Rosiglitazone Restores G-Protein Coupling, Recruitment, and Function of Renal Dopamine D₁A Receptor in Obese Zucker Rats

Meghna Trivedi, Aditi Marwaha, Mustafa Lokhandwala

Abstract—Hypertension related to insulin resistance results from increased sodium retention. Dopamine, by activating D₁A receptors in renal proximal tubules, increases sodium excretion. Recently, dopamine has been shown to augment its own signaling by recruiting intracellular D₁A receptors to cell surface in proximal tubules. In this study, we hypothesized that coupling of D₁A receptors to G proteins and dopamine-induced recruitment of D₁A receptors to the plasma membrane are impaired in obese Zucker rats, resulting in a diminished natriuretic and diuretic response to D₁A receptor agonist, SKF-38393. We also examined effects of rosiglitazone (3 mg/kg per day, 15 days) in restoring the defects in D₁A receptor signaling and function in these animals. In obese rats, D₁A receptors did not couple to G proteins, as shown by a lack of fenoldopam-sensitive [³⁵S] GTPγS binding. In addition, we observed, by using radioligand binding and immunoblotting, that dopamine recruited D₁A receptors to cell surface in lean Zucker rats but failed to do so in obese rats. Rosiglitazone treatment resulted in restoration of G-protein coupling of D₁A receptors and their recruitment by dopamine in obese rats similar to that seen in lean rats. Furthermore, SKF-38393 failed to increase natriuresis and diuresis in obese rats compared with lean rats. However, in rosiglitazone-treated obese rats, SKF-38393 elicited a diuretic and natriuretic response similar to that in lean rats. Collectively, these results suggest that insulin resistance may be responsible for impaired renal dopamine D₁A receptor signaling and function as treatment with an insulin-sensitizer, rosiglitazone, normalizes these parameters in obese Zucker rats. (Hypertension. 2004;43[part 2]:376-382.)

Key Words: dopamine ■ kidney ■ insulin resistance ■ obesity

Insulin resistance is a common factor that links obesity, type 2 diabetes, and hypertension. Obesity promotes insulin resistance, which can lead to development of type 2 diabetes and hypertension.¹,² Hypertension in patients with insulin resistance is usually associated with increased sodium retention,³,⁴ which is mainly owing to inability of the kidney to excrete sodium appropriately, leading to development of hypertension.

One of the hormones that increase sodium excretion is dopamine. Dopamine, by activating D₁A receptors, inhibits sodium transporters (Na,K-ATPase and Na,H-exchanger) in renal proximal tubules and promotes sodium excretion.⁵ Impaired D₁A receptor function in the kidney has been reported in hypertensive animals,⁶,⁷ as well as hypertensive humans,⁸ possibly contributing to development of hypertension.⁹ We have previously shown that there is reduced abundance and impaired G-protein coupling of cell surface D₁A receptors, resulting in inability of dopamine to inhibit sodium transporters in proximal tubules of insulin-resistant obese Zucker rats.¹⁰ Interestingly, rosiglitazone treatment of obese rats corrected the decrease in cell surface D₁A receptor abundance¹¹ and the impairment in dopamine-induced inhibition of sodium transporters¹¹ in proximal tubules. It is possible that impairment in G-protein coupling of D₁A receptors in obese rats is also corrected by rosiglitazone.

We have recently shown that dopamine increases abundance of cell surface D₁A receptors by recruiting them to the plasma membrane from an intracellular pool in proximal tubules.¹² This recruitment of D₁A receptors to the plasma membrane may augment natriuretic and diuretic response to dopamine in proximal tubules.¹² Dopamine-induced recruitment of D₁A receptors occurs through activation of D₁A receptors-Gs-cAMP pathway.¹² Because in obese rats G-protein coupling of D₁A receptors is impaired,¹⁰ it is likely that dopamine-induced D₁A receptor recruitment is impaired in obesity. Moreover, it is possible that rosiglitazone treatment corrects the defect in dopamine-induced D₁A receptor recruitment in obese rats by restoring receptor-G-protein coupling.

Dopamine D₁A receptor activation in proximal tubules leads to inhibition of sodium transporters, increasing sodium and water excretion.⁵ Because signaling of D₁A receptors is impaired in proximal tubules of obese rats compared with lean rats, it is likely that activation of D₁A receptors by an
agonist, SKF-38393, will not increase sodium and water excretion in obese rats. If rosiglitazone treatment restores D1A receptor signaling, it is possible that natriuretic and diuretic response to SKF-38393 is restored in obese rats treated with rosiglitazone.

In the present study, we first tested the hypothesis that coupling of D1A receptors to G proteins and recruitment of D1A receptors to the plasma membrane by dopamine are impaired in proximal tubules of obese rats compared with lean rats. This, in turn, contributes to reduced sodium and water excretion by SKF-38393. Second, we tested the hypothesis that correcting insulin resistance by an insulin sensitizer, rosiglitazone, restores coupling of D1A receptors to G proteins, dopamine-induced recruitment of D1A receptors to the plasma membrane, and sodium and water excretion by SKF-38393 in obese rats.

**Methods**

**Materials**

The source of materials is indicated in parenthesis after the name of materials. All the other chemicals were purchased from Sigma Chemical.

**Animals and Drug Treatment**

Male obese and lean rats (Charles River Laboratory, Wilmington, Mass) were maintained in animal care facility, with a 12-hour light/12-hour dark cycle and were provided standard rat chow (Purina Mills) and tap water ad libitum. Obese and lean rats (9 to 10 weeks old) were randomly assigned to either rosiglitazone maleate (3 mg/kg per day) treatment or vehicle (1% carboxymethylcellulose) (Purina Mills) and tap water ad libitum. Obese rats were significantly heavier than were lean rats in the fat pads and weight. The animals were treated daily by oral gavage for 15 days. All experimental protocols were approved by the University of Houston Institutional Animal Care and Use Committee. One set of rats, 6 in each group, was used for in vitro studies, and another set, 5 to 7 rats in each group, was used for in vivo renal function studies.

**Experimental Design for In Vitro Studies**

After the treatment period, proximal tubules were prepared from kidneys of all 4 groups of rats and treated with vehicle, dopamine, or forskolin. First, plasma membranes were prepared from vehicle- and dopamine-treated proximal tubules and were used for fenoldopam-stimulated [35S] GTPγS binding to determine G-protein coupling of D1A receptors on the plasma membranes. Second, plasma membranes from vehicle-, dopamine-, and forskolin-treated proximal tubules were used for [3H] SCH23390 binding and immunoblotting to study recruitment of D1A receptors.

**Blood Glucose and Plasma Insulin Analysis**

Rats were fasted overnight after treatment and were anesthetized with sodium pentobarbital (50 to 100 mg/kg IP). Blood (≈50 μL) was collected from aorta after a midline abdominal incision. Blood glucose values were determined by using Accu-Chek Advantage glucose monitoring system. Plasma insulin levels were measured by a rat insulin radioimmunoassay kit (Linco Research Inc).

**Isolation of Proximal Tubules and Drug Treatment**

After collecting blood samples, proximal tubules were prepared from kidney cortex by Ficoll gradient method as previously described.13,14 The enriched proximal tubules from each group of rats were suspended in modified Krebs-Henseleit buffer A13 (KHB A). The proximal tubular suspension (1 mg/mL) was incubated with KHB A, dopamine (10 nmol/L), or forskolin (1 μmol/L) at 37°C for 15 minutes. The drug treatment was terminated by rapid freezing in dry ice/acetone.

**Preparation of Plasma Membranes**

Plasma membranes were prepared from the frozen-thawed proximal tubular lysate as described before.11 The plasma membranes were used for radioligand binding, immunoblotting, and [3H] GTPγS binding experiments.

**Measurement of [35S] GTPγS Binding**

[35S] GTPγS binding was performed as described previously. Briefly, plasma membranes were incubated with either vehicle or various concentrations (10 nmol/L to 1 μmol/L) of fenoldopam to stimulate [35S] GTPγS binding, in presence of 6 nM of [3H] GTPγS (specific activity, 1250 Ci/mmol; Dupont NEN) and 5 μg plasma membrane for 1 hour. Nonspecific [3H] GTPγS binding was determined in presence of 100 μmol/L unlabeled GTPγS.

**Radioligand ([3H] SCH-23390) Binding in Plasma Membranes**

Radioligand binding experiments were performed according to previously described methods. Briefly, plasma membrane samples (50 μg) were incubated with 100 nmol/L [3H] SCH-23390 (specific activity, 86 Ci/mmol; Dupont NEN) at 25°C for 1.5 hours. Nonspecific binding was defined by using 10 μmol/L SCH-23390.

**Immunoblotting for D1A Receptor Protein in Plasma Membranes**

Immunoblotting analysis was performed in the plasma membrane samples by using D1A receptor specific antibodies (Alpha Diagnostics) as described earlier.16 The D1A receptor bands were densitometrically quantified by using Scion Image software provided by National Institutes of Health.

**Experimental Protocol for Renal Function Studies**

The effect of SKF-38393 on sodium and water excretion was determined in all 4 groups. The protocol consisted of 45-minute stabilization period after surgery followed by 5 consecutive 30-minute collection periods. During C1 and C2, saline alone was infused; during D, SKF-38393 (3 μg/kg per minute in saline) was infused; during R1 and R2 (recovery), only saline was infused. Surgical procedures were carried out as described earlier.17 Urine samples were collected throughout the 30-minute periods, and blood samples were collected at the end of each period. Renal function was evaluated as described previously.17

**Data Analysis**

Data are represented as mean±SEM of number (N) of experiments. The results were analyzed by using either Student unpaired t test, 1-way ANOVA, or repeated-measures ANOVA (functional studies) followed by Newman-Keuls multiple comparison test to assess significance of difference between the groups (lean and obese treated with either vehicle or rosiglitazone) and within groups. Statistical analysis was done by using Graph Pad Prism, version 3.02 (GraphPad Software). Statistical significance was considered at P<0.05.

**Results**

**Body Weight, Fasting Blood Glucose, and Plasma Insulin**

Obese rats were significantly heavier than were lean rats in both the treatment groups (Table). Rosiglitazone (3 mg/kg per day for 15 days) treatment caused significant weight gain in obese rats but not in lean rats (data not shown). Rosiglitazone treatment normalized fasting blood glucose levels and significantly reduced plasma insulin levels in obese treated rats.
Rosiglitazone Treatment Restores G-Protein Coupling of D1A Receptors on the Plasma Membrane in Obese Rats

In plasma membranes from obese control rats, fenoldopam did not stimulate \(^{35}\)S GTP\(^{\gamma}\)S binding (Figure 1A). However, there was a concentration-dependent stimulation of \(^{35}\)S GTP\(^{\gamma}\)S binding by fenoldopam at all concentrations (10 nM to 1 \(\mu\)M) in plasma membranes from obese treated rats (Figure 1B). There was no significant difference in percent increase in fenoldopam-stimulated \(^{35}\)S GTP\(^{\gamma}\)S binding in plasma membranes from lean control and treated rats. Basal \(^{35}\)S GTP\(^{\gamma}\)S binding in plasma membranes from lean and obese treated rats was elevated 5-fold compared with that in lean and obese control rats.

Rosiglitazone Treatment Restores Dopamine-Induced Recruitment of D1A Receptors in Obese Rats

In lean control rats, dopamine increased \(^{3}H\) SCH-23390 binding (Figure 2A) and D1A receptor protein density (Figure 3A) in plasma membranes, indicating recruitment of D1A receptors to the plasma membrane. In contrast, dopamine increased neither \(^{3}H\) SCH-23390 binding (Figure 2B) nor D1A receptor protein density (Figure 3B) in plasma membranes from obese control rats. The increase in specific \(^{3}H\) SCH-23390 binding or in D1A receptor protein density in plasma membranes by dopamine in lean treated rats (Figures 2C and 3C) was not significantly different from that in lean control rats (Figures 2A and 3A). Rosiglitazone treatment of obese rats restored increase in specific \(^{3}H\) SCH-23390 binding (Figure 2D) and D1A receptor protein density (Figure 3D) in plasma membranes when proximal tubules were treated with dopamine compared with KHB A.

Forskolin Recruits D1A Receptors to the Plasma Membrane in Obese Rats

Recruitment of D1A receptors occurs via activation of D1A receptor–cAMP pathway. To determine whether bypassing the D1A receptors and increasing cAMP by direct activation of adenyl cyclase can recruit D1A receptors, we performed \(^{3}H\) SCH-23390 binding and immunoblotting experiments in plasma membranes isolated from KHB A–treated or forskolin-treated proximal tubules from all 4 groups of rats. In obese control rats, specific \(^{3}H\) SCH-23390 binding (Figure 2B) and D1A receptor protein density (Figure 3B) in plasma membranes increased by \(~100\) fmole/mG protein and 1 U density, respectively, when proximal tubules were treated with forskolin compared with KHB A. However, this increase in \(^{3}H\) SCH-23390 binding and D1A receptor protein density in obese control rats was lower than that (\(~150\) to 200 fmole/mG protein and \(~1.7\) to 2.2 U density) in other 3 groups of rats (Figures 2A, 2C, and 2D and Figures 3A, 3C, and 3D). Similar results were obtained by treatment of

![Figure 1](http://hyper.ahajournals.org/)

**Figure 1.** Effect of dopamine on fenoldopam-sensitive \(^{35}\)S GTP\(^{\gamma}\)S binding in the plasma membranes from lean and obese Zucker rats treated with either vehicle or rosiglitazone. LC indicates lean control; OC, obese control; LT, lean treated; and OT, obese treated. Control and dopamine indicate treatment of proximal tubules. The basal \(^{35}\)S GTP\(^{\gamma}\)S binding (in pmole/mG protein) was 0.217±0.036 in LC, 0.162±0.031 in OC, 1.1±0.36 in LT, and 0.924±0.306 in OT rats. Dopamine treatment of proximal tubules did not change the basal \(^{35}\)S GTP\(^{\gamma}\)S binding significantly compared with vehicle treatment. *Fenoldopam-treated vs vehicle-treated groups; †dopamine-treated vs vehicle-treated groups; 1-way ANOVA followed by Newman-Keuls multiple comparison test (N=6).
proximal tubules with cell-permeable cAMP analog, dibutyryl cAMP, suggesting that effects of forskolin are mediated by activation of adenyl cyclase and subsequently increasing cAMP (data not shown).

**Newly Recruited D<sub>1A</sub> Receptors Couple to G Proteins**

To examine whether the newly recruited D<sub>1A</sub> receptors on the plasma membrane can couple to G proteins, [³⁵S] GTPγS binding in plasma membranes prepared from dopamine-treated proximal tubules was performed and compared with that in plasma membranes prepared from vehicle-treated proximal tubules in all 4 groups of rats. In obese control rats, fenoldopam failed to stimulate [³⁵S] GTPγS binding in plasma membranes prepared from dopamine-treated proximal tubules compared with that from KHB A-treated proximal tubules (Figure 1A). In contrast, fenoldopam, at all concentrations (10 nmol/L to 1 μmol/L), significantly increased [³⁵S] GTPγS binding in plasma membranes when proximal tubules were treated with dopamine compared with KHB A in obese treated rats (Figure 1B). The percentage increase in fenoldopam-stimulated [³⁵S] GTPγS binding in both KHB

---

**Figure 2.** Effects of dopamine and forskolin on [³⁵S] SCH-23390 binding in plasma membranes in lean and obese Zucker rats treated with either vehicle or rosiglitazone. Vehicle-treated lean rats (A), vehicle-treated obese rats (B), rosiglitazone-treated lean rats (C), and rosiglitazone-treated obese rats (D). *Dopamine-treated vs vehicle-treated groups; #1-way ANOVA followed by Newman-Keuls multiple comparison test (N=4).

**Figure 3.** Effects of dopamine and forskolin on the density of D<sub>1A</sub> receptor protein on plasma membranes in lean and obese Zucker rats treated with either vehicle or rosiglitazone. C indicates control (KHB A); D, dopamine; or F, forskolin treatment of proximal tubules. Vehicle-treated lean rats (A), vehicle-treated obese rats (B), rosiglitazone-treated lean rats (C), and rosiglitazone-treated obese rats (D). *Dopamine-treated vs vehicle-treated groups; #forskolin-treated vs dopamine-treated groups; 1-way ANOVA followed by Newman-Keuls multiple comparison test (N=4).
A–treated or dopamine-treated proximal tubules was similar in lean control and treated rats.

**SKF-38393 Increases Urine Flow, Urinary Sodium Excretion, and Fractional Excretion of Sodium in Rosiglitazone-Treated Obese Rats**

Intravenous administration of SKF-38393 (3 μg/kg per minute) failed to increase urine flow (UF), urinary sodium excretion ($U_{NaE}$), and fractional excretion of sodium ($FE_{Na}$) in obese control rats (Figure 4A-C and Figure 5A-C). However, in obese treated rats, SKF-38393 significantly increased UF, $U_{NaE}$, and $FE_{Na}$ in obese treated rats (Figure 5A through 5C). Rosiglitazone treatment did not alter the response to SKF-38393 in lean rats (data not shown). Mean arterial pressure was significantly elevated in obese control rats compared with lean control rats; rosiglitazone treatment reduced mean arterial pressure in obese control rats to normal levels seen in lean control and treated rats (Table). Heart rate was not significantly different in all 4 groups of rats (Table). No changes in mean arterial pressure and heart rate were produced by SKF-38393 in any of the groups (data not shown).

**Discussion**

Our results show that rosiglitazone restores natriuretic and diuretic response to SKF-38393, a D$_{1A}$ receptor agonist, by improving G-protein coupling of D$_{1A}$ receptors on the plasma membrane and dopamine-induced recruitment of intracellular D$_{1A}$ receptors to the plasma membrane in obese rats. Dopamine D$_{1A}$ receptors on the plasma membrane did not couple to G proteins in obese rats; however, rosiglitazone treatment restored the receptor–G-protein coupling in these animals. In addition, dopamine recruited D$_{1A}$ receptors to the plasma membrane in proximal tubules isolated from lean rats but failed to do so in obese rats. Rosiglitazone treatment in obese rats restored the ability of dopamine to recruit D$_{1A}$ receptors to the plasma membrane. These newly recruited D$_{1A}$ receptors were normal in terms of coupling to G proteins. It has been reported that dopamine-induced D$_{1A}$ receptor recruitment occurs through a D$_{1A}$ receptor–cAMP pathway. Therefore, when we bypassed the D$_{1A}$ receptors and directly elevated cAMP by forskolin, D$_{1A}$ receptors were recruited to the plasma membrane even in obese control rats, in which activation of D$_{1A}$ receptors by dopamine did not recruit D$_{1A}$ receptors. Furthermore, SKF-38393 failed to promote natriuresis and diuresis in obese rats. However, rosiglitazone treatment restored SKF-38393–mediated natriuresis and diuresis in obese rats.

Inability to recruit D$_{1A}$ receptors by dopamine in obese rats is owing to impaired D$_{1A}$ receptor–G-protein coupling. Previously, we have reported that dopamine-induced D$_{1A}$ receptor recruitment occurs through activation of D$_{1A}$ receptor-G$_{i}$/cAMP–PKA pathway in proximal tubules of Sprague Dawley rats. We show in the present study that coupling of D$_{1A}$ receptors to G proteins was impaired in proximal tubules of obese rats. Because of impaired coupling of these receptors to G proteins, it is likely that recruitment of D$_{1A}$ receptors would not occur in obese rats. As hypothesized, we observed that recruitment of D$_{1A}$ receptors by dopamine in obese control

![Figure 4](image1.png)

*Figure 4. UF (A), $U_{NaE}$ (B), and $FE_{Na}$ (C) before, during, and after 3 μg/kg per minute SKF-38393 in lean control (□) and obese control (□) rats. *Significant difference from control values (C1 + C2/2) within the same group; 1-way ANOVA followed by Newman-Keuls multiple comparison test. *Significant difference from lean control rats; Student unpaired t test (N=5).*

![Figure 5](image2.png)

*Figure 5. UF (A), $U_{NaE}$ (B), and $FE_{Na}$ (C) before, during, and after 3 μg/kg per minute SKF-38393 in obese control (○) and obese treated (△) rats. *Significant difference from control values (C1 + C2/2) within the same group; 1-way ANOVA followed by Newman-Keuls multiple comparison test. *Significant difference from obese control rats; Student unpaired t test (N=5 to 7).*
rats was impaired. When obese rats were treated with rosiglitazone, recruitment of D₁A receptors by dopamine was restored in proximal tubules of these animals. At the same time, rosiglitazone treatment restored G-protein coupling of the receptors in these animals. Therefore, correction in receptor–G-protein coupling by rosiglitazone treatment results in restoration of dopamine-induced D₁A receptor recruitment in obese treated rats.

Direct activation of adenylyl cyclase by forskolin led to recruitment of D₁A receptors to the plasma membrane in obese control rats, suggesting intact signaling through adenylyl cyclase in these animals. However, fewer D₁A receptors were recruited to the plasma membranes by treatment of proximal tubules with forskolin in obese control rats compared with other 3 groups. This may be owing to inability of forskolin to stimulate PKA in proximal tubules of obese rats. Reduced availability of D₁A receptors in the cell could not be a reason because in cytosol D₁A receptor density is not reduced in proximal tubules of obese control rats compared with lean control rats (data not shown). Interestingly, the fact that forskolin recruits D₁A receptors to the plasma membranes in obese rats argues for an alternative PKA-independent pathway for recruiting D₁A receptors to the plasma membrane. Improving insulin sensitivity with rosiglitazone restores forskolin-induced recruitment of D₁A receptors to the plasma membrane in obese rats.

Restoration of D₁A receptor signaling by rosiglitazone treatment is further seen as normalization of SKF-38393–induced natriuresis and diuresis in obese rats. In obese control rats, mean arterial pressure was significantly elevated. This could possibly contribute to pressure natriuresis resulting in increased basal U₅₀V. Moreover, higher weight and therefore higher sodium and water intake of obese rats may also cause increased sodium and water excretion as compared with that in lean rats. Similar to obese control rats, basal UF and U₅₀V were significantly greater in obese treated rats, even though rosiglitazone treatment reduces blood pressure in these animals. This could be explained by the fact that thiazolidinedione derivatives have been shown to increase U₅₀V. Attenuated natriuretic and diuretic response to SKF-38393 in obese control rats could be partly owing to inability of dopamine and D₁A receptor agonist to inhibit sodium transporters in proximal tubules. This may result from (1) reduced abundance of D₁A receptors on the plasma membrane, (2) impaired coupling of D₁A receptors to G proteins, and, subsequently, (3) impaired dopamine-induced recruitment of D₁A receptors to the plasma membrane in obese rats.

Our G-protein coupling experiments showed that there was a 5-fold increase in basal [³⁵S]GTPγS binding in plasma membranes from rosiglitazone-treated lean and obese rats. Activation of PPARγ receptors, a nuclear transcription factor, has been shown to increase transcription of several genes. It is possible that rosiglitazone, a PPARγ agonist, increases transcription of one or several G proteins, resulting in increased levels of total G protein pool. It is also possible that the ratio of high-affinity (GTP-bound) to low-affinity (GDP-bound) G proteins is increased after rosiglitazone treatment, resulting in an increase in basal [³⁵S]GTPγS binding. Because rosiglitazone increases basal [³⁵S]GTPγS binding in both obese and lean rats, this effect is not caused by its insulin sensitizing effect. However, this effect of rosiglitazone does not contribute to restoration of D₁A receptor function in obese rats.

Rosiglitazone improves insulin sensitivity in animals as well as patients. This has been evaluated by oral glucose tolerance test or hyperinsulinemic-euglycemic clamp techniques. However, hyperglycemia with hyperinsulinemia is an indicator of insulin resistance, and a decrease in fasting blood glucose and plasma insulin levels suggests an improvement in insulin sensitivity. Our observations of reduction in fasting blood glucose and plasma insulin levels in obese treated rats indicate that the dose and duration of rosiglitazone treatment (3 mg/kg per day for 15 days) was adequate for improving insulin sensitivity. Previously, we and others have used higher doses of rosiglitazone (≥10 mg/kg per day) for longer duration (28 days) to improve insulin sensitivity. However, the present study as well as some recent reports show that even lower doses (3 to 5 mg/kg per day) of rosiglitazone for 7 to 14 days improve insulin sensitivity to the same extent. In obese rats, fasting blood glucose levels were completely normalized to levels seen in lean rats, and plasma insulin values were reduced to similar extent with both treatment regimens of rosiglitazone.

Hyperglycemia has been shown to be responsible for decreased renal function before it causes an increase in blood pressure. On the other hand, increases in insulin cause decreased renal sodium and water excretion by increasing renal sympathetic activity, in addition to a direct effect on sodium transporters. Hyperinsulinemia and hyperglycemia may have a synergistic effect on D₁A receptor dysfunction. Although plasma insulin levels are not reduced to normal levels by rosiglitazone, coupling of D₁A receptors to G proteins and their recruitment to the plasma membrane are restored to normal in obese rats. This suggests that moderately elevated insulin levels in obese rats treated with rosiglitazone may not influence D₁A receptor recruitment and coupling. Moreover, rosiglitazone, by improving insulin sensitivity, lowers triglyceride and free fatty acid levels in obese rats. It is possible that the triglyceride-lowering effect of rosiglitazone is partly responsible for normalizing dopaminergic response in obese rats. The role of insulin and glucose in D₁A receptor dysfunction has been established earlier; however, what role triglycerides and free fatty acids play in D₁A receptor signaling remains to be elucidated.

In conclusion, we have shown that defective G-protein coupling of D₁A receptors in obese Zucker rats is corrected by treatment with rosiglitazone. Moreover, dopamine-induced recruitment of D₁A receptors to the plasma membrane is impaired in obese Zucker rats, and rosiglitazone treatment of these rats corrects this defect. Further, impaired D₁A receptor signaling in renal proximal tubules contributes to reduced natriuretic and diuretic response to D₁A receptors agonist, SKF-38393 in obese Zucker rats. Rosiglitazone treatment corrects D₁A receptor signaling and, subsequently, natriuretic and diuretic response to SKF-38393 in these animals.

Perspectives
The pathogenesis of obesity-related hypertension is extremely complex and certainly cannot be ascribed to dysfunc-
tion of one pathway. Overactivity of antinatriuretic hormones and underactivity of natriuretic hormones can lead to increased sodium retention. Dopamine, a natriuretic hormone, plays a central role in regulation of sodium homeostasis. Moreover, dopamine enhances the effects of other natriuretic hormones, such as atrial natriuretic peptide and counteracts the effects of antinatriuretic hormones such as angiotensin II. Increasing sodium retention in obesity may be owing to loss of dopaminergic control over antinatriuretic and natriuretic hormones. The loss of dopaminergic control could result from impaired $D_1\alpha$ receptor G-protein coupling, recruitment, and function caused by insulin resistance. Therefore, rosiglitazone, in addition to being of therapeutic benefit in insulin resistance, may also prevent abnormality in renal sodium handling associated with renal $D_1\alpha$ receptor dysfunction as it restores coupling of $D_1\alpha$ receptors to G proteins, $D_1\alpha$ receptor recruitment to the plasma membrane, and natriuretic and diuretic response to $D_1\alpha$ receptor activation. Because insulin resistance develops before diseases such as diabetes and hypertension appear, rosiglitazone treatment offers a valuable therapeutic approach in prevention and treatment of these diseases.

Acknowledgment
This project was supported by National Institutes of Health (NIDDK) grant DK-58743.

References
Rosiglitazone Restores G-Protein Coupling, Recruitment, and Function of Renal Dopamine D_1A Receptor in Obese Zucker Rats
Meghna Trivedi, Aditi Marwaha and Mustafa Lokhandwala

Hypertension. 2004;43:376-382; originally published online January 12, 2004; doi: 10.1161/01.HYP.0000111587.51185.fe

Hypertension is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 2004 American Heart Association, Inc. All rights reserved.
Print ISSN: 0194-911X. Online ISSN: 1524-4563

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://hyper.ahajournals.org/content/43/2/376

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in Hypertension can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to Hypertension is online at:
http://hyper.ahajournals.org//subscriptions/