs. 18.9 ± 3.0 µmol/kg/min). Glucose specific activity remained constant before and during the clamp both prior to and following treatment with either pioglitazone or metformin thereby permitting accurate measurements of glucose turnover.

Endogenous glucose production and glucose disappearance (figure 3)

Endogenous glucose production before the clamp did not differ prior to and during treatment with either pioglitazone (15.8 ± 0.6 vs. 16.1 ± 0.6 µmol/kg/min) or metformin (17.2 ± 0.7 vs. 17.6 ± 1.1 µmol/kg/min). Suppression of endogenous glucose production during the final thirty minutes of the clamp was greater (p<0.001) during treatment with pioglitazone (6.0 ± 1.0 vs. 0.2 ± 1.6 µmol/kg/min) but did not differ during treatment with metformin (5.8 ± 1.0 vs. 5.0 ± 0.8 µmol/kg/min).

Glucose disappearance before the clamp did not differ prior to and during treatment with either pioglitazone (15.8 ± 0.6 vs. 16.1 ± 0.6 µmol/kg/min) or metformin (17.2 ± 0.7 vs. 17.6 ± 1.1 µmol/kg/min). Glucose disappearance during the final thirty minutes of the clamp also did not differ prior to and during treatment with either pioglitazone (22.8 ± 2.8 vs. 24.4 ± 1.8 µmol/kg/min) or metformin (22.2 ± 2.5 vs. 23.8 ± 2.8 µmol/kg/min).

Gluconeogenesis and glycogenolysis (figure 4)

Gluconeogenesis before the clamp did not differ prior to and during treatment with
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either pioglitazone \( (10.6 \pm 0.5 \text{ vs. } 11.1 \pm 0.7 \mu\text{mol/kg/min}) \) or metformin \( (9.8 \pm 0.7 \text{ vs. } 10.3 \pm 0.9 \mu\text{mol/kg/min}) \). On the other hand, gluconeogenesis during the final thirty minutes of the clamp was lower \( (p<0.01) \) during than before treatment with pioglitazone \( (4.5 \pm 0.9 \text{ vs. } 0.8 \pm 1.2 \mu\text{mol/kg/min}) \) but did not differ during treatment with metformin \( (3.7 \pm 0.8 \text{ vs. } 2.6 \pm 0.7 \mu\text{mol/kg/min}) \). Glycogenolysis did not differ before the clamp prior to and during treatment with pioglitazone \( (6.3 \pm 0.5 \text{ vs. } 6.3 \pm 0.4 \mu\text{mol/kg/min}) \) or metformin \( (7.6 \pm 0.7 \text{ vs. } 7.0 \pm 0.7 \mu\text{mol/kg/min}) \). On the other hand, suppression of glycogenolysis was greater during the final thirty minutes of the clamp \( (p<0.01) \) following treatment with pioglitazone \( (2.0 \pm 0.4 \text{ vs. } -0.3 \pm 0.9 \mu\text{mol/kg/min}) \) but did not differ following treatment with metformin \( (2.2 \pm 0.5 \text{ vs. } 1.5 \pm 0.4 \mu\text{mol/kg/min}) \).

**Free fatty acid and glycerol concentrations (figure 5)**

Plasma free fatty acid concentrations before the clamp did not differ prior to and during treatment with pioglitazone \( (0.39 \pm 0.05 \text{ vs. } 0.34 \pm 0.05 \text{ mmol/L}) \). On the other hand, treatment with pioglitazone resulted in greater \( (p<0.05) \) suppression of plasma free fatty acid concentrations during the final thirty minutes of the clamp \( (0.14 \pm 0.03 \text{ vs. } 0.06 \pm 0.01 \text{ mmol/L}) \). In contrast, plasma free fatty acids during treatment with metformin were higher \( (p<0.05) \) before the clamp \( (0.26 \pm 0.04 \text{ vs. } 0.47 \pm 0.08 \text{ mmol/L}) \) but did not differ during the final thirty minutes of the clamp \( (0.12 \pm 0.03 \text{ vs. } 0.15 \pm 0.07 \text{ mmol/L}) \). Plasma glycerol concentrations did not differ before or during the final thirty minutes of the clamp prior to or during treatment with either pioglitazone or metformin.

**Total and HMW plasma adiponectin concentrations (figure 6)**

Treatment with pioglitazone resulted in a marked increase \( (p<0.01) \) in both total and HMW plasma adiponectin concentrations. In contrast, neither total nor HMW plasma adiponectin concentrations changed following treatment with metformin.

**Discussion**

Thiazolidinediones and metformin both are considered insulin “sensitizers” \( (1-5) \). The present data indicate that when compared to one another, pioglitazone and metformin have equivalent effects on insulin induced stimulation of glucose uptake assessed at insulin concentrations that are commonly present under the conditions of daily living. On the other hand, relative to metformin, pioglitazone increases insulin induced suppression of endogenous glucose production at least in part by enhancing suppression of gluconeogenesis.

Multiple studies have shown that thiazolidinediones improve hepatic insulin action relative to placebo \( (6,16,30,31) \). The present studies establish that thiazolidinediones also improve hepatic insulin action relative to metformin. Insulin induced suppression of glucose production was greater following four months of treatment with pioglitazone but did not differ following four months of treatment with metformin. Glucose production was assessed under conditions designed to optimize comparison between therapies. Consistent with previous reports \( (39-41) \), treatment with metformin increased fasting glucagon concentrations. However this does not explain the difference in hepatic insulin sensitivity following treatment with pioglitazone or metformin since endogenous glucagon secretion was inhibited during the clamps with somatostatin. Insulin was infused at rates anticipated to result in sub-maximal suppression of glucose production so that
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differences in hepatic response could be evaluated. This design resulted in peripheral (and presumably portal) insulin concentrations during the clamps that were comparable before and during treatment in both groups. Furthermore, the gluconeogenic precursor glycerol, infused as part of a separate protocol evaluating the effects of elevated free fatty acids on glucose metabolism, was maintained at comparable concentrations during treatment with pioglitazone or metformin despite differences in plasma free fatty acid concentrations. Consistent with previous studies, treatment with pioglitazone resulted in both an increase in plasma adiponectin concentration and an improvement in hepatic insulin action (16, 43). In contrast, treatment with metformin did not alter either plasma adiponectin concentration (32, 42) or hepatic insulin action. These data further support the premise that thiazolidinediones improve insulin action at least in part by increasing adiponectin concentrations (16).

The observation in the present study that hepatic insulin action was enhanced after treatment with pioglitazone but not metformin differs from the report by Tikkainen et al that glucose production was lower and percent suppression greater after treatment with metformin but not after treatment with rosiglitazone. While it is possible that rosiglitazone and pioglitazone have different effects on hepatic insulin action, we doubt this is the case. More likely, the apparent discordance between the conclusions of the two studies is due to the fact that in the study of Tikkainen et al a) insulin concentrations were significantly lower on the rosiglitazone than metformin study days thereby accounting for lesser suppression of glucose production, b) glucose concentrations were clamped at hyperglycemic levels (~8 mmol/l) making it difficult to distinguish between effects of these agents on glucose effectiveness from those due to changes in insulin action, and c) endogenous insulin secretion was not inhibited leaving open the possibility that differences in suppression of glucose production following treatment were due to differences in portal insulin concentrations rather than differences in hepatic insulin action. Taken together, the current data indicate that when measured under the appropriate conditions, (i.e. glucose concentrations clamped at euglycemic levels with portal insulin and glucagon concentrations matched) pioglitazone enhances insulin induced suppression of glucose production whereas metformin does not.

Glucose production equals the sum of glycogenolysis and gluconeogenesis. Previous studies have shown that thiazolidinediones and metformin inhibit multiple enzymes in the gluconeogenic pathway and when compared to placebo, lower gluconeogenesis in humans (9,10,12-15,43). In the present studies, insulin induced suppression of gluconeogenesis and glycogenolysis improved following treatment with pioglitazone but did not change following treatment with metformin. The enhanced suppression of glucose production was accompanied by enhanced insulin induced suppression of plasma free fatty acid concentration. Since plasma free fatty acids modulate rates of both gluconeogenesis and glycogenolysis, it is possible that pioglitazone improved hepatic insulin sensitivity by enhancing suppression of lipolysis and/or increasing plasma free fatty acid clearance (44-46). However, association or lack thereof does not prove causality. Therefore, future studies will be required to distinguish between the direct effects of thiazolidinediones on hepatic insulin action from those due to their effect in fat metabolism.

Of note, plasma free acid concentrations prior to the clamp were higher after than before metformin treatment. On the
other hand, free fatty acid concentrations suppressed to comparable concentrations during the clamp on both occasions. We have no explanation for the higher basal free fatty acid concentrations following treatment with metformin. However, the higher free fatty acid concentrations may explain, at least in part, why basal rates of glucose production following treatment with metformin did not differ from those present before treatment despite the fact that plasma insulin concentrations tended to be lower. In any case, the current data emphasize the need for further study of the effects of metformin on free fatty acid metabolism in people with type 2 diabetes.

Insulin induced stimulation of glucose uptake did not differ before and during treatment in either the pioglitazone or metformin groups. This indicates that under the present experimental conditions (i.e. low physiologic insulin concentrations) both agents had a comparable effect on extra-hepatic insulin action. Although effects of metformin on insulin action have been inconsistent (18-21), at first glance the present data may appear at variance with previous reports that thiazolidinediones increase insulin stimulated glucose uptake (16,29,30). However, whereas an increase in glucose uptake following treatment with a thiazolidinediones almost always has been observed when assessed in the presence of very high insulin concentrations, enhanced uptake was not observed in the same studies when assessed at lower insulin concentrations that still were substantially higher than those used in the present experiments (16,29,30). The one exception was the study of Tikkanen et al where an improvement in insulin induced stimulation of glucose uptake following treatment with rosiglitazone was detected at insulin concentrations similar to those used in the present experiments (32). However in those experiments, HbA1c fell during treatment with rosiglitazone so reduced glucotoxicity may have contributed to the improvement in insulin action. Taken together, these data suggest that in the presence of low physiologic insulin concentrations, the effects of pioglitazone on hepatic insulin action is likely as, if not more important, than the effects of pioglitazone on extra-hepatic insulin action in the regulation of glucose metabolism. The present study has certain limitations. Twenty four of the thirty one subjects were being treated with metformin prior to study reflecting the standard of practice in our community. Although all glucose-lowering agents were discontinued at least 10 days before the pretreatment study, it could be argued that in these subjects the design in effect was continuation of their current therapy with metformin versus treatment with pioglitazone. However, this does not detract from the observation that treatment with pioglitazone resulted in an improvement in hepatic insulin action whereas treatment with metformin did not. Furthermore, the conclusions were the same when the twenty four individuals who had been treated with metformin alone prior to study were analyzed as a separate subset. HbA1c increased slightly in the metformin group but remained unchanged in the pioglitazone group. Plasma free fatty acids also increased. Therefore it is possible that the deterioration in glycemic control and the associated increase in free fatty acid concentrations blunted the effects of metformin on insulin action. On the other hand, since metformin is not known to have a direct effect on insulin secretion, these changes presumably occurred because insulin action was lower during treatment with metformin than with pioglitazone consistent with the results observed during the clamps.

Subjects in the metformin groups were treated with a maximum of 2000 mg per day. It is possible that higher doses of metformin would have improved hepatic insulin action.
However, we doubt if this would be the case since the efficacy of metformin appears to plateau at 2000 mg per day (47). The maximum dose of pioglitazone was reached within one week whereas the maximum dose of metformin was not reached until one month. We believe that it is unlikely that the difference in time to maximum dose affected the conclusions since subjects were on stable doses of both drugs for three months before study. Insulin was infused during the night to ensure glucose concentrations were comparable in all groups on all study days thereby avoiding the confounding effects of “glucotoxicity” due to differences in glycemic control (48). We only compared metformin to pioglitazone. We do not know if the results would have been the same if we used a different thiazolidinedione (e.g. rosiglitazone) since previous studies have suggested that the effects of these agents on blood lipids differ (49). On the other hand, since when compared to placebo, the effects of pioglitazone and rosiglitazone on insulin action are the same (30,31), we believe the current data are representative of the response to the thiazolidinedione class of drugs rather than specific for pioglitazone.

In summary, in the presence of low physiologic insulin concentrations, insulin induced stimulation of glucose uptake is comparable during treatment with pioglitazone or metformin. In contrast, insulin induced suppression of glucose production is greater during treatment with pioglitazone than during treatment with metformin, at least in part, due to enhanced suppression of gluconeogenesis. Free fatty acids also were lower during the clamp after treatment with pioglitazone but not after treatment with metformin implying that pioglitazone enhances suppression of lipolysis and/or increases FFA clearance. Thus, the common belief that pioglitazone improves glycemic control in people with type 2 diabetes primarily by increasing glucose uptake whereas metformin does so by suppressing glucose production needs to be reexamined.

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Pioglitazone compared to metformin

References


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Table 1. Subject Characteristics

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* p < 0.05 pre treatment vs. post treatment
Figure Legends

Figure 1
Plasma glucose, insulin, C-peptide and glucagon concentrations observed prior to and during infusion of somatostatin in diabetic subjects both prior to and following treatment with pioglitazone (left panel) and metformin (right panel). Somatostatin, and replacement infusions of glucagon and growth hormone were started at time 0.

Figure 2
The intravenous glucose infusion rate required to maintain target glucose concentrations (upper panel) and plasma [3-3H] glucose-specific activity (lower panel) observed in the diabetic subjects both prior to and following treatment with pioglitazone (left panel) and metformin (right panel) during final 40 minutes of a baseline period (basal) and a euglycemic hyperinsulinemic (clamp) period. Somatostatin, insulin and replacement infusions of glucagon and growth hormone were started at time 0.

Figure 3
Rates of endogenous glucose production (upper panel) and glucose disappearance (lower panel) observed in the diabetic subjects both prior to and following treatment with pioglitazone (left panel) and metformin (right panel) during final thirty minutes of a baseline period (basal) and a euglycemic hyperinsulinemic (clamp) period. * represents p<0.001 versus pre-treatment.

Figure 4
Rates of gluconeogenesis (upper panel) and glycogenolysis (lower panel) observed in the diabetic subjects both prior to and following treatment with pioglitazone (left panel) and metformin (right panel) during final thirty minutes of a baseline period (basal) and a euglycemic hyperinsulinemic (clamp) period. * represents p<0.001 versus pre-treatment.

Figure 5
Free fatty acid concentrations observed in the diabetic subjects both prior to and following treatment with pioglitazone (left panel) and metformin (right panel) during final thirty minutes of a baseline period (basal) and a euglycemic hyperinsulinemic (clamp) period. * represents p<0.01 versus pre-treatment.

Figure 6
Total and HMW adiponectin concentrations observed in the diabetic subjects both prior to and following treatment with pioglitazone (left panels) and metformin (right panels) during final thirty minutes of a baseline period (basal) and a euglycemic hyperinsulinemic (clamp) period. * represents p<0.01 versus pre-treatment.
Figure 1

Pioglitazone compared to metformin

Glucose

Insulin

C-Peptide

Glucagon

Pre treatment
Post treatment

Figure 1
Minutes

Figure 1
Minutes
Pioglitazone compared to metformin

Figure 2

Glucose Infusion Rate

Specific Activity

Pre treatment
Post treatment

18
Pioglitazone compared to metformin

**Endogenous Glucose Production**

- **Pioglitazone**
  - Basal: □ Pre treatment, ■ Post treatment
  - Clamp: □ Pre treatment, ■ Post treatment
  - Basal: 15 ± 2 μmol/kg/min, Clamp: 5 ± 2 μmol/kg/min

- **Metformin**
  - Basal: □ Pre treatment, ■ Post treatment
  - Clamp: □ Pre treatment, ■ Post treatment
  - Basal: 20 ± 3 μmol/kg/min, Clamp: 10 ± 3 μmol/kg/min

**Glucose Disappearance**

- **Pioglitazone**
  - Basal: □ Pre treatment, ■ Post treatment
  - Clamp: □ Pre treatment, ■ Post treatment
  - Basal: 25 ± 2 μmol/kg/min, Clamp: 20 ± 2 μmol/kg/min

- **Metformin**
  - Basal: □ Pre treatment, ■ Post treatment
  - Clamp: □ Pre treatment, ■ Post treatment
  - Basal: 30 ± 3 μmol/kg/min, Clamp: 25 ± 3 μmol/kg/min

*Figure 3*
Pioglitazone compared to metformin

**Pioglitizone**

**Metformin**

**Gluconeogenesis**

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<tr>
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**Glycogenolysis**

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<td><strong>Clamp</strong></td>
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Figure 4
Pioglitazone compared to metformin

Free Fatty Acids

Pioglitazone
Metformin

Basal Clamp Basal Clamp

mmol/L

0 0.2 0.4 0.6

Figure 5

Pre treatment Post treatment

Glycerol

Basal Clamp Basal Clamp

mmol/L

0 100 200 300 400 500

Figure 5
Figure 6