Sex Difference in Presynaptic Adrenergic Inhibition of Norepinephrine Release During Normoxia and Ischemia in the Rat Heart

Xiao-Jun Du, Anthony M. Dart, Rudolph A. Riemersma, and Michael F. Oliver

Using a perfused innervated rat heart model, we studied the sex difference in the sympathetic nerve stimulation-induced norepinephrine release and its presynaptic $\alpha_2$-adrenergic inhibition in normoxic and ischemic conditions. During normoxic perfusion, the $\alpha_2$-adrenoceptor antagonist rauwolscine resulted in a higher overflow of norepinephrine during nerve stimulation in females than in males ($p<0.05$). This more marked potentiation of norepinephrine overflow in females was accompanied by an increased chronotropic and inotropic response ($p<0.01$). During early stop-flow ischemia neural norepinephrine overflow was lower in female than in male hearts ($p<0.005$). Rauwolscine enhanced norepinephrine overflow more in females than in males ($p<0.05$), thereby eliminating the initial difference in norepinephrine overflow during ischemia between the two sexes. Ovariectomy attenuated the presynaptic $\alpha_2$-adrenergic inhibition of norepinephrine release compared with sham-operated females ($p<0.02$). No sex difference was found in either cardiac norepinephrine content or nonexocytotic norepinephrine overflow induced by a 40-minute period of stop-flow ischemia. Thus, presynaptic $\alpha_2$-adrenergic inhibition of myocardial norepinephrine release is greater in female than in male rats. This difference persists into the early phase of ischemia and is largely responsible for the lower neural norepinephrine release in the female heart. Female hormones may increase presynaptic $\alpha_2$-adrenergic activity in the heart. (Circulation Research 1991;68:827–835)

Clinical studies have suggested sex-related differences in the adrenergic reactivity to various stressors, including mental stress,1,2 head-up tilt,2 lower-body negative pressure,3 exercise,1,2,4,5 and insulin-induced hypoglycemia.5 When exposed to these stressful conditions, men show a more marked sympathoadrenal activation than do women, as evidenced by higher plasma catecholamine concentrations and higher urinary catecholamine outputs.1-4 Plasma concentrations of epinephrine and norepinephrine are also higher in males under resting conditions.2-4

Results from experimental studies of rats support these clinical observations. Cardiac sympathetic activation by drug-induced hypotension is more pronounced in males than in females.6 Furthermore, the female cardiovascular system is more tolerant to several pathological conditions such as circulatory shock,7 isoproterenol-induced myocardial necrosis,8 and myocardial ischemia and infarction.9,10 These pathological stimuli all activate the sympathoadrenal system.

The mechanisms responsible for these sex differences in adrenergic activation by stressful factors are largely unknown. A lower central sympathetic activity level in females has been proposed.2,5,11 Catecholamine content and metabolism in the central nervous system and adrenal medulla may also differ.12,13 Many studies have demonstrated that females have a higher density and activity of $\alpha$-adrenoceptors in various tissues.14-21 It is unclear whether presynaptic $\alpha_2$-adrenoceptors are more numerous in females, as one study suggests.22 Estrogen treatment may increase, and ovariectomy decrease, the number of $\alpha_2$-adrenoceptors.15-21 However, testosterone treatment has no such effect.17 One clinical study showed that estrogen attenuated the pressor response to mental stress.11

Because of these findings, it is logical to hypothesize that a sex difference in the mechanisms controlling sympathoadrenal function exists. One possibility is that there is a more potent presynaptic inhibition of norepinephrine release from sympathetic nerves in females. We have examined this possibility by using...
an in situ perfused, innervated rat heart model. In particular, we have studied the effects of $\alpha_2$-adrenoceptor inhibition and stimulation on sympathetic nerve stimulation–induced norepinephrine release under normoxic and ischemic conditions. In addition, we have also examined the effect of gonadectomy in female rats on these processes.

**Materials and Methods**

**Preparation**

Male and female Sprague-Dawley rats (9–10 weeks old, 200–290 g) were used. Experiments were performed alternately on male and female rats. No attempt was made in these studies to ascertain the phase of the estrous cycle of female rats at the time of study. Rats were anesthetized with pentobarbital (60 mg/kg i.p.) and heparinized (200 units i.v.). Hearts were perfused in situ with a modified Krebs-Henseleit solution containing (mM) $\text{Na}^+$ 148, $K^+$ 4.0, $Ca^{2+}$ 1.85, $Mg^{2+}$ 1.05, $HCO_3^{-}$ 25, $PO_4^{3-}$ 0.5, glucose 11, and EDTA 0.027. The perfusate was gassed continuously with 95% $O_2$-5% $CO_2$. Oxygenation of the perfusate was checked hourly with an IL 1302 pH/blood gas analyzer (Instrumentation Laboratory, Milan, Italy), giving a $P_O_2$ of 615±34 mm Hg ($\pm$SD), a pH of 7.40±0.02, and a $P_CO_2$ of 37±1 mm Hg. The temperature of the perfusate at the point it entered the heart was 37°C. Hearts were heavier in male ($0.77\pm0.07$ g wet wt, $n=73$) than in female ($0.72\pm0.06$ g wet wt, $n=74$, $p<0.01$) rats. Perfusion flow rates were carefully adjusted to estimated heart weight and were $5.24\pm0.33$ ml/min/kg for males and $5.18\pm0.35$ ml/min/kg for females (mean±SD, NS). At this flow rate, perfusion pressure was $42\pm2$ mm Hg and did not differ between the sexes. A cannula was introduced into the right atrium for collection of coronary venous effluent. Another 4F cannula connected to a pressure transducer (EM751, Elema, Stockholm, UK) was inserted into the left ventricular cavity through the apex. Left ventricular pressure, $dP/dt$, epicardial ECG, and perfusion pressure were recorded using an ink-jet recorder (Type EM 34, Elema-Schönander, Stockholm) or Gould TA 2000 recorder (Gould Inc., Cleveland, Ohio).

The left cervicothoracic stellate ganglion (with intact cardiac nerves) was prepared for subsequent electrical stimulation. The nerves were constantly superfused with warmed, oxygenated perfusate except when stimulated. Nerve stimulation was performed using a Model S88 stimulator (Grass Instrument Co., Quincy, Mass.) with a stimulus isolation unit (SIU 7). Stimulation was set at a pulse width of 2 msec, a current of 0.8 mA, and a frequency of 5 Hz. A 15-minute recovery period was allowed between successive nerve stimulations.

In ischemic series, global ischemia was produced by stopping the perfusion pump. During ischemia, myocardial temperature was kept between 36°C and 37°C by covering hearts with a thermostatic chamber. At the end of the ischemic period, hearts were reperfused at the preischemic flow rate. Finally, hearts were excised, blotted dry, and weighed.

The neural norepinephrine reuptake inhibitor desipramine (Sigma Chemical Co., St. Louis) was added to the perfusate (0.1 $\mu$M) in all nerve stimulation experiments. The drugs were infused by a pump (model 22, Harvard Apparatus, South Natick, Mass.) through a side port close to the heart at least 12 minutes before the next nerve stimulation (normoxic series) or before the second episode of ischemia (ischemic series).

A 20-minute stabilization period was always allowed before each experiment. Coronary effluent for norepinephrine analysis was collected over a 2-minute period starting with the nerve stimulation in normoxic perfused hearts. Coronary effluent was collected during the first 2.5 minutes of ischemia/reperfusion in the ischemic hearts (because there was little effluent collected during the first 30 seconds). Norepinephrine overflow in the absence of nerve stimulation was always low and did not differ in male and female groups ($0.82\pm0.17$ and $0.77\pm0.14$ pmol/g/min, respectively, NS). The reproducibility of successive nerve stimulations was examined in two groups of normoxic perfused male rat hearts. Six ($n=9$) or four ($n=7$) repetitive nerve stimulations of 30- or 60-second duration, respectively, were performed. A constant amount of norepinephrine overflow was observed (Figure 1).

**Biochemical Analysis**

Norepinephrine concentrations in the heart and in coronary effluent were measured, in duplicate, using a

![Figure 1. Reproducibility of norepinephrine (NE) overflow induced by repetitive sympathetic ganglion stimulation (six times for 30 seconds or four times for 60 seconds) in normoxic perfused male rat hearts. $S_2$-evoked NE overflow was 31.0±2.5 pmol/g for a 30-second stimulation and 79.7±13.5 pmol/g for a 60-second stimulation; both were set at 1 (first bar). NE overflow induced by subsequent nerve stimulation is presented as the ratio of that obtained during the first nerve stimulations ($S_1$). There was no significant decline in the amount of NE overflow (analysis of variance).](attachment:image.png)
radioenzymatic method. The coefficient of variation determined by repeated analysis of a sample with a norepinephrine concentration of 2 pmol/ml was 7%. The levels of lactate and lactate dehydrogenase in the effluent were analyzed enzymatically using commercial kits (Boehringer Mannheim, Mannheim, FRG) on a Cobas Bio centrifugal analyzer (Roche Analytic, Basel, Switzerland).

Protocols for Normoxic Series

**Experiment 1.** Total content of norepinephrine was measured in eight male and eight female rat hearts. After 20 minutes of in situ perfusion, hearts were excised, weighed, and quickly frozen on solid CO₂ and stored at −40°C until assayed.

**Experiment 2.** The effect of incremental doses of rauwolscine on neural norepinephrine overflow was examined in 10 male and 10 female rat hearts. Sympathetic nerve stimulation (30-second duration) was applied five times (S₁–S₅) at 15-minute intervals. The first (S₁) and last (S₅) nerve stimulations served as controls (no drug). Concentrations of rauwolscine were 0.1 µM (S₂), 1 µM (S₃), and 10 µM (S₄).

**Experiment 3.** Presynaptic α₂-adrenergic receptor–mediated inhibition of norepinephrine was studied in 12 male and 12 female rat hearts by using rauwolscine and clonidine. After an initial control stimulation (S₁, 60 seconds), a second stimulation (S₂, 60 seconds) was performed in the presence of rauwolscine (1 µM) and a third (S₃, 60 seconds) in the presence of clonidine (3 µM).

Protocol for Ischemic Series

**Experiment 1.** The effect of ischemia on nerve stimulation–induced norepinephrine release was studied in male and female hearts (n=9/group). A first nerve stimulation (S₁, 30 seconds) was used as an individual normoxic control. Hearts were then subjected to three episodes of ischemia in the sequence of 1-, 3-, and 6-minute duration and separated by 15-minute periods of normoxic reperfusion. Nerve stimulation (S₅–S₂) was applied during the final 30 seconds of each ischemic period.

**Experiment 2.** Effects of the α₂-adrenergic antagonist rauwolscine (1 µM) and the agonist clonidine (3 µM) on norepinephrine release during ischemia were studied in male (n=23) and female (n=24) rat hearts. Each heart underwent two episodes of 3-minute ischemia separated by a 15-minute recovery period. Norepinephrine overflow was determined during an initial nerve stimulation (60 seconds) in the final minute of ischemia (S₅). The hearts were then assigned to treatment with rauwolscine (male n=13, female n=13) or clonidine (male n=10, female n=11). Another nerve stimulation (S₃) was performed in the final minute of the second ischemic period.

**Experiment 3.** After 15–40 minutes of stop-flow ischemia, nonexocytotic norepinephrine release occurs in the isolated rat heart and is inhibited by desipramine. This ischemia-induced norepinephrine release was examined in male and female hearts (n=10/group). Desipramine was omitted from the perfusate. Hearts were subjected to 40 minutes of ischemia followed by reperfusion. Coronary effluent was collected during the first 3 minutes of reperfusion.

Protocol for Gonadectomy Series

Female Sprague-Dawley rats (6 weeks old) were gonadectomized or sham-operated using the techniques described by Waynforth. Body weights were measured weekly. The perfusion experiments were conducted during 7–8 weeks after the operation. Adequacy of the gonadectomy was assessed by inspection of possible residual ovarian tissue, atrophy of the uterus, and changes in body weight.

Nerve stimulation–induced norepinephrine overflow and the effects of rauwolscine (1 µM) and clonidine (3 µM) were studied using the same protocol as described for experiment 3 of the normoxic series. All the experiments were carried out in the presence of desipramine (0.1 µM) to inhibit the neuronal reuptake of norepinephrine, except in the study examining the effect of 40 minutes of ischemia on nonexocytotic norepinephrine release (experiment 3 of the ischemic series).

Statistical Analysis

The data are presented as mean±SEM. When possible, each animal served as its own control to eliminate between-animal variations and to improve the statistical power. Therefore, norepinephrine overflow data are also expressed as a ratio of values measured before and after an intervention. Statistical methods included one- or two-way analysis of variance (ANOVA) and unpaired and paired Student’s t test (with Bonferroni correction).
Results

Effect of Sympathetic Nerve Stimulation During Normoxia

Sympathetic nerve stimulation (60 seconds) caused a marked overflow of norepinephrine into the coronary effluent in male and female rat hearts. This increase in catecholamine release was reflected by an increased inotropic and chronotropic response (Table 1). None of these measurements were different between male and female hearts. Myocardial content of norepinephrine was also similar in males and females (7.7±0.8 versus 8.1±0.7 nmol/g).

Sex Difference in the Presynaptic Modulation of Norepinephrine Release During Normoxia

Rauwolscine increased norepinephrine overflow at each of the concentrations studied \((p<0.01)\) in both male and female hearts. The maximum effect of rauwolscine was observed at 1 \(\mu\)M (Figure 2).

At the optimum concentration of 1 \(\mu\)M, rauwolscine significantly enhanced norepinephrine overflow during the second nerve stimulation in both sexes \((p<0.01, \text{Table 2})\) and the \(S_2/S_1\) ratio was significantly higher in females than in males \((p<0.05, \text{Figure 3})\). The prestimulation levels of \(\pm dP/dt\) and heart rate were similar between and within groups (correction for follow-up tests). Differences were considered significant at a level of \(p<0.05\).

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related to differences in norepinephrine overflow (Figures 4A–4C).

In comparison to the effect of the antagonist rauwolscine, the agonist clonidine had little effect on norepinephrine overflow in normoxic perfused hearts. A small reduction in norepinephrine overflow was observed in females ($p<0.05$) but not in males (Table 2). This difference between the two sexes was not significant whether expressed in absolute terms or as a ratio of norepinephrine overflow during the control stimulation (Table 2, Figure 5).

**Sex Difference in Norepinephrine Overflow During Ischemia**

A progressive decline in norepinephrine overflow was observed within the first 6 minutes of ischemia in male and female hearts ($p<0.001$ by ANOVA). In males this reduction was not significant at 1 minute of ischemia compared with the preischemic control value ($32.5\pm7.6$ versus $40.4\pm6.2$ pmol/g, NS), but it was significant at 3 and 6 minutes of ischemia ($27.0\pm5.5$ and $20.1\pm3.0$ pmol/g, respectively, both $p<0.01$). An identical pattern emerged when the results were expressed as the ratio of the preischemic overflow (Figure 6). In females, norepinephrine overflow was significantly inhibited by ischemia at all times examined (1, 3, and 6 minutes of ischemia versus the preischemic control value: $19.8\pm3.8$, $14.9\pm2.6$, and $10.8\pm1.5$ versus $39.3\pm5.5$ pmol/g, $p<0.001$). The ratio of norepinephrine overflow during ischemia relative to the preischemic control was significantly lower in females than in males ($p<0.01$ by ANOVA, Figure 6). Meanwhile, nerve stimulation–induced increase in heart rate was also lower during ischemia in females than in males (combined data, +35±5 versus +52±6 beats/min, $p<0.05$ by ANOVA).

Norepinephrine overflow was also significantly increased by rauwolscine in male and female groups during ischemia, and the initial difference between the sexes in ischemic norepinephrine overflow was no longer significant (Table 2). The ratio of norepinephrine overflow in the presence and absence of rauwolscine was significantly higher in the female group ($p<0.05$, Figure 3), an effect mainly attributable to the difference in norepinephrine overflow between males and females in the absence of this drug. As in the normoxic experiments, changes in heart rate induced by nerve stimulation with and without rauwolscine were also related to the changes in norepinephrine overflow during ischemia (Figure 4D).

The effect of clonidine during ischemia differed from that during normoxia. Clonidine increased norepinephrine overflow in male hearts with an in-
creased ratio of norepinephrine overflow (Table 2, Figure 5), whereas no change in norepinephrine overflow had been observed during normoxia. In contrast to the clonidine-mediated reduction in norepinephrine overflow in normoxic female hearts, an insignificant increase in the ratio of norepinephrine overflow (S2/S1) in ischemia was observed (Figure 5). The ischemia-induced difference in neural norepinephrine overflow between both sexes was not affected by clonidine (Table 2).

Reperfusion after 40 minutes of ischemia evoked spontaneous norepinephrine overflow in both male and female hearts (186±21 versus 183±31 pmol/g, NS). Overflow of lactate and lactate dehydrogenase were also observed in male and female hearts (lactate, 19.6±0.7 versus 19.7±1.2 µmol/g; lactate dehydrogenase, 214±46 versus 208±32 microunits/g). No sex difference was found in all these changes.

Gonadectomy Series

Gonadectomy was associated with an increase in body weight, which was discernible starting from the second week after the operation. At the time the rats were killed, gonadectomized animals had a higher body weight (320±7 versus 258±5 g, p<0.001) and heart weight (1.02±0.3 versus 0.86±0.02 g, p<0.001) and a marked uterine atrophy (83±3 versus 463±26 mg, p<0.001) than sham-operated female rats. The perfusion flow rate was similar in the two groups (4.93±0.13 versus 4.73±0.11 ml/g/min, NS).

Norepinephrine overflow evoked by a control nerve stimulation (S1) did not differ between sham-operated and gonadectomized females. Norepinephrine overflow was significantly increased by rauwolscine in sham-operated controls (from 70.5±8.4 to 164.7±22.7 pmol/g, p<0.001) and in gonadectomized rats (from 88.7±13.2 to 140.5±18.7 pmol/g, p<0.01). However, the potentiation of norepinephrine overflow by rauwolscine was reduced after gonadectomy as the S2/S1 ratio was significantly lower (p<0.02, Figure 7). Norepinephrine overflow was not significantly affected by clonidine in gonadectomized and sham-operated animals and S2/S1 ratios were similar (0.94±0.17 versus 1.04±0.15, NS).

Prestimulation heart rate was similar in sham-operated and gonadectomized rat hearts (195±11 versus 180±13 beats/min, NS). The chronotropic response to nerve stimulation was not significantly affected by

![Figure 5](http://circres.ahajournals.org/)

**Figure 5.** Effect of the α-adrenoceptor agonist clonidine (3 µM) on norepinephrine (NE) overflow induced by sympathetic ganglion stimulation (NE) in male and female rat hearts during normoxia and during the final minute of ischemia (3 minutes). Results are presented as the ratio of individual control values measured during normoxia or during ischemia without clonidine (S1, open bars). Desipramine (0.1 µM) was used to inhibit neural NE uptake. n=10 to 12 hearts/group. *p<0.05 vs. S1-induced NE overflow.

![Figure 6](http://circres.ahajournals.org/)

**Figure 6.** Norepinephrine (NE) overflow from male and female rat hearts induced by sympathetic ganglion stimulation (5 Hz for 30 seconds) during normoxia and 1, 3, and 6 minutes of stop-flow ischemia. NE overflow during ischemia is expressed as the ratio of individual normoxic values (S1=1). Desipramine (0.1 µM) was used to inhibit neural NE uptake. Sex difference was significant (overall, p<0.001 by analysis of variance; at 3 and 6 minutes of ischemia, p<0.05 and p<0.01, respectively, by unpaired t test with Bonferroni correction; n=9 hearts/group).

![Figure 7](http://circres.ahajournals.org/)

**Figure 7.** Effect of female gonadectomy (FGX, n=14) or sham-operation (FSH, n=13) on the ratio of norepinephrine (NE) overflow evoked by sympathetic ganglion stimulation (5 Hz for 60 seconds) without (S1) and with (S2) the α2-adrenoceptor antagonist rauwolscine (1 µM) in normoxic perfused rat hearts.
gonadectomy. Changes in heart rate with and without rauwolscine were related to the changes in the amount of norepinephrine overflow (Figure 4E).

**Discussion**

**Sex Difference in the Presynaptic α2-Adrenergic Inhibition of Norepinephrine Release**

The present study has demonstrated, for the first time, a more potent α2-adrenergic presynaptic inhibition of exocytotic norepinephrine release in female than in male rat hearts. In the presence of the α2-adrenoceptor antagonist rauwolscine, norepinephrine overflow increased proportionally more in female rat hearts. These sex differences are not due to a leftward shift in the dose–response curve of rauwolscine in female rat hearts. The maximum effect of rauwolscine on norepinephrine overflow was achieved at 1 μM in both male and female rats. Clonidine, an α2-adrenergic agonist, significantly reduced norepinephrine overflow in females but not in males.

One may argue that the sex differences in norepinephrine release could simply be caused by a denser sympathetic innervation in female rat hearts. However, if this had been the case, then we would have expected norepinephrine overflow in females also to be higher in the absence of rauwolscine and the ratio of norepinephrine overflow with and without rauwolscine to remain unchanged. This was not so. Myocardial norepinephrine levels were also not different in both sexes. Another possibility is a less efficient norepinephrine uptake system in females. Our studies were always carried out in the presence of the neural reuptake inhibitor desipramine, and therefore, a sex difference in neural reuptake of norepinephrine, if it exists, seems to be an unlikely explanation. There are no published data to support this possibility, and one clinical study actually found a higher clearance of catecholamines from the circulation in females. In addition, estrogen could inhibit the extraneural norepinephrine uptake in rat heart in vitro. However, because this uptake mechanism plays a minor role in norepinephrine clearance from the synaptic cleft in our model, the magnitude of the sex difference in norepinephrine overflow seems too large to be explained this way.

Our evidence for the sex difference of presynaptic control of norepinephrine release does not rely only on the results of norepinephrine overflow. The inotropic and chronotropic potentiation by rauwolscine was also more marked in female than in male hearts. The possibility that this was due to a difference in postsynaptic α-adrenergic responsiveness between the sexes seems unlikely because α-receptors contribute little to the inotropic response to catecholamines. In gonadectomized rats the inotropic state of the heart is reduced, and therefore, a difference in the response to nerve stimulation between sham-operated and gonadectomized rat hearts cannot simply reflect norepinephrine overflow under these circumstances.

Taken together, our results suggest that the presynaptic α2-adrenergic inhibition of norepinephrine release plays a more important role in female than in male rat hearts.

**Mechanism**

Both the increase in norepinephrine overflow and the associated greater chronotropic response by rauwolscine was attenuated after gonadectomy, indicating a less effective α2-adrenergic presynaptic inhibition after removal of the ovaries. The adequacy of gonadectomy was documented by a marked uterine atrophy that has been attributed to the fall in estrogen. Therefore, a female hormone–mediated modulation of presynaptic α2-adrenoceptors is indicated. To the best of our knowledge, this is the first study to suggest a modulation by sex hormones of neurotransmission in the heart.

Sex differences in adrenoceptors have been extensively studied using pharmacological and radioligand techniques. Most studies have found a higher sensitivity and density of α-adrenoceptors in various tissues (blood vessels, neurons, platelets, uterus, bladder, and urethra) in females compared with males. Some studies show that this sex difference in α-adrenoceptors is restricted to the α2-subtype, but others found that α1-receptors also differed in both sexes. β-Adrenoceptors were similar in the two sexes. Furthermore, this sex difference in α-adrenoceptors does not persist after ovariectomy. Conversely, elevating estrogen levels in males and females increases the density of α1-receptors but has no such effect on β-receptors. Progesterone prevents the estrogen-induced rise in myometrial α2-adrenoceptors. Testosterone treatment does not affect α-adrenoceptors.

**Sex Difference in Norepinephrine Release During Ischemia**

A reduction in neural norepinephrine overflow during acute ischemia has been documented and is explained by presynaptic inhibition, enhanced neural reuptake, and energy depletion of sympathetic nerves.

Another important observation in our study is that ischemia reduces neural norepinephrine release to a greater extent in females than in males. In contrast, no sex difference exists in nonexocytotic norepinephrine release induced by a 40-minute period of ischemia. Interestingly, at 3 minutes of ischemia, administration of the α2-adrenoceptor antagonist rauwolscine led to a higher net increase in norepinephrine overflow in females and the initial difference in the total amount of norepinephrine overflow between the two sexes was no longer observed. Thus, the lower norepinephrine release during ischemia in female hearts is largely due to the greater α2-adrenoceptor–mediated presynaptic inhibition in female than in male hearts. This sex difference is observed only during ischemia and not during normoxia. It is probably the result of a higher prevailing norepinephrine level in the synaptic cleft.
caused by the ineffectiveness of washout during stop-flow ischemia. Our clonidine data also support this proposition. Because clonidine is a partial agonist, it will show an antagonistic effect when norepinephrine concentrations are high. Indeed, during ischemia, clonidine increased neural norepinephrine overflow in males. Although the rise in the female group was not significant, it was also not different from the male group. However, the overall effect of clonidine during ischemia is to enhance neural norepinephrine overflow (p < 0.01, combined data from both groups). The observed sex difference in presynaptic inhibition of neural norepinephrine release during ischemia may be an explanation for the early loss in catecholamines from adrenergic nerves after coronary ligation in one study with male rats (0.5 hour) but not in two other studies of female rats (3–8 hours). However, it remains to be seen whether there is a continuing nervous activity and efficient presynaptic inhibition on catecholamine release during prolonged ischemia.

**Implications**

The sympathetic nervous system plays an important role in the development of serious ventricular arrhythmias during acute myocardial ischemia. In anesthetized rats, the onset of ventricular arrhythmias is delayed and arrhythmia-induced death within the first 20 minutes of coronary ligation is less frequent in females than in males. In the Framingham Study, sudden cardiac death rates are higher in men than in women. However, sudden cardiac death, as a percentage of total mortality from coronary heart disease, is not significantly lower in women. Although ventricular tachyarrhythmias are more likely to be induced in electrophysiological testing in men than in women, these studies were not controlled for differences in the extent of coronary artery disease or extent of ventricular dysfunction. In addition, the majority of women in all these studies were of postmenopausal age. Thus, adequate clinical studies comparing the vulnerability to arrhythmias or sudden cardiac death in men and women do not appear to have been conducted. The difference in presynaptic inhibition shown in this study might lead to a reduced adrenergic stimulation and fewer serious arrhythmias and/or a reduction in sudden cardiac death in women. It would be interesting to test this possibility in men and premenopausal women.

**Acknowledgments**

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**References**

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