Differential Effects of T- and L-Type Calcium Antagonists on Glomerular Dynamics in Spontaneously Hypertensive Rats

Yasuyuki Nakamura, Hidehiko Ono, Edward D. Frohlich

Abstract—To determine whether there is a difference in the effects of T- and L-type calcium antagonists on systemic, renal, and glomerular hemodynamics, the pathological changes of N⁵-nitro-L-arginine methyl ester (L-NAME)–exacerbated nephrosclerosis and clinical alterations were investigated in spontaneously hypertensive rats (SHR). Seven groups of 17-week-old male SHRs were studied: Group 1, control; Group 2, mibefradil, 50 mg · kg⁻¹ · d⁻¹; Group 3, L-NAME in drinking water, 50 mg/L; Group 4, L-NAME (50 mg/L) plus mibefradil (50 mg · kg⁻¹ · d⁻¹); Group 5, L-NAME (50 mg/L) plus amlodipine (10 mg · kg⁻¹ · d⁻¹); Group 6 and 7, L-NAME (50 mg/L) for 3 weeks followed by mibefradil (50 mg · kg⁻¹ · d⁻¹) or amlodipine (10 mg · kg⁻¹ · d⁻¹), respectively, for the subsequent 3 weeks. Both the T- and L-channel calcium antagonists similarly reduced mean arterial pressure and total peripheral resistance index. These changes were associated with significant decreases in afferent and efferent glomerular arteriolar resistances and the ultrafiltration coefficient (P<0.01). Furthermore, the histopathological glomerular and arterial injury scores and urinary protein excretion were also significantly improved (P<0.01), and left ventricular and aortic masses were significantly diminished in all treated groups. Both drugs, mibefradil and amlodipine, had effects of increasing the single-nephron glomerular filtration ratio (SNGFR), and single-nephron plasma flow (SNPF), and of reducing glomerular afferent arteriolar resistance and urinary protein excretion. Thus, the T-type (mibefradil) and L-type (amlodipine) calcium antagonists each prevented and reversed the pathophysiological alterations of L-NAME–exacerbated hypertensive nephrosclerosis in SHR. The T-type calcium antagonist (mibefradil) seemed to have been more effective than the L-type amlodipine antagonist and it produced a greater reduction in afferent arteriolar resistance while preserving SNGFR. (Hypertension. 1999;34:273-278.)

Key Words: L-type receptor calcium antagonist ■ mibefradil ■ amlodipine ■ systemic hemodynamics ■ L-NAME ■ glomerular arteriolar injury ■ proteinuria ■ T-type receptor calcium antagonist ■ renal hemodynamics ■ glomerular dynamics ■ arteriolar injury

The calcium antagonists are widely used for the treatment of hypertension and coronary artery disease.¹⁻³ They have been said to belong to 3 chemical subgroups (dihydropyridines, phenylalkylamines, and benzothiazepines), although at least 5 types of receptors (L, T, N, P/Q, and R), have been identified and cloned that control their respective voltage-gated calcium channels. Moreover, their intracellular effects on controlling free calcium ions are also extremely variable. This class of compounds is highly heterogeneous, and each agent may be distinguished by location and function, structure, pharmacological sensitivities, and electrophysiological and physiological characteristics.¹ Indeed, in no one organ is their variability more evident than with respect to their renal effects.⁴⁻¹⁴

Recently, the T-type calcium antagonist mibefradil was demonstrated to have developed an ability to inhibit both L- (long lasting) and T- (transient) types of calcium channel receptor sites, with greater selectivity for the T-type channel receptor.¹⁵⁻¹⁸

Little is known on the comparative or differential effects of these 2 receptor antagonists on systemic, renal, and glomerular hemodynamic effects.¹⁹,²⁰ The present study, therefore, was designed to determine whether differences exist between these 2 types of calcium antagonists. To this end, the effects of the L-type calcium antagonist amlodipine and the T-type calcium antagonist mibefradil on systemic and renal hemodynamics and glomerular dynamics were studied in spontaneously hypertensive rats (SHR) with N⁵-nitro-L-arginine methyl ester (L-NAME)–exacerbated nephrosclerosis.²¹

Methods

Male SHR (Charles River Laboratories, Wilmington, Mass) aged 17 weeks were housed in plastic cages and maintained at 20°C in a light-controlled room with free access to food, standard rat chow (PMI Feeds Inc, St. Louis, Mo), and tap water. All experimental
studies had been approved previously by our institutional (Alton Ochsner Medical Foundation) animal care and use committee. The rats were divided into 7 groups: Group 1, control (n = 10); Group 2, mibefradil (50 mg·kg⁻¹·d⁻¹) by gastric gavage for 3 weeks; n = 9); Group 3, L-NNAME (Sigma Chemical Co, St. Louis, Mo); (50 mg/L in drinking water for 3 weeks; n = 13); Group 4, L-NNAME (50 mg/L) plus mibefradil (50 mg·kg⁻¹·d⁻¹) for three weeks by gastric gavage; n = 10); Group 5, L-NNAME (50 mg/L) plus mibefradil (10 mg·kg⁻¹·d⁻¹) for 3 weeks by gastric gavage; n = 7); Group 6, L-NNAME (50 mg/L) for 3 weeks; and mibefradil (50 mg·kg⁻¹·d⁻¹) for 3 weeks; and Group 7 (n = 8) L-NNAME (50 mg/L) for 3 weeks then mibefradil (10 mg·kg⁻¹·d⁻¹) for 3 weeks. The average daily dose of L-NNAME (7.6±0.7 mg/d in drinking water) was calculated from water intake and determined in our earlier studies.11,21–23 The 24-hour urinary protein (U prot V, Lowry method) and sodium (U Na V, saline at room temperature. Cardiac output was displayed on a digital recorder (Cardiotherm 500; Columbus Instruments; Columbus, Ohio) for 10 min. The arterial pressure was measured through a transducer (model 2263, Grass Instruments; Quincy, Mass) that was connected to a thermodilution device (Model 200; Medex Instruments; West Point, Pa) at a rate of 0.4 mL/100 g body weight per hour. 11

Micropuncture Technique

Rats were anesthetized with thiobutabarbital (Inactin, 100 mg/kg; Byk-Gulden) and then placed on a temperature-regulated table to maintain rectal temperature at 37°C throughout the study. Inactin was selected for the anesthetic agent because, in our past studies, it as well as other notable agents9,25–26 this agent had no effect on renal function. However, micropuncture studies are extensive and long, and we cannot exclude this factor in altering such renal phenomena such as glomerulotubular feedback. After a tracheostomy (with insertion of polyethylene tubing), an indwelling polyethylene catheter (PE-50) was placed into the right femoral artery to permit blood sampling and measurement of artery pressure and heart rate. Arterial pressure was measured through a transducer (model P23 Dd, Statham Instruments; Oxnard, Calif) that was connected to a multichannel polygraph (Sensor Medics R612, Beckman Instruments; Schiller Park, Ill). The right carotid artery was cannulated with a thermistor microprobe (Type IT-18; Physitemp Instruments, Inc; Clifton, NJ) that was connected to a thermodilution device (Cardiotherm 500; Columbus Instruments; Columbus, Ohio) for determination of cardiac output. The right jugular vein was also cannulated with a polyethylene catheter (PE-50) for infusion of solutions. A high-precision syringe (CR-700-200; Hamilton Co; Reno, Nev) was connected to that venous catheter for injection of saline at room temperature. Cardiac output was displayed on a digital screen, which was recorded simultaneously; the calculated cardiac output was normalized for body weight and expressed as cardiac index (CI, mL·min⁻¹·kg⁻¹). Total peripheral resistance (TPR) was calculated as the quotient of mean arterial pressure (MAP) and CI. After these hemodynamic measurements were obtained, the bladder was cannulated with a polyethylene catheter (PE-100) for collection of urine. The left kidney was then exposed through a flank incision and suspended in a Lucite cup (packed with cotton) while warm agar was dripped around the kidney to form a saline (0.9% NaCl) well at the midpoint of each collection period. Two or three “star vessels” were punctured directly for collection of efferent glomerular arteriolar blood. To determine single-nephron glomerular filtration rate (SNGFR), precisely timed (90-second) samples of fluid were collected from 4 to 6 superficial proximal tubules. Efferent (P G), tubular (P T), and stopped-flow (SFP) pressures were measured directly by a servo-null system (Instrumentation for Physiology and Medicine; San Diego, Calif) as reported previously.11,21–23 The P G and P T were obtained from the proximal tubule and the “star vessel”, respectively. P T was calculated from the sum of the P SFP and the plasma π T. Arterial plasma protein concentration (C A) was determined, and π T and π G were calculated using the Landis-Pappenheimer equation.21 The pressure gradient across the glomerular capillary wall was calculated as ΔP = P T – P G; the transmembrane colloid osmotic pressure difference (Δπ) was calculated according to the equation of Deen et al23 as modified by Arendshorst and Gottschalk.22 The P SFP, P T, and P G measurements were made 3 times and their averages were determined. [3H]Inulin radioactivity of all tubular fluid, urine, and plasma samples were counted to determine SNGFR, GFR, and ERPF. These measurements were used to calculate π T, π G, R A, and R G, and glomerular capillary K F. At the termination of each study, blood was withdrawn for measurement of serum creatinine and uric acid concentrations using a 747-100 Analyzer (Boehringer Mannheim/Hitachi).

Renal Morphology

After being fixed in 10% buffered formalin and embedded in paraffin for light microscopy, the kidneys were cut at a thickness of 2 to 3 μm and stained with hematoxylin and eosin, periodic acid-Schiff, and periodic acid-methenamine-silver as also reported previously.11,21–23 Histological examination was conducted in a blinded fashion, and glomerular and arteriolar injury scores were calculated as described previously.11,21–23 Approximately 30 subcapsular and 50 juxtamedullary glomeruli from each specimen were analyzed for glomerular injury as described in previous studies: Grade 1, normal glomerulus by light microscopy; Grade 2, involvement of up to one-third of the glomerular area; Grade 3, involvement of one to two thirds of the glomerulus; and Grade 4, two thirds to global sclerosis. Each scoring permitted calculation of a glomerular injury score (GIS): [(1× number of Grade 2 glomeruli)+ (2× number of Grade 3 glomeruli) + (3× number of Grade 4 glomeruli)]×100/(number of glomeruli studied).

Fifty to 50 afferent arterioles were also examined from each specimen in order to determine an arteriolar injury score (AIS) using the serial sections stained with periodic-acid-Schiff. Grading was performed as described previously:11,21–23 Grade 1, no arteriolar changes; Grade 2, arteriolar wall hyalinosis up to 50% of circumference; Grade 3, 50% to 100% hyalinosis of the wall circumference but without luminal narrowing; and Grade 4, complete hyalinosis of the wall with evidence of luminal encroachment. Each score was then calculated according to the formula for arteriolar injury score: [(1× number of Grade 2 arterioles)+(2× number of Grade 3 arterioles) + (3× number of Grade 4 arterioles)]×100/(number of arterioles observed).

Statistical Analysis

All data are expressed as mean±SE. A 1-way ANOVA, followed by Duncan’s multiple range test, was performed for between-group significance.30 The confidence level was considered to be statistically significant when the probability value was less than 0.05.

Results

Organ Weights

Left ventricular and aortic masses were significantly increased by L-NNAME and reduced by mibefradil, with respect to the control group (P<0.001). Moreover, left ventricular and aortic masses were also reduced by both calcium antagonists (with and after L-NNAME). In contrast, right ventricular and left kidney masses did not change (Table 1). Body weight was greater in Group 6 rats because they were 3 weeks older.

Systemic and Cardiac, and Whole Kidney Hemodynamics

Mibefradil (Group 2) slightly decreased MAP and TPR1 as compared with the control group (Table 2). On the other
TABLE 1. Body and Organ Weights

<table>
<thead>
<tr>
<th></th>
<th>Group 1, Control (n=10)</th>
<th>Group 2, Mibefradil (n=9)</th>
<th>Group 3, L-NAME (n=13)</th>
<th>Group 4, L-NAME+Mibefradil (n=10)</th>
<th>Group 5, L-NAME+Amlodipine (n=7)</th>
<th>Group 6, L-NAME then Mibefradil (n=9)</th>
<th>Group 7, L-NAME then Amlodipine (n=8)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body weight, g</td>
<td>354.0±1.0</td>
<td>354.0±1.0</td>
<td>354.0±1.0</td>
<td>348.0±1.0</td>
<td>368.0±1.0</td>
<td>354.0±1.0</td>
<td>368.0±1.0</td>
</tr>
<tr>
<td>Left ventricular, mg/g</td>
<td>2.7±0.0</td>
<td>2.7±0.0</td>
<td>2.7±0.0</td>
<td>2.7±0.0</td>
<td>2.7±0.0</td>
<td>2.7±0.0</td>
<td>2.7±0.0</td>
</tr>
<tr>
<td>Right ventricular, mg/g</td>
<td>0.5±0.0</td>
<td>0.5±0.0</td>
<td>0.5±0.0</td>
<td>0.5±0.0</td>
<td>0.5±0.0</td>
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<td>0.5±0.0</td>
</tr>
<tr>
<td>Aorta, mg · mm⁻¹ · kg⁻¹</td>
<td>3.1±0.1</td>
<td>3.1±0.1</td>
<td>3.1±0.1</td>
<td>3.1±0.1</td>
<td>3.1±0.1</td>
<td>2.9±0.1</td>
<td>3.0±0.1</td>
</tr>
</tbody>
</table>

Data is mean±1 SEM.

*P<0.01, **P<0.05 vs Group 1; †P<0.01, ‡P<0.05 vs Group 3; §P<0.01 vs Group 6.

hand, ERPF and GFR increased significantly (P<0.01, P<0.05, respectively). L-NAME treatment (Group 3) significantly increased MAP, TPRI, and renal vascular resistance (RVR; P<0.01) and decreased ERPF and GFR (P<0.05 at least). Significantly, treatment with mibefradil and L-NAME (Group 4) prevented L-NAME-induced alterations in MAP, GFR (P<0.05), and TPRI, RVR (P<0.01), although ERPF remained unaltered. Amlodipine (with and after L-NAME) also significantly prevented and reversed the adverse hemodynamic alterations on MAP, TPRI, GFR, and RVR (as compared with L-NAME treatment; Group 3). Moreover, when either mibefradil or amlodipine followed the 3-week administration of L-NAME (Groups 6 and 7), the increases in MAP, TPRI, and RVR and decreases in GFR were reversed significantly as compared with L-NAME treatment (Group 3; Table 2). There were no significant differences between Groups 6 and 7.

Glomerular Dynamics

SNGFR and SNPF were increased (P<0.05 at least) and R₅ and R₆ decreased (P<0.01) by mibefradil (Group 2). L-NAME (Group 3) decreased SNGFR, SNPF, and Kₑ, whereas SFP, ∆P, Pₒ, R₅, and R₆ were increased significantly (Table 3). L-NAME co-treatment with mibefradil (Group 4) or amlodipine (Group 5) reduced SFP, ∆P, Pₒ, Rₒ, and Kₑ significantly (Table 3); however, the preventive effects of mibefradil were greater than amlodipine. Moreover, when mibefradil or amlodipine were administered after the 3-week course treatment with L-NAME (Groups 6 and 7), SNGFR, SNPF, ∆P, Pₒ, R₅, R₆, and Kₑ were reversibly and beneficially changed (as compared with the L-NAME treatment Group 3; at least P<0.05). These reversal effects in SNGFR, SNPF, R₅, and Kₑ seemed to be greater with mibefradil than with amlodipine, but they were not significant statistically. Although the concentrations of serum creatinine and uric acid were increased by L-NAME (Group 3), they were not increased significantly so as compared with the control group (Group 1). U₉, V were significantly increased by L-NAME; these changes were prevented and reversed by mibefradil or amlodipine (Table 4). These effects seemed to be greater with mibefradil rather than amlodipine.

Glomerular and Arteriolar Injury Scores

Histological study demonstrated that L-NAME (Group 3) exacerbated both the GIS (29±6 versus 126±28; P<0.01) and AIS (43±2 versus 104±24; P<0.01) as compared with controls (Group 1). The GIS of both subcapsular (14±2 versus 62±14; P<0.01) and juxtaglomerular (14±4 versus 28±2 versus 104±24; P<0.01) as compared with controls (Group 1). The GIS of both subcapsular (14±2 versus 62±14; P<0.01) and juxtaglomerular (14±4 versus

TABLE 2. Systemic and Whole Kidney Hemodynamics

<table>
<thead>
<tr>
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<th>Group 1, Control (n=10)</th>
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<th>Group 7, L-NAME then Amlodipine (n=8)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean arterial pressure, mm Hg</td>
<td>184±2</td>
<td>174±4</td>
<td>213±4</td>
<td>188±5</td>
<td>187±5</td>
<td>181±4</td>
<td>182±3</td>
</tr>
<tr>
<td>Heart rate, bpm</td>
<td>335±9</td>
<td>335±10</td>
<td>346±8</td>
<td>344±11</td>
<td>316±3</td>
<td>339±9</td>
<td>321±5</td>
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<td>Cardiac index, mL · min⁻¹ · kg⁻¹</td>
<td>192±6</td>
<td>208±6</td>
<td>176±9</td>
<td>204±12</td>
<td>184±6</td>
<td>200±15</td>
<td>196±8</td>
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<tr>
<td>Total peripheral resistance index, U/kg</td>
<td>0.96±0.03</td>
<td>0.84±0.03</td>
<td>1.28±0.09</td>
<td>0.95±0.06</td>
<td>1.03±0.06</td>
<td>0.96±0.09</td>
<td>0.94±0.04</td>
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<td>Stroke Index, mL · beat⁻¹ · kg⁻¹</td>
<td>0.58±0.03</td>
<td>0.63±0.03</td>
<td>0.52±0.03</td>
<td>0.55±0.06</td>
<td>0.58±0.02</td>
<td>0.58±0.05</td>
<td>0.61±0.02</td>
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<tr>
<td>Effective renal plasma flow, mL/min</td>
<td>2.9±0.1</td>
<td>3.9±0.2</td>
<td>1.7±0.3</td>
<td>2.2±0.2</td>
<td>2.4±0.3</td>
<td>2.3±0.4</td>
<td>2.4±0.3</td>
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<tr>
<td>Glomerular filtration rate, mL · min⁻¹ · 100 g⁻¹</td>
<td>0.26±0.02</td>
<td>1.35±0.01</td>
<td>0.16±0.02</td>
<td>0.25±0.02</td>
<td>0.23±0.03</td>
<td>0.22±0.03</td>
<td>0.25±0.03</td>
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<tr>
<td>Filtration fraction, %</td>
<td>32±2</td>
<td>32±2</td>
<td>43±9</td>
<td>41±3</td>
<td>32±3</td>
<td>34±5</td>
<td>37±3</td>
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<tr>
<td>Renal vascular resistance, U</td>
<td>30±2</td>
<td>22±1</td>
<td>103±25</td>
<td>48±7</td>
<td>43±7</td>
<td>39±4</td>
<td>41±5</td>
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<tr>
<td>Hematocrit, %</td>
<td>53±0.4</td>
<td>52±1.0</td>
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<td>51±0.7</td>
<td>51±0.6</td>
<td>51±1</td>
<td>52±0.3</td>
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</table>

Data is mean±1 SEM.

*P<0.01, **P<0.05 vs Group 1; †P<0.01, ‡P<0.05 vs Group 3.
TABLE 3. Glomerular Dynamics

<table>
<thead>
<tr>
<th></th>
<th>Group 1, Control (n=10)</th>
<th>Group 2, Mibefradil (n=9)</th>
<th>Group 3, L-NAME+Mibefradil (n=13)</th>
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<th>Group 7, L-NAME then Amlopidine (n=8)</th>
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</thead>
<tbody>
<tr>
<td>SNGFR, nL/min</td>
<td>31.2±1.1</td>
<td>36.9±0.8*</td>
<td>23.7±1.9*</td>
<td>33.8±0.6†</td>
<td>28.0±1.7‡§</td>
<td>31.5±1.5†</td>
<td>28.3±1.3‡</td>
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<tr>
<td>SNPF, nL/min</td>
<td>105±4</td>
<td>134±7**</td>
<td>79±8*</td>
<td>121±10†</td>
<td>91±8</td>
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<td>104±10†</td>
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<tr>
<td>SNFF, %</td>
<td>30±0.4</td>
<td>28±1.4</td>
<td>31±1.4</td>
<td>29±2.0</td>
<td>32±1.3</td>
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<td>31±1.4</td>
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<tr>
<td>P1, mm Hg</td>
<td>12.1±0.5</td>
<td>12.1±0.3</td>
<td>11.3±0.4</td>
<td>12.5±0.2</td>
<td>12.4±0.4</td>
<td>13.1±0.4‡</td>
<td>13.1±0.5‡</td>
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<tr>
<td>P2, mm Hg</td>
<td>17.2±0.8</td>
<td>17.5±0.5</td>
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<td>16.5±0.4</td>
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<td>18.6±0.4</td>
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<tr>
<td>SFP, mm Hg</td>
<td>33±0.7</td>
<td>32±0.6</td>
<td>38±0.9*</td>
<td>32±0.7†</td>
<td>30±0.9†</td>
<td>33±0.6‡</td>
<td>33±0.4†</td>
</tr>
<tr>
<td>s, mm Hg</td>
<td>19±0.3</td>
<td>21±0.5</td>
<td>18±0.5</td>
<td>22±1.2†</td>
<td>18±0.4§</td>
<td>20±0.8</td>
<td>19±0.4</td>
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<tr>
<td>o, mm Hg</td>
<td>33±1.1</td>
<td>34±1.2</td>
<td>33±1.6</td>
<td>39±2.7**†</td>
<td>33±1.0</td>
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<td>36±1.4</td>
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<td>ΔP, mm Hg</td>
<td>41±0.9</td>
<td>38±1.4</td>
<td>45±1.0*</td>
<td>41±1.3‡</td>
<td>36.1±1.0††§</td>
<td>39±0.8†</td>
<td>38±0.9†</td>
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<tr>
<td>P0</td>
<td>52.8±0.9</td>
<td>52.0±0.7</td>
<td>56.4±1.2**</td>
<td>53.7±1.4</td>
<td>48.1±1.2**†§</td>
<td>52.5±0.8‡</td>
<td>51.6±0.7‡</td>
</tr>
<tr>
<td>R0, u</td>
<td>4.6±0.2</td>
<td>3.7±0.1*</td>
<td>9.3±1.2*</td>
<td>4.7±0.4†</td>
<td>6.3±0.7†</td>
<td>4.9±0.6‡</td>
<td>5.6±0.4†</td>
</tr>
<tr>
<td>R0, u</td>
<td>1.5±0.1</td>
<td>1.2±0.1*</td>
<td>2.7±0.3*</td>
<td>1.6±0.2†</td>
<td>1.7±0.1†</td>
<td>1.8±0.2†</td>
<td>1.7±0.1†</td>
</tr>
<tr>
<td>Kf,nL·s⁻¹·mm Hg⁻¹</td>
<td>0.040±0.004</td>
<td>0.060±0.005*</td>
<td>0.021±0.002*</td>
<td>0.051±0.005†</td>
<td>0.046±0.003†</td>
<td>0.047±0.003†</td>
<td>0.041±0.004†</td>
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</tbody>
</table>

Data is mean±1 SEM.
*P<0.01, **P<0.05 vs Group 1; †P<0.01, ‡P<0.05 vs Group 3, §P<0.01, ||P<0.05 vs Group 4.

64±15 P<0.01) cortical glomeruli were more severe in the L-NAME-treated SHR than in controls. Both calcium antagonists significantly improved the subcapsular and juxtaglomerular cortical glomeruli GIS (P<0.01). Furthermore, the AIS was also reduced significantly by either mibefradil or amlopidine (Table 5).

**Discussion**

The results of this study clearly demonstrate that both mibefradil and amlopidine improved systemic and renal hemodynamics as well as intrarenal glomerular dynamics. These findings are similar to our previous findings concerning the renoprotective effects of felodipine, an L-type channel calcium receptor antagonist. In that study, felodipine not only prevented but also reversed L-NAME-exacerbated hypertensive nephrosclerosis in SHR. On the other hand, prior reports describing the effects by amlopidine, another L-type channel antagonist, have been inconsistent. Dworkin et al found no renoprotective effects of amlopidine in uninephrectomized SHR since there was no improvement in proteinuria, FF, and GIS. On the other hand, Saruta et al demonstrated a protective action in 5/6 nephrectomized SHR. These workers also reported that most L-type calcium antagonists, including amlopidine, diluted only the afferent glomerular arterioles.

In general, most reports on the effects of calcium antagonists, such as an L-type agent, suggest that these agents dilate only the afferent arterioles and have little effect on the efferent vessels (eg, nifedipine in the isolated perfused hydromeophic model, diltiazem in vasoconstricted isolated perfused kidney, and verapamil in rats in which responses to angiotensin II were determined from perfused juxtaglomular nephron).

In contrast, L-type receptor antagonists, such as efonidipine and mibefradil, have been shown to dilate both afferent and efferent glomerular arterioles. Similarly, Ménard et al reported that mibefradil diminished proteinuria and prevented glomerular lesions in DOCA-salt hypertensive rats. These investigators also suggested that mibefradil may dilate both afferent arterioles and efferent arterioles. Our data support this finding since mibefradil reduced P0, R0, and R2. Our data further suggest that amlopidine also dilated both afferent and efferent arterioles, findings which are consistent with the report by Hayashi et al.

**TABLE 4. Serum Creatinine and Uric Acid Concentrations**

<table>
<thead>
<tr>
<th></th>
<th>Group 1, Control (n=10)</th>
<th>Group 2, Mibefradil (n=9)</th>
<th>Group 3, L-NAME (n=13)</th>
<th>Group 4, L-NAME+Mibefradil (n=10)</th>
<th>Group 5, L-NAME+Amlopidine (n=7)</th>
<th>Group 6, L-NAME then Mibefradil (n=9)</th>
<th>Group 7, L-NAME then Amlopidine (n=8)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Creatinine, mg/dL</td>
<td>0.55±0.05</td>
<td>0.60±0.05</td>
<td>0.74±0.05</td>
<td>0.74±0.06</td>
<td>0.71±0.07</td>
<td>0.71±0.06</td>
<td>0.68±0.08</td>
</tr>
<tr>
<td>Uric acid, mg/dL</td>
<td>1.6±0.2</td>
<td>1.2±0.2</td>
<td>2.1±0.4</td>
<td>1.6±0.2</td>
<td>2.1±0.3</td>
<td>1.5±0.2</td>
<td>1.3±0.1</td>
</tr>
<tr>
<td>Uo, V, mg/24 h</td>
<td>15±1.1</td>
<td>14±1.1</td>
<td>55±10*</td>
<td>13±1.9†</td>
<td>21±1.6†</td>
<td>17±1.6†</td>
<td>19±1.9†</td>
</tr>
<tr>
<td>Ua, V, mg/24 h</td>
<td>1.4±0.1</td>
<td>1.3±0.1</td>
<td>1.5±0.1</td>
<td>1.6±0.1</td>
<td>1.8±0.1*</td>
<td>1.5±0.1</td>
<td>1.7±0.1</td>
</tr>
</tbody>
</table>

Data is mean±1 SEM.
*P<0.01, **P<0.05 vs Group 1; †P<0.01 vs Group 3.
Our study does not define the mechanism by which either mibebradil or amlodipine dilate afferent and efferent arterioles although these 2 agents clearly reduce the arteriolar smooth muscle tone. Some studies have suggested that angiotensin II-induced afferent arteriolar vasoconstriction may be mediated by activation of the voltaged calcium channels.25,34,35 Both voltage-operated T- and L-type calcium channels prevail in afferent arterioles, although similar actions may be lacking in the efferent arterioles. Saruta et al,5 however, demonstrated that efonidipine and manidipine not only inhibited voltage-operated calcium channels but also seem to affect other mechanisms involved in arteriolar smooth muscle contraction, because these agents inhibited angiotensin II-induced vasoconstriction of efferent arterioles.

In a recent report,13 we have summarized the results of those studies that examined the nephroprotective effects of calcium antagonists in hypertension. In that review we emphasized that the nephroprotective effect of all calcium antagonists were consistent and appeared to vary with the experimental model, the type and dose of the calcium antagonist used, and their possible differences in their renal microcirculatory effects. Bidani and Griffin36 suggested that glomeruloprotection with calcium antagonists may depend on the net balance between the protective arterial pressure lowering effects and the deleterious pressure transmission effects on renal vasculature, as they generally decreased afferent arterioles resistance. However, our data clearly demonstrated that the level of arterial pressure was not important in the L-NAME exacerbated SHR nephrosclerosis model, since mean arterial pressure was still remained intensely increased despite its significant reduction in response to the 2 different calcium antagonists. Moreover, both agents prevented and reversed the pathophysiologival renal effects although quantitative differences may exist.

**Conclusion**

These data, therefore, suggest that despite their action through 2 calcium channel receptors and similar systemic hemodynamic effects, the T- and L-type calcium antagonists each prevented and reversed L-NAME exacerbated hypertensive nephrosclerosis in SHR. Although both T- and L-type agents studied herein dilated the afferent and efferent arterioles, the T-channel antagonist mibebradil seemed to have a greater effect than the L-type antagonist amlodipine in reducing afferent arteriolar resistance and on maintaining and preserving SNGFR. Mibebradil and amlodipine each reduced left ventricular and aortic mass without changing right ventricular mass.

**References**


Differential Effects of T- and L-Type Calcium Antagonists on Glomerular Dynamics in Spontaneously Hypertensive Rats
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