Sex Hormone Modulation of Ventricular Hypertrophy in Sinoaortic Denervated Rats

ANTONIO M. CABRAL, ELISARDO C. VASQUEZ, MARGARETH R. MOYSÉS, AND ABILIO ANTONIO

SUMMARY The influence of sex hormones on the development of left ventricular hypertrophy was investigated in baroreceptor-denervated rats. A significant increase \((p<0.01)\) in the left ventricular weight/body weight ratio was observed in male but not in female rats 15 days after operation, compared to age- and sex-matched sham-operated rats. This differential hypertrophy occurred despite the development of a significant elevation in arterial blood pressure in both sexes. Castration prior to sinoaortic denervation did not change the level of arterial hypertension but caused a significant reduction \((p<0.01)\) in left ventricular weight in male rats and a significant increase \((p<0.01)\) in female rats. The pretreatment of male and female sinoaortic denervated and castrated rats with testosterone resulted in ventricular hypertrophy similar to that observed in intact male sinoaortic denervated rats. Pretreatment with estradiol, however, suppressed the left ventricular hypertrophy in intact male rats but did not change the normal ventricular mass observed in intact female sinoaortic denervated rats. These results indicate that the development of left ventricular hypertrophy in sinoaortic denervated rats is modulated by sex hormones, and that testosterone exerts a facilitatory and estradiol an inhibitory action. (Hypertension 11 [Suppl I]: I-93–I-97, 1988)

KEY WORDS • baroreceptors • neurogenic hypertension • heart rate • arterial pressure

LEFT ventricular hypertrophy is observed in many different models of experimental hypertension and in hypertensive humans. However, it is generally recognized that the condition does not depend solely on variations in the level of blood pressure. It has been observed in spontaneously hypertensive rats during the prehypertensive stage, and after the development of hypertension has been prevented by peripheral sympathectomy and by antihypertensive drugs. Lindpaintner and Sen demonstrated the reversal of cardiac hypertrophy by dietary sodium restriction in renovascular hypertensive rats, despite persisting hypertension.

Recently, we observed left ventricular hypertrophy in male, but not in female, sinoaortic denervated (SAD) rats, when compared to age- and sex-matched sham-operated rats. However, the influence of sex on the cardiac hypertrophy of SAD rats has not been investigated. The aim of the present investigation was to verify the modulatory actions of sex hormones testosterone and estradiol on the development of ventricular hypertrophy in SAD rats.

Materials and Methods

Experiments were carried out on 42 male and 42 female rats (Wistar strain) weighing 200 to 260 g. The rats were obtained from the Physiological Sciences Department. They were housed in standard size metabolic cages in an air-conditioned, light-cycled room. Rats were randomly divided into groups containing eight to nine animals each. Sinoaortic denervation was performed under ether anesthesia in a group of male and female rats as described in detail elsewhere. In brief, aortic baroreceptors were denervated by bilaterally resecting a 0.5-mm strip of superior laryngeal, cervical sympathetic, and aortic depressor nerves. The carotid sinus baroreceptors were denervated by stripping the carotid bifurcation and its branches. The same procedure was used on the male and female sham-operated (SO) rats, but the nerves were not dissected out. As with the SAD rats, surgery lasted 15 to 30 minutes. To study the effects of endogenous sex hormones, a group of female and male rats were castrated under ether anesthesia, and 15 days later underwent sinoaortic denervation.

To investigate the effects of the administration of exogenous sex hormones, oophorectomized female...
and orchiectomized male rats were given testosterone propionate (Schering 0.5 mg/kg) or 17β-estradiol enolate (Schering 0.5 μg/kg) subcutaneously. The hormones were diluted in 0.1 ml corn oil and given daily during the 15 days before and after sinoaortic denervation. Control animals received either injection of corn oil or no injection.

After surgical denervation of the baroreceptors, animals have a transient reduction in both food and water intake. Therefore, the daily intake of all groups was monitored after surgery. Male and female SO rats received the same daily quantities of water and food that the SAD rats had ingested on the previous day.

Blood pressure measurements were recorded directly through an indwelling catheter. A polyethylene catheter (PE-50 with tapered end) filled with heparinized saline, 40 U/ml, was placed in the abdominal aorta through the femoral artery under ether anesthesia. A second catheter (PE-50) was placed in the right jugular vein to permit intravenous injection of drugs. The catheters were tunneled subcutaneously and brought to the exterior at the dorsal midcervical region of the neck. Mean arterial pressure (MAP) and heart rate (HR) were recorded in the conscious, unrestrained animals 6 hours later. The arterial catheter was connected to a Statham P23Dd pressure transducer (Statham Instruments, Oxnard, CA, USA) by flexible connecting tubing, and the MAP was recorded continuously on a polygraph (Hewlett-Packard, Lexington, MA, USA). The HR was computed from the arterial pressure pulse waves.

The baroreceptor reflex activity in SAD rats was tested by determining the decrease in HR in response to an increase in MAP caused by bolus injection of methoxamine (Wellcome, 10-40 μg/kg i.v.).

Immediately after MAP and HR measurements and baroreceptor function tests, animals were killed by decapitation, and the right and left cardiac ventricles (including the septum) were separated and weighed. The net weight of seminal vesicle and prostate in male rats and the uterus in female rats was also measured.

All data are reported as means ± SEM. Statistical analysis was performed using analysis of variance and Tukey tests, and Student’s t test when appropriate.

**Results**

Sinoaortic denervation resulted in a significant increase of the left ventricular weight/body weight ratio in male (2.323 ± 0.031 mg/g) but not female (2.065 ± 0.057 mg/g) rats compared to age- and sex-matched SO rats (2.031 ± 0.093 and 2.171 ± 0.042 mg/g, respectively) as shown in Figure 1. The values observed in right ventricles were not significantly different between SAD (males, 0.551 ± 0.016; females, 0.497 ± 0.014 mg/g) and SO (males, 0.555 ± 0.018; females, 0.554 ± 0.012 mg/g) rats. The MAP observed in conscious, unrestrained animals was significantly higher in male (121 ± 5 mm Hg) and female (127 ± 2 mm Hg) SAD rats than in both male (108 ± 3 mm Hg) and female (114 ± 3 mm Hg) SO rats. As previously observed, the HR of hypertensive male SAD rats (410 ± 19 beats/min) was not significantly different from that of male SO rats (370 ± 8 beats/min); the difference between female hypertensive SAD (424 ± 10 beats/min) and female SO (384 ± 9 beats/min) groups, however, was significant (Tables 1 and 2). The test of baroreceptor reflex activity in male and female SAD groups showed no significant bradycardia in response to increases in MAP caused by methoxamine, 10 to 40 μg/kg, contrasting with the reflex bradycardia observed in the SO group (Table 3).

The castrated male SAD rats showed a significant reduction (6.5%) of left ventricular hypertrophy (see Figure 1) without significant changes in MAP and HR (see Table 1). On the other hand, castration of female SAD rats significantly increased the left ventricular weight/body weight ratio (7%), resulting in the same levels of ventricular mass for male and female castrated SAD groups. The values of arterial hypertension and HR in female castrated SAD rats were similar to those in male castrated SAD rats.

Treatment with testosterone in male and estradiol in female castrated SAD rats restored the values of left ventricular weight/body weight ratio (2.374 ± 0.045 and 2.098 ± 0.043 mg/g, respectively) observed in noncastrated SAD rats (see Figure 1). When the male rats were treated with estradiol, left ventricular hypertrophy was completely suppressed (1.997 ± 0.022 mg/g) despite the same level of arterial hypertension (see Table 1).

Treatment with estradiol in female castrated SAD rats resulted in a return of left ventricular weight to levels observed in SO and SAD groups. When this group was treated with testosterone, however, left ven-
tricular hypertrophy developed (2.322 ± 0.038 mg/g) to the same levels observed in male SAD rats (see Figure 1). Neither testosterone nor estradiol treatment changed the level of arterial hypertension in male and female castrated SAD rats as compared to noncastrated SAD animals (see Tables 1 and 2).

Discussion

Sinoaortic denervation in the rat results in immediate arterial hypertension and tachycardia, followed by progressive reduction in the level of hypertension and a return to normal HR. We have also observed a rapid development of desensitization of cardiac β-adrenergic receptors and ventricular hypertrophy. The present results demonstrate that the development of left ventricular hypertrophy in SAD rats is modulated by sex hormones, where testosterone exerts a facilitatory and estradiol an inhibitory action.

As previously reported, the present results show left ventricular hypertrophy in male but not in female rats 15 days after sinoaortic denervation. This difference between sexes could not be attributed to baroreceptor denervation because the test of baroreceptor reflex activity shows, both in male and female SAD rats, no bradycardia in response to increases in MAP elicited by methoxamine, compared to age- and sex-matched SO groups (see Figure 2). Vasquez and Krieger reported a marked decrease in food and water consumption after sinoaortic denervation. However, the paired feeling and water intake of age- and sex-matched SO and SAD animals resulted in a similar body weight gain both in male and female SAD rats. The present study, showing similar levels of arterial hypertension in male (hypertrophied heart) and female (nonhypertrophied heart) SAD rats, supports the concept that factors other than arterial pressure may contribute to ventricular hypertrophy. Dissociation of arterial pressure and cardiac hypertrophy has also been demonstrated in various animal models.

Considering that sinoaortic baroreceptor denervation results in cardiac sympathetic hyperactivity and that cardiac hypertrophy may directly or indirectly be influenced by adrenergic activity, and also considering that the sex hormones exert a modulatory action on the sympathetic nervous system, it was expected that differences in the ventricular mass would occur between male and female hypertensive rats.

The orchiectomy of male and oophorectomy of female SAD rats developed (2.322 ± 0.038 mg/g) to the same levels observed in male SAD rats (see Figure 1). Neither testosterone nor estradiol treatment changed the level of arterial hypertension in male and female castrated SAD rats as compared to noncastrated SAD animals (see Tables 1 and 2).

Table 1. Influence of Sex, Castration, and Testosterone (0.5 mg/kg) or Estradiol (0.5 μg/kg) Administration in Male Sinoaortic Denervated Rats

<table>
<thead>
<tr>
<th>Measurement</th>
<th>Male sham-operated (n = 9)</th>
<th>Non-castrated (n = 9)</th>
<th>Castrated + testosterone (n = 9)</th>
<th>Castrated + estradiol (n = 9)</th>
</tr>
</thead>
<tbody>
<tr>
<td>MAP (mm Hg)</td>
<td>108 ± 3</td>
<td>121 ± 5*</td>
<td>123 ± 3*</td>
<td>125 ± 3*</td>
</tr>
<tr>
<td>HR (beats/min)</td>
<td>370 ± 8</td>
<td>410 ± 19</td>
<td>384 ± 8</td>
<td>377 ± 9</td>
</tr>
<tr>
<td>Body wt (g)</td>
<td>245 ± 7</td>
<td>231 ± 7</td>
<td>244 ± 12</td>
<td>250 ± 10</td>
</tr>
<tr>
<td>Left ventricular wt (mg)</td>
<td>498 ± 9</td>
<td>537 ± 10†</td>
<td>523 ± 17</td>
<td>594 ± 15‡</td>
</tr>
<tr>
<td>Right ventricular wt (mg)</td>
<td>136 ± 5</td>
<td>128 ± 5</td>
<td>131 ± 9</td>
<td>136 ± 7</td>
</tr>
<tr>
<td>Seminal vesicle wt (mg)</td>
<td>288 ± 21</td>
<td>259 ± 23</td>
<td>52 ± 5</td>
<td>489 ± 31§</td>
</tr>
<tr>
<td>Prostate wt (mg)</td>
<td>235 ± 19</td>
<td>219 ± 23</td>
<td>8 ± 2</td>
<td>328 ± 24§</td>
</tr>
</tbody>
</table>

Data are means ± SEM. HR = heart rate.

Analysis of variance: *p<0.01 or †p<0.05 compared with sham-operated rats; ‡p<0.01 compared with noncastrated sinoaortic denervated rats; §p<0.01 compared with castrated sinoaortic denervated rats.

Table 2. Influence of Sex, Castration, and Testosterone (0.5 mg/kg) or Estradiol (0.5 μg/kg) Administration in Female Sinoaortic Denervated Rats

<table>
<thead>
<tr>
<th>Measurement</th>
<th>Female sham-operated (n = 9)</th>
<th>Non-castrated (n = 9)</th>
<th>Castrated + testosterone (n = 9)</th>
<th>Castrated + estradiol (n = 9)</th>
</tr>
</thead>
<tbody>
<tr>
<td>MAP (mm Hg)</td>
<td>114 ± 3</td>
<td>127 ± 2*</td>
<td>123 ± 3*</td>
<td>126 ± 4*</td>
</tr>
<tr>
<td>HR (beats/min)</td>
<td>384 ± 9</td>
<td>424 ± 10†</td>
<td>387 ± 6‡</td>
<td>394 ± 8‡</td>
</tr>
<tr>
<td>Body wt (g)</td>
<td>219 ± 6</td>
<td>224 ± 7</td>
<td>237 ± 9*</td>
<td>244 ± 6‡</td>
</tr>
<tr>
<td>Left ventricular wt (mg)</td>
<td>475 ± 15</td>
<td>463 ± 13</td>
<td>497 ± 17‡</td>
<td>567 ± 16‡</td>
</tr>
<tr>
<td>Right ventricular wt (mg)</td>
<td>119 ± 3</td>
<td>112 ± 3</td>
<td>124 ± 5‡</td>
<td>130 ± 4*‡</td>
</tr>
<tr>
<td>Uterus wt (mg)</td>
<td>312 ± 14</td>
<td>331 ± 25</td>
<td>83 ± 13</td>
<td>157 ± 5</td>
</tr>
</tbody>
</table>

Data are means ± SEM. HR = heart rate.

Analysis of variance: *p<0.01 or †p<0.05 compared with sham-operated rats; ‡p<0.01 compared with noncastrated sinoaortic denervated rats; §p<0.01 compared with castrated sinoaortic denervated rats.
male SAD rats significantly reduced the ventricular hypertrophy in male rats and significantly increased the ventricular weight in female rats. Our data indicate that the observed changes in ventricular mass are the result of a direct action of sex hormones on the heart, since the levels of arterial hypertension were not modified by castrating SAD rats.

In an attempt to investigate the relative role played by sex hormones, groups of male and female castrated SAD rats were treated with either testosterone or estradiol. The efficacy of the substitutive hormonal therapy was proved by the heavier uteri, seminal vesicles, and prostates observed in treated rats compared to those of untreated, castrated animals (see Tables 1 and 2). The fact that the treatment of male and female castrated SAD rats with testosterone restored left ventricular hypertrophy to the same levels observed in male non-castrated SAD rats supports the view that this hormone exerts a facilitatory action on the development of ventricular hypertrophy. The findings of others in mice and rats and in baboons, together with the present results, also corroborate the concept that myocardial cells possess specific androgen receptors that are influenced by circulating testosterone, resulting in the growth of cells in the heart.

On the other hand, when other groups were treated with estradiol, complete suppression of ventricular hypertrophy was observed both in castrated and noncastrated males and castrated females, indicating an inhibitory influence of this hormone on ventricular hypertrophy.

Contrasting with other experimental models of hypertension, sinoaortic denervation-induced hypertension is exclusively characterized by an immediate and intense increase in sympathetic outflow. Thus the cardiac hypertrophy in our model must certainly be sympathetic nerve-dependent and not caused by pressure overload, since only a slight elevation of blood pressure occurs in SAD rats. The lack of cardiac hypertrophy in female rats and in estradiol-treated male rats indicates a possible inhibitory influence of this hormone on cardiac sympathetic activity. This view is supported by several studies demonstrating that long-term administration of estrogens to rats reduced their response to β-adrenergic agonists.

The sex differences we show in the present work and the evidence of androgen and estrogen receptors suggest that sex hormones may affect cardiac function directly and explain some differences between men and women relative to cardiopathy.

References

1. Tarazi RC, Levy MN. Cardiac responses to increased afterload: state of the art review. Hypertension 1982;4(suppl II): II-8–II-18
15. Salt PJ. Inhibition of noradrenaline uptake, in the isolated rat.
heart by steroids, clonidine and methoxylated phenylethyla-
16. Roberts JM, Insel PA, Goldfen BD, Goldfen A. α-Adrenore-
ceptors but not β-adrenoreceptors increase in rabbit uterus with
17. Koenig H, Goldstone A, Lu CY. Testosterone-mediated sexual
dimorphism of the rodent heart: ventricular lysosomes, mito-
chondria, and cell growth are modulated by androgens. Circ
Res 1982;50:782-787
18. Krieg M, Smith K, Bartsch W. Demonstration of a specific
androgen receptor in rat heart muscle: relationship between
binding, metabolism, and tissue levels of androgens. En-
docrinology 1978;103:1686-1694
19. Schaible TF, Malhotra A, Ciambrone G, Scheuer J. The ef-
fects of gonadectomy on left ventricular function and cardiac
contractile proteins in male and female rats. Circ Res
1984;54:38-49
20. McGill HC Jr, Sheridan PJ. Nuclear uptake of sex steroid
hormones in the cardiovascular system of the baboon. Circ Res
1981;48:238-244
21. Black DJ, Fregly MJ, Thrasher TN, Moreland AF. Reduced
beta-adrenergic responsiveness in rats treated with estrogenic
22. McGill HC Jr, Anselmo VC, Buchanan JM, Sheridan PJ. The
heart is a target organ for androgen. Science 1980;207:775-777
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