Angiotensin-II Type 1 Receptor–Mediated Hypertension in D4 Dopamine Receptor–Deficient Mice


Abstract—Dopamine receptors are important in systemic blood pressure regulation. D1 receptors are expressed in the kidney and brain, but their role in cardiovascular regulation is unknown. In pentobarbital-anesthetized mice, systolic and diastolic blood pressures were elevated in sixth-generation D4 receptor–deficient (D4−/−) mice and in tenth-generation D4−/− mice compared with D4 wild-type (D4+/+) littermates. The conscious blood pressures measured via a chronic (femoral) catheter or telemetry (carotid) were also higher in D4−/− mice than in D4 littermates. Basal renal and plasma renin concentrations were similar in the 2 mouse strains. The protein expression of angiotensin II type 1 receptor was increased in homogenates of kidney (330±53%, n=5) and brain (272±69%, n=5) of D4−/− mice relative to D4+/+ mice (kidney: 100±12%, n=5; brain: 100±32%, n=5). The expression of the receptor in renal membrane was also increased in D4−/− mice (289±28%, n=8) relative to D4+/+ mice 100±14%, n=10. In contrast, the expression in the heart was similar in the 2 strains. Bolus intravenous injection of angiotensin II type 1 receptor antagonist losartan initially decreased mean arterial pressures to a similar degree in D4−/− and D4+/+ littermates. However, the hypotensive effect of losartan dissipated after 10 minutes in D4−/− mice, whereas the effect persisted for >45 minutes in D4−/− mice. We conclude that the absence of the D4 receptor increases blood pressure, possibly via increased angiotensin II type 1 receptor expression. (Hypertension. 2006;47:288-295.)

Key Words: dopamine receptors, angiotensin II mice hypertension angiotensin II endothelin vasopressins

Essential hypertension is a major risk factor for the development of cardiovascular disease.1 It is a heterogeneous disease in which both genetics and environment influence blood pressure.2 Dopamine affects cardiovascular regulatory mechanisms by its actions on renal hemodynamics and ion and water transport and by its regulation of hormones and humoral agents, such as aldosterone, catecholamines, endothelin, prolactin, proopiomelanocortin, renin, and vasopressin. In addition, dopamine can control blood pressure by acting on neuronal cardiovascular centers, heart, and arterial and venous vessels.3-9 Dopamine exerts its actions by occupation of the D1-like (D1 and D5) and D2-like (D2, D3, and D4) family of cell surface G protein–coupled receptors. We have reported that disruption of the D1, D2, D3, and D4 receptors leads to hypertension in mice, via specific pathophysiologic mechanisms.10-13 The cardiovascular consequences of disruption of the D4 receptor have not been reported. D4 receptors are expressed in the heart, renal collecting ducts, juxtaglomerular cells, and in brain nuclei known to affect blood pressure, but their role in cardiovascular regulation is unknown.14-17 Loci near the D4 receptor gene (11p15.5) have been linked to hypertension,18,19 and polymorphisms of the D4 receptor gene are associated with hypertension.20 Therefore, we tested the hypothesis that the D4−/− mouse has a cardiovascular phenotype.

Methods

Generation of D4 Dopamine–Receptor Mutant Mice

The original F1 hybrid strain (129/Sv×C57BL/6) carrying a mutant form of the D4 dopamine receptor was initially generated and backcrossed to C57BL/6 mice. Heterozygous (D4+/−) mice were mated to obtain D4−/− and D4+/+ littermates, and the D4−/− mice were backcrossed with C57BL/6 mice to obtain sixth- and tenth-generation mice in the Department of Physiology and Pharmacology, Oregon Health and Science University.21 We used sixth-generation mice for acute studies and tenth-generation mice for chronic and immunoblotting studies. All of the animals were genotyped21 and treated in accordance with National Institutes of Health guidelines for ethical treatment and handling of animals in research.

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Blood Pressure Measurement

Blood Pressures Under Anesthesia
Mice were anesthetized with pentobarbital (50 mg/kg IV) and tracheotomized (PE100). Catheters (PE50 heat-stretched to 180-μm tip) were inserted into the femoral vessels for fluid administration and blood pressure monitoring. Blood pressures were recorded (CardioMaxII, Columbus Instrument) after 1 hour of equilibration.

Blood Pressures Without Anesthesia
Conscious blood pressures were measured in 2 sets of mice. In the first set of studies, blood pressures were measured via a femoral artery catheter, coated with 5% heparin complex, threaded upward and out of a 5-mm incision at the nape of the neck. The catheter was flushed immediately (1/2 mg plasmin and 1000 U heparin/mL of sterile saline) and every 2 days thereafter. One to 3 days after catheter placement, blood pressures were measured in freely moving, unanesthetized mice. In the second set of studies, TA-PAC20 catheter placement, blood pressures were measured in freely moving, sterile saline) and every 2 days thereafter. One to 3 days after

Acute Saline Loading Study
After a 60-minute stabilization period after the catheter insertion and a baseline 60-minute period for blood pressure measurement, a normal saline load equivalent to 5% body weight was infused intravenously for 30 minutes. Urine was collected during saline loading via suprapubic cystostomy for another 30 minutes; 3 more urine collection periods of 60 minutes each were obtained after loading. Blood (50 μL) was obtained from the femoral artery before the load and at the end of the last urine collection. The kidneys were obtained for determination of renin concentration.

Acute Drug Infusion Studies
In additional experiments, several antagonists of receptors known to influence blood pressure were infused via a central venous catheter, and blood pressure and heart rate were monitored. The drugs were α-adrenergic antagonist phentolamine (5 ng/kg per minute), angiotensin II type 1 receptor (AT1) antagonist losartan (3 mg/kg per minute for 10 minutes), endothelin B receptor (ETB) antagonist BQ788 (6.6 μg/kg per minute for 15 minutes), and V1 vasopressin receptor (V1) antagonist [1-(β-mercapto-β, β-cyclopentamethylenepropionic acid)-2-(O-methyl)-tyrosine] arginine vasopressin (10 μg/kg over 30 s). The rationale and dosages of these drugs have been validated. The dose of losartan had been shown previously to have an effect of limited duration in wild-type mice. We also studied the acute effect on blood pressure of varying doses of bolus intravenous injections of angiotensin II (0.1, 0.3, 1, 3, and 10 ng/kg per mouse). Mice were euthanized with pentobarbital (100 mg/kg) at the end of the experiments.

Measurement of Renin, Na+/K+-ATPase, and Catechols
Plasma and renal renin concentrations were assessed by radioimmunoassay measuring the generation of angiotensin 1-10 Na/K+-ATPase activity in renal cortex or medulla was measured as the ouabainsensitive dephosphorylation of (tris)-p-nitrophenyl phosphate by K+-p-nitrophenyl phosphatase. The kidneys were homogenized with 0.1 mol/L HClO4 and centrifuged at 6000g for 20 minutes at 4°C. The supernatant and urine catechol concentrations were determined by high-performance liquid chromatography and electrochemical detection.

Chronic Sodium Balance Study With Ration Feeding in Metabolic Cages
The mice were maintained in metabolic cages to allow quantitative urine collections and ration feeding, modified from a rat-diet protocol. The baseline sodium diet (5 g/25 g body weight per day, TD.90228, Harlan Teklad) consisted of a gelled mixture of distilled water (10 mL/5 g of mouse chow), agar (0.04 g/10 mL of water), and 0.4% NaCl (added before gelation). The sodium replete diet was the same, except for the addition of 0.8% NaCl. All of the animals received the same amount of gelled food, determined by weighing the gelled mixture. On day 3, the mice were ration-fed 0.4% NaCl gelled food. On day zero, the ration was changed to 0.8% NaCl for 1 week. Seven days later, the mice were euthanized after blood pressures were recorded with anesthesia. Serum and urinary samples were analyzed for Na, K, Cl (E4A Electrolyte system, Beckman) and creatinine concentrations (Creatinine Analyzer2,

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Figure 1. Blood pressures (BPs) of D4+/+ and D4−/− mice measured under pentobarbital anesthesia. BPs were obtained in mice of both genders from the sixth generation (D4+/+: n=27; D4−/−: n=18, 8- to 12-months old; A) and tenth generation (4- to 6-months old; 7 pairs; B). Data are mean±SE. *P<0.05 vs D4+/+ mice, t test.

Figure 2. BPs of conscious D4−/− and D4+/+ mice (tenth generation, 4- to 5-months old). (A) BPs of mice measured via a chronic femoral artery catheter (3 pairs). The bihourly SBP (B) and nighttime SBP and MAP (C) of mice measured by telemetry (3 pairs). Data are mean±SE. *P<0.05 vs D4+/+ mice, t test, with Bonferroni correction (B).

Blood Pressures - 6th Generation

Blood Pressures - 10th Generation

Conscious Blood Pressures - 10th Generation

Telemetry
TABLE 1. The Effect of Saline Loading on Renal Function in D4+/+ (n=6) and D4−/− (n=18) Mice

<table>
<thead>
<tr>
<th>Collection Periods</th>
<th>GFR (nL/g kidney weight per minute)</th>
<th>V (μL/min)</th>
<th>UNaV (nEq/min)</th>
<th>FENa (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baseline</td>
<td>1225±256</td>
<td>1598±123</td>
<td>1.21±0.09*</td>
<td>1.11±0.07*</td>
</tr>
<tr>
<td>Loading</td>
<td>1390±107</td>
<td>1823±136</td>
<td>2.94±0.27</td>
<td>4.28±0.84</td>
</tr>
<tr>
<td>Postload 1</td>
<td>1240±77</td>
<td>1493±203</td>
<td>1.72±0.33*</td>
<td>3.36±0.72</td>
</tr>
<tr>
<td>Postload 2</td>
<td>1146±167</td>
<td>1304±80*</td>
<td>1.32±0.06*</td>
<td>2.53±0.52</td>
</tr>
<tr>
<td>Postload 3</td>
<td>1260±133</td>
<td>1247±102*</td>
<td>1.19±0.10*</td>
<td>1.32±0.21*</td>
</tr>
</tbody>
</table>

Data are mean±SE. Each period lasted 60 minutes. MAPs were not affected by time or saline infusion. GFR, glomerular filtration rate; V, urine flow; UNaV, sodium excretion; FENa, fractional sodium excretion.

*P<0.05 vs loading period or postload 1, ANVR, Newman Keuls test.

TABLE 2. The Effect of Saline Loading on Urinary Catechols (pg/min) in D4+/+ (n=5) and D4−/− (n=17) Mice

<table>
<thead>
<tr>
<th>Collection Periods</th>
<th>L-DOPA</th>
<th>DOPAC</th>
<th>Dopamine</th>
<th>Epinephrine</th>
<th>Norepinephrine</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baseline</td>
<td>10.6±2.3</td>
<td>15.2±2.4</td>
<td>105±26</td>
<td>82±9</td>
<td>498±87*</td>
</tr>
<tr>
<td>Loading</td>
<td>15.1±4.0</td>
<td>18.0±3.9</td>
<td>231±61</td>
<td>149±31</td>
<td>884±107</td>
</tr>
<tr>
<td>Postload 1</td>
<td>14.0±4.5</td>
<td>20.5±5.5</td>
<td>183±67</td>
<td>143±37</td>
<td>608±112*</td>
</tr>
<tr>
<td>Postload 2</td>
<td>12.5±3.7</td>
<td>31.3±14.0</td>
<td>131±51</td>
<td>144±44</td>
<td>492±37*</td>
</tr>
<tr>
<td>Postload 3</td>
<td>9.0±2.3</td>
<td>33.7±9.7</td>
<td>140±59</td>
<td>151±43</td>
<td>518±86*</td>
</tr>
</tbody>
</table>

Data are mean±SE. Each period lasted 60 minutes. DOPA indicates dihydroxyphenylalanine; DOPAC, dihydroxyphenylacetic acid.

*P<0.05 vs loading.

#P<0.05 vs baseline, ANVR, Newman Keuls test.
Renal and Plasma Renin Concentrations in D4\textsuperscript{+/+} Mice

Renal renin concentrations (µg A/g kidney per hour) were similar in D4\textsuperscript{+/+} mice (57.5±7.2, n=18) and D4\textsuperscript{−/−} mice (70.2±18.3, n=5). Plasma renin concentrations were also not significantly different between D4\textsuperscript{+/+} (3835±097 ng A/mL per hour, n=18) and D4\textsuperscript{−/−} (3540±2881, n=5) mice.

Na\textsuperscript{+}/K\textsuperscript{+}-ATPase Activity

Renal cortical and medullary Na\textsuperscript{+}/K\textsuperscript{+}-ATPase activities were not significantly changed in D4\textsuperscript{+/+} mice (cortex=34.2±0.5 nmol Pi/mg protein per minute, medulla=34.2±0.5, n=17) compared with D4\textsuperscript{−/−} mice (cortex=33.4±2.0 nmol Pi/mg protein per minute, medulla=31.5±2.6, n=5). The D4 receptor agonist, SKF81297 (1 μmol/L), decreased Na\textsuperscript{+}/K\textsuperscript{+}-ATPase activity to a similar extent in cortex (D4\textsuperscript{+/+}=25.98±2.91%, D4\textsuperscript{−/−}=21.60±1.41%) and medulla (D4\textsuperscript{+/+}=23.17±3.34%, D4\textsuperscript{−/−}=20.02±1.37%; P>0.05, ANOVA) in the 2 mouse strains.

Role of Blood Pressure–Regulating Systems in the Hypertension of D4\textsuperscript{−/−} Mice

Bolus intravenous injection of losartan decreased MAP promptly and to a similar degree initially in both strains. However, the effect dissipated quickly with recovery toward baseline 10 minutes after the injection in D4\textsuperscript{−/−} mice, whereas the hypotensive effect of losartan persisted for >45 minutes in D4\textsuperscript{+/+} mice (Figure 3a). In contrast, the V1 vasopressin receptor antagonist (Figure 3b) and ETB receptor antagonist (Figure 3c) increased MAP in D4\textsuperscript{−/−} but not in D4\textsuperscript{+/+} mice, albeit modestly. Blockade of endothelin A receptors had no effect on blood pressure in either mouse strain, whereas blockade of α-adrenergic receptors with phentolamine decreased blood pressure to a similar extent in D4\textsuperscript{−/−} and D4\textsuperscript{+/+} mice (Figure 3d). Bolus injections of angiotensin II (0.1, 0.3, 1, 3, and 10 ng/kg per mouse) dose-dependently increased blood pressure in both D4\textsuperscript{−/−} mice [Yc=16.45+(5.6×dose), n=5] and D4\textsuperscript{+/+} mice [Yc=13.89+(3.37×dose), n=4; P<0.05]. Although, a higher MAP was found in D4\textsuperscript{−/−} mice than in D4\textsuperscript{+/+} mice in each dose group.

Figure 4. Effect of angiotensin II on MAP in anesthetized D4\textsuperscript{+/+} and D4\textsuperscript{−/−} mice. Angiotensin II dose-dependently (0.1, 0.3, 1, 3, and 10 ng/kg per mouse) increased the blood pressure in both D4\textsuperscript{−/−} [Yc=16.45+(5.6×dose), n=5] and D4\textsuperscript{+/+} mice [Yc=13.89+(3.37×dose), n=4]. MAPs were always higher in D4\textsuperscript{−/−} than in D4\textsuperscript{+/+} mice (t P<0.05, ANOVA, Newman Keuls test). However, the dose-dependent increase in MAP caused by angiotensin II was greater in D4\textsuperscript{−/−} than in D4\textsuperscript{+/+} mice and reached significance at 10 ng (t P<0.05, t test).
TABLE 3. Body Weight, Blood Pressure, Creatinine Clearance, and Sodium and Water Excretion in Ration-Fed Mice

<table>
<thead>
<tr>
<th>Variable</th>
<th>D4+/− (n=5)</th>
<th>D4−/− (n=5)</th>
</tr>
</thead>
<tbody>
<tr>
<td>BW (g) day 3</td>
<td>26.8±0.5</td>
<td>20.7±0.7*</td>
</tr>
<tr>
<td>Day 0</td>
<td>27.6±0.7</td>
<td>21.4±0.6*</td>
</tr>
<tr>
<td>Day 10</td>
<td>27.8±0.5</td>
<td>22.3±0.4*</td>
</tr>
<tr>
<td>SBP (mm Hg)</td>
<td>96±3</td>
<td>119±1*</td>
</tr>
<tr>
<td>DBP (mm Hg)</td>
<td>81±5</td>
<td>90±2*</td>
</tr>
<tr>
<td>Serum Na (mM)</td>
<td>150±2</td>
<td>151±2</td>
</tr>
<tr>
<td>Creatinine clearance (mL/g BW per day)</td>
<td>0.6±1.0</td>
<td>4.8±0.7</td>
</tr>
<tr>
<td>Water excretion (mL/g BW per day)</td>
<td>0.10±0.02</td>
<td>0.08±0.02</td>
</tr>
<tr>
<td>Na excretion (mmol/g BW per day)</td>
<td>0.017±0.002</td>
<td>0.014±0.002</td>
</tr>
</tbody>
</table>

Values are mean±SE. BW indicates body weight; BP, blood pressure. Mice: female, 6 months old. BP's were measured under pentobarbital anesthesia. Salt intake: 0.8% NaCl for 10 days, 5 g mouse chow plus 10 mL water/25g of BW per day.

*P<0.05 vs D4+/−, t test.

mice at any dose, the MAP tended to increase to a greater extent in D4+/− than in D4−/− mice, reaching significance at 10 ng (ΔMAP, mm Hg, D4+/−: 151±4, n=5; D4−/−: 151±4, n=4; P<0.05, t test; Figure 4).

Chronic Sodium Balance Study

Age- and gender-matched D4+/− mice and D4−/− littermates (tenth-generation) and C57/BL6 mice (Taconic, Germantown, NY) were housed in the animal facility under the same conditions for >1 month before use. Backcrossing to the sixth generation or to more than the tenth generation results in a C57/BL6 genetic background >98%30 or >99% congenic, respectively. In agreement with data shown in Figure 1A and 1B, SBP and DBP under pentobarbital anesthesia were higher in D4−/− than in D4+/− mice (female, 6 months old). D4−/− mice weighed less than D4+/− mice possibly because of the ration feeding (sodium intake = 0.8% NaCl) in metabolic cages. Sodium and water excretions (normalized by body weight) tended to be lower in D4−/− than in D4+/− mice, but the differences did not reach statistical significance. Creatinine clearance and serum sodium concentration were similar in D4+/− and D4−/− mice (Table 3).

AT1 Receptor Protein Expression

To determine a mechanism for the involvement of the AT1 receptor in the hypertension of D4−/− mice, we measured AT1 receptor protein expression in the ration-fed tenth-generation D4+/− and C57/BL6 mice. AT1 receptor expression (45 kDa) in whole-kidney homogenates of D4−/− mice was increased (330±53%, normalized by the band density of D4+/− set to 100%, n=5; P<0.05) compared with D4+/− mice (100±12%, n=5; Figure 5A). AT1 receptor expression was also increased in whole-brain homogenates of D4−/− mice (272±69%; P<0.05) compared with D4+/− mice (100±32%; Figure 5B). In contrast, there were no differences in AT1 expression in whole heart homogenates between the 2 mouse strains (Figure 5C).

In additional experiments, tenth-generation D4+/− and D4−/− mice littermates (mixed gender, 4 to 5 months old) were studied to achieve a >99% C57/BL6 genetic background.31 There were no differences in body weight between D4+/− mice (31±2 g, n=10) and D4−/− littermates (27±2 g, n=8), whereas blood pressures were higher in D4+/− mice than in D4−/− littermates (Figure 1B). AT1 receptor expression was measured in kidney membrane fractions; receptors in the cytosol would not be responsive to angiotensin II stimulation. AT1 receptor expression in kidney membrane fractions was increased in D4−/− mice relative to D4+/− littermates (D4+/− mice: 100±14%; D4−/− mice: 289±28; % of D4+/−; P<0.05; Figure 6), in agreement with studies using renal homogenates.

Discussion

The present studies show that the complete lack of D4 dopamine receptors resulted in increased blood pressure. Furthermore, the increased blood pressure in the mutant mice was associated with a prolonged depressor response caused by the AT1 receptor antagonist losartan. The transient effect of losartan in the D4−/− mice was related to the dose and similar to the hypertensive effect in D4+/− mice.12 The prolonged hypotensive effect of losartan in the D4−/− mice is also
Evidence has accumulated that the increase in blood pressure caused by central AT1 activation is mediated with37–40 or without41–44 activation of the sympathetic nervous system. In the present study, α-adrenergic blockade with phentolamine decreased blood pressure to a similar extent in D4−/− and D4+/+ mice. This observation may be specific to the D4−/− mice, because the α-adrenergic blockade decreased blood pressure to a greater extent in D3−/− and D3+/+ mice than in their wild-type counterparts.11,13 There were also no differences in renal or urinary catechol excretion between D4+/+ and D4−/− mice. The pressor effect mediated by AT1 receptors expressed in the central nervous system has also been shown to be mediated, in part, by activation of central V1 receptors.38,45,46 The increased blood pressure caused by disruption of the D4 dopamine receptor is characterized by activation of central V1 vasopressin receptors.13 However, in the current study, V1 vasopressin receptor blockade did not decrease but actually produced a slight increase in blood pressure in D4−/− mice, suggesting a different mechanism to be involved.

Endothelin receptors may mediate pressor and depressor effects.37–49 We have reported that disruption of the D2 dopamine receptor in mice increased blood pressure possibly via the vasoconstrictor ETB2 (presumably in vascular smooth muscle or in brain centers).11 The D2 receptor, like the D2 receptor, is not expressed in endothelial cells.50 The pressor effect mediated by AT1 receptors in mice increased blood pressure expression corrected for α-actin was decreased in D4−/− mice (n=10) relative to D4+/+ littermates (n=8). *P<0.05 vs D4+/+, t test.

Figure 6. Immunoblots of AT1 receptors of renal membranes from tenth-generation D4−/− mice and D4+/+ littermates. Top, immunoblot of AT1 receptors in membranes from whole kidneys of D4−/− (left, n=6) and D4+/+ (right, n=4) littermates. Bottom, bar graphs; AT1 receptor expression corrected by α-actin was increased in D4−/− mice (n=10) relative to D4+/+ littermates (n=8). *P<0.05 vs D4+/+, t test.

similar to the effect in D3−/− mice.12 The duration of the hypertensive effect was longest in the D3−/− mice, intermediate in the D3+/− mice, and shortest in the D3+/+ mice. There were no differences in plasma or renal renin concentrations between D4−/− and D4+/+ mice. This contrasts with the elevation of renal renin concentration in the hypertensive D3−/− mice.12

The mechanism underlying the AT1-dependent high blood pressure in the D4−/− mice is not readily apparent. Aberrant interactions between D4 and AT1 receptors were found in spontaneously hypertensive rats.28 It is possible that the D4 receptor similarly regulates the AT1 receptor. D4 receptors are expressed in brain areas that contribute to the regulation of blood pressure, for example, nucleus tractus solitarius.32–35 Because the differential effect of AT1 receptor blockade on blood pressure became significant at later rather than earlier time points, we presumed that the AT1 receptor-mediated hypertension in D4−/− mice is, in part, central in origin. This may also explain why the acute hypertensive effect of angiotensin II was reduced in D4 receptor-deficient mice compared with wild-type mice. However, AT1 protein expression in whole brain and renal homogenates and membranes of D4−/− mice were increased relative to D4+/+ mice. The finding that the increase in AT1 receptor protein in the kidneys of D4−/− mice was not associated with sodium retention may indicate a counteracting effect of another receptor stimulated by angiotensin II outside the central nervous system, possibly the AT2 receptor.36 The absence of a difference in AT1 receptor expression in the heart of D4−/− and D4+/+ mice also suggests tissue-specific D4 regulation of the AT1 receptor. These remain speculative at this time.

Perspectives
This study demonstrates that disruption of the D4 dopamine receptor results in increased blood pressure that may be related to activation of AT1 receptors in the brain. However, determination of AT1 receptor expression and function in specific brain nuclei are needed to understand how the D4
receptor interacts with the AT₁ receptor in the central regulation of blood pressure. AT₁ receptors are also increased in the kidneys of D₄/−/− mice, but these mice do not have an impaired ability to excrete an acute sodium load. It is possible that D₄/−/− mice may not be able to excrete a chronic sodium load, but that remains to be determined. Because the D₃ receptor gene locus is linked to and D₄ receptor variants are associated with hypertension, the relevance of the D₁ receptor in the pathogenesis of human essential hypertension needs to be evaluated.

Acknowledgments

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References


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In the article by Bek et al in the February 2006 issue of Hypertension (Bek MJ, Wang X, Asico LD, Jones JE, Zheng S, Li X, Eisner GM, Grandy DK, Carey RM, Soares-da-Silva P, Jose PA. Angiotensin-II type 1 receptor–mediated hypertension in D₄ dopamine receptor–deficient mice. Hypertension 2006;47:288–292) the symbols in the legend for Figure 2B were incorrect. The correct Figure 2 appears below. The authors regret the error.

**Figure 2.** BPs of conscious D₄⁺/+ and D₄⁻/⁻ mice (tenth generation, 4- to 5-months old). (A) BPs of mice measured via a chronic femoral artery catheter (3 pairs). The bihourly SBP (B) and nighttime SBP and MAP (C) of mice measured by telemetry (3 pairs). Data are mean±SE. *P<0.05 vs D₄⁺/+ mice, t test, with Bonferroni correction (B).