Cyclosporine A is a potent immunosuppressant agent used in organ transplantation to prolong the survival of allogenic grafts and in the treatment of autoimmune diseases (Cohen et al., 1984). However, the clinical use of cyclosporine is often associated with serious cardiovascular disorders such as hypertension (Schachter, 1988; Scherrer et al., 1990). Several mechanisms have been proposed to explain the hypertensive effect of cyclosporine such as sympathetic activation (Scherrer et al., 1990; Chiu et al., 1992), direct vasoconstriction (Xue et al., 1987), and/or endothelin release (Kon et al., 1990). Attenuation of arterial baroreceptor responsiveness is another mechanism that may contribute to cyclosporine-induced hypertension. Gerhardt et al. (1999) have shown that cyclosporine elevates blood pressure in kidney transplant recipients, and this effect was associated with a reduced baroreflex function. Cyclosporine has also been shown to shift the midranges of baroreflex control of sympathetic nerve activity and heart rate to higher arterial pressures (Ryuzaki et al., 1997). Spectral analysis of the arterial pressure-heart period relationship in patients with heart transplantation showed that cyclosporine reduces the overall gain of baroreflex responsiveness to orthostatic stimulus (Lucini et al., 2000). Notably, the attenuation of baroreflex responsiveness is considered as an important independent risk factor for cardiac mortality (Schwartz et al., 1984; Tsuji et al., 1994).

The exact mechanism by which cyclosporine attenuates the baroreflex function, and whether it involves central or peripheral pathways, is not clear. It has been suggested that an imbalance between the sympathetic and parasympathetic activities due to an enhanced sympathetic tone may explain cyclosporine-induced baroreflex dysfunction (Gerhardt et al., 1999). Cyclosporine increases the production of thromboxane A2 and disturbs the prostaglandin (prostaglandin I2 and thromboxane A2) balance (Perico et al., 1986). Given that prostaglandin I2 increases the baroreceptor sensitivity (Chen et al., 1990), the altered prostaglandin profile may result in reduced baroreceptor sensitivity.

Reported findings have shown that cyclosporine causes testicular dysfunction. Cyclosporine produces dose-dependent reductions in serum testosterone levels and intratesticular testosterone contents (Krueger et al., 1991; Bowman et al., 1997), and impairs spermatogenesis (Srinivas et al., 1998). The alterations in testicular function by cyclosporine...
can be reversed by testosterone replacement therapy (See-thalakshmi et al., 1990). Interestingly, testosterone has been shown in a recent study from our laboratory to selectively modulate the baroreceptor control of reflex bradycardia versus no effect on reflex tachycardia (El-Mas et al., 2001). In the latter study, we provided the first experimental evidence that testosterone facilitates baroreflex function through a mechanism that involves enhancement of cardiac vagal activity. With this idea in mind, the ability of cyclosporine to reduce baroreflex responsiveness and serum testosterone levels raises the possibility that these two effects of cyclosporine are correlated. This assumption has not yet been investigated.

The present study, therefore, addressed two important questions pertinent to the hypothesis that testosterone modulates cyclosporine-induced baroreflex impairment: first, whether cyclosporine reduces baroreceptor responsiveness via inhibition of the facilitatory effect of testosterone on baroreflexes; and second, whether alterations in the cardiac autonomic control contribute to cyclosporine-testosterone baroreflex interaction. To accomplish these goals, baroreflex curves relating reflex HR responses to increments in blood pressure evoked by phenylephrine were established in conscious, freely moving, sham-operated, castrated rats and in CAS + T rats treated with cyclosporine or vehicle. Baroreflex responsiveness was measured in the absence and presence of atropine (muscarinic blocker) or propranolol (β-adrenergic blocker) to evaluate the roles of cardiac vagal and sympathetic activity, respectively, in the HR responses. The slopes of the curves were taken as an index of BRS. Plasma testosterone levels were also measured and correlated to changes in BRS.

Materials and Methods

Male Wistar rats (230–280 g; High Institute of Public Health, Alexandria, Egypt) were used in the present study.

Castration. Castration was performed as described in our previous studies (El-Mas et al., 2001) and by others (Milla et al., 1992). A single 2- to 3-cm incision was made in the scrotum. The testes were isolated, tied off with sterile suture, and removed. The skin was sutured, and the rats were allowed 10 days prior to intravascular cannulation. Sham operation involved exposure of the testes without isolation. Each rat received an intramuscular injection of 60,000 U of penicillin G benzathine and penicillin G procaine (Penicid) and was housed in a separate cage. Intravascular cannulation was performed 10 days later.

Intravascular Cannulation. The method described in our previous studies (El-Mas and Abdel-Rahman, 1992; El-Mas et al., 1997a,b) for measurement of blood pressure was adopted. Briefly, the rats were anesthetized by thiopental (50 mg/kg i.p.). Catheters (polyethylene 50) were placed in the abdominal aorta and vena cava via the femoral artery and vein for measurement of blood pressure and i.v. administration of drugs, respectively. The catheters were inserted about 5 cm into the femoral vessels and secured in place with sutures. The arterial catheter was connected to a Gould-Statham (Oxnard, CA) pressure transducer, and blood pressure was displayed on a Grass polygraph (model 7D; Grass Instruments, Quincy, MA). The heart rate was computed from blood pressure waveforms by a Grass tachograph and displayed on another channel of the polygraph.

Finally, the catheters were tunneled subcutaneously and exteriorized at the back of the neck between the scapulae. The catheters were flushed with heparin (100 U/ml) and plugged by stainless steel pins. Incisions were closed by surgical clips and swabbed with povidone-iodine solution. Each rat received an intramuscular injection of 60,000 U of penicillin G benzathine and penicillin G procaine in an aqueous suspension (Penicid) and was housed in a separate cage. The experiment started 24 and 72 h later. Experiments were performed in strict accordance with institutional animal care and use guidelines.

Measurement of Plasma Testosterone. A blood sample (0.4 ml) was withdrawn from the arterial line of each rat on the morning of the primary experiment day immediately before baroreflex testing. The plasma testosterone level was measured by radioimmunoassay (Diagnostic Products Corp., Los Angeles, CA).

Protocols and Experimental Groups. A total of six groups of male rats (sham, castrated, CAS + T, CyA, CAS + CyA, and CAS + CyA + T+) were used in this study to investigate the modulatory effect of testosterone on cyclosporine-induced attenuation of reflex HR responses and the relative contributions of vagal and sympathetic activities to these responses. Each rat in a particular group was employed in two experiments (24 and 72 h after instrumentation) to test the effect of atropine or propranolol on reflex HR responses to peripherally mediated increases in MAP evoked by phenylephrine. In the primary experiment, approximately 50% of the rats in a given group received atropine and the other 50% received propranolol. In the secondary experiment, the administration of atropine and propranolol was crossed over. Cyclosporine (20 mg/kg, dissolved in sesame oil) or an equal volume of vehicle was injected subcutaneously in single daily doses for 13 consecutive days. The first dose of cyclosporine or vehicle was given 11 days before the primary experiment of baroreflex testing. Testosterone (1 mg dissolved in sesame oil) was injected subcutaneously in single daily doses for 8 consecutive days starting 5 days before the primary experiment (Seidenfeld et al., 1980; El-Mas et al., 2001). The last dose of cyclosporine, testosterone, or vehicle was injected on the morning of the secondary experiment day.

On the day of the experiment (24 or 72 h after intravascular cannulation), the arterial catheter was connected to a pressure transducer for measurement of blood pressure and HR as mentioned above. A period of 30 min was allowed at the beginning of the experiment for stabilization of blood pressure and HR. Baroreflex curves of phenylephrine were generated as previously described (Abdel-Rahman, 1999; El-Mas et al., 2001) in all rats before and 10 min after atropine or propranolol (1 mg/kg each, i.v.). Comparison of the BRs before and after atropine or propranolol would allow a proper assessment of the relative contributions of the vagal and sympathetic autonomic components to the reflex HR responses (Abdel-Rahman, 1999; El-Mas et al., 2001). The doses of atropine and propranolol used in the present study have been shown to be adequate for muscarinic and β-adrenergic blockade, respectively (Coleman, 1980; Abdel-Rahman, 1999; El-Mas et al., 2001).

For the generation of the baroreflex curves, randomized i.v. doses of phenylephrine (0.5–8 μg/kg) were injected at 5-min intervals as in our previous studies (El-Mas and Abdel-Rahman, 1997; El-Mas, 1998, 1999). This time interval was adequate for blood pressure and HR to regain baseline levels. Phenylephrine was dissolved in saline, and the injection volume was kept constant at 0.05 ml/100 g of body weight with a flush volume of approximately 0.1 ml of saline. The MAP (diastolic plus one-third pulse pressure) and HR values were measured, and the peak changes in both variables (ΔMAP and ΔHR) were used for construction of the baroreflex curves.

Drugs. Testosterone (Organon NV, Oss, The Netherlands), phenylephrine hydrochloride, atropine sulfate, propranolol hydrochloride (Sigma-Aldrich, St. Louis, MO), thiopental (Biochimie GmbH, Vienna, Austria), povidone-iodine solution (Betadine; Nile Pharmaceutical Co., Cairo, Egypt), and Penicid (Cid Pharmaceutical Co., Cairo, Egypt) were purchased. Cyclosporine A was a gift from Novartis Pharma, AG (Basel, Switzerland). A fresh solution of cyclosporine in sesame oil was prepared every 3 days and kept in the refrigerator.
Statistical Analysis. Values are expressed as mean ± S.E.M. The relationship between increases in MAP and associated decreases in HR was assessed by regression analysis for individual animals as described in our previous studies (El-Mas and Abdel-Rahman, 1992, 1998; El-Mas et al., 2001). The regression coefficient (slope of the regression line) expressed as beats per minute per mm Hg was taken as an index of BRS. Analysis of variance (ANOVA) followed by a Newman-Keuls post hoc analysis was used for multiple comparisons with the level of significance set at P < 0.05.

Results

Cyclosporine-Testosterone Baroreflex Interaction. The baseline values of MAP measured in conscious, freely moving rats on the day of the experiment were similar in all groups of rats (Table 1). The baseline HR values were not altered by castration or testosterone replacement, whereas they were significantly (P < 0.05) increased in cyclosporine-treated groups (CyA, CAS + CyA, and CAS + CyA + T) compared with sham-operated values (Table 1). Pooled data obtained prior to atropine or propranolol administration showed that i.v. administration of phenylephrine (0.5–8 µg/kg) produced dose-related increases in MAP that were associated with reflex decreases in HR (Table 2). Multiple comparisons (ANOVA) of the mean pressor responses to phenylephrine revealed that these responses were not affected by short-term castration but showed significant (P < 0.05; ANOVA) reductions by cyclosporine treatment (20 mg/kg/day for 11–13 days) compared with sham-operated values (Table 2). The reflex bradycardic responses were significantly (P < 0.05) reduced by castration and by cyclosporine treatment (Table 2). Analysis of the baroreflex curves, relating decreases in HR responses to phenylephrine-induced increases in MAP, revealed a lesser steep regression line in the case of castrated or cyclosporine-treated rats; i.e., for a comparable rise in MAP, revealed a greater pressor response and by cyclosporine treatment (Table 2). The correlation coefficients of the regression lines were highly significant (P < 0.001) and ranged from 0.89 to 0.99.

To eliminate any role for the reduced pressor responsiveness to phenylephrine observed in cyclosporine-treated rats and its possible contribution to cyclosporine-induced impairment of reflex bradycardia, the HR responses to similar increases (approximately 35 mm Hg) in MAP were computed for individual rats, regardless of the dose of phenylephrine used, and the BRS was measured by calculation of the ratio ∆HR/∆MAP (El-Mas and Abdel-Rahman, 1993). The results showed that the reflex bradycardic (Fig. 3B) and BRS (Fig. 3C) responses, evoked by comparable increases in MAP (Fig. 3A), were significantly (P < 0.05) reduced by cyclosporine compared with the sham-operated rats. Similar reductions in the baroreflex bradycardia (Fig. 3B) and BRS (Fig. 3C) were demonstrated in CAS and CAS + CyA rats.

Changes in plasma testosterone levels evoked by castration, testosterone replacement, cyclosporine, or their combinations are shown in Fig. 2B. Plasma testosterone levels were significantly (P < 0.05) reduced by castration or cyclosporine compared with sham-operated levels (15.6 ± 5.3, 41.6 ± 9.9, and 105.4 ± 21.2 ng/dl, respectively). Treatment of CAS or CAS + CyA rats with testosterone restored the physiological levels of the hormone (Fig. 2B).

Effects of Muscarinic or β-Adrenergic Blockade on Reflex Heart Rate Responses. The effects of muscarinic or β-adrenergic blockade with atropine and propranolol, respectively, on peripherally mediated increases in MAP and reciprocal changes in HR are shown in Table 3 and Figs. 4 and 5. In sham-operated rats, muscarinic blockade by atropine (1 mg/kg, i.v.) caused an upward shift of the baroreflex curve relating the pressor responses of phenylephrine to the associated reflex bradycardic responses and elicited a significant reduction in the BRS from −1.36 ± 0.14 to −0.59 ± 0.11 beats/min/mm Hg (Fig. 4A). Qualitatively similar effects for atropine were demonstrated in castrated and in cyclosporine-treated rats (Fig. 4, B and D). However, the percentage of reductions in BRS by atropine in castrated and cyclosporine-treated rats (30.34 ± 9.98 and 40.15 ± 7.99%, respectively) were significantly less than that of sham-operated rats (57.67 ± 4.37%; Table 3). The atropine-induced reductions in BRS of CAS + CyA rats were not statistically different from those of castrated or cyclosporine-treated rats (Table 3). The effects of atropine on the baroreflex curves and BRS were restored to sham-operated levels after testosterone replacement in CAS but not in CAS + CyA rats (Fig. 4, C and F; Table 3).

Contrary to the effect of atropine, β-adrenergic blockade by propranolol (1 mg/kg, i.v.) in sham-operated rats caused a slight upward shift in the baroreflex curve and insignificant decreases in the BRS from −1.59 ± 0.14 to −1.24 ± 0.21 beats/min/mm Hg (Fig. 5A). This represented a 22.35 ± 10.55% reduction in the BRS (Table 3), which suggests a minor contribution of the cardiac sympathetic activity in the reflex bradycardia. The effects of propranolol on the baroreflex curve of phenylephrine (Fig. 5) and the percentage re-

### Table 1

<table>
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<tr>
<th>Group</th>
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<th>MAP</th>
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<tr>
<td>Sham</td>
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<td>108.6 ± 3.9</td>
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<td>402.5 ± 11.6</td>
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<td>CyA</td>
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<td>427.2 ± 10.7*</td>
</tr>
<tr>
<td>CAS + CyA</td>
<td>12</td>
<td>103.1 ± 2.8</td>
<td>475.0 ± 10.9*</td>
</tr>
<tr>
<td>CAS + CyA + T</td>
<td>13</td>
<td>99.2 ± 3.1</td>
<td>452.5 ± 11.6*</td>
</tr>
</tbody>
</table>

* P < 0.05 compared with sham-operated value.
duction in BRS (Table 3) were not significantly altered by castration, cyclosporine, or their combination.

**Discussion**

Cyclosporine A, one of the most effective immunosuppressant agents, has been shown in clinical and experimental studies to impair the arterial baroreceptor function (Ryuzaeki et al., 1997; Gerhardt et al., 1999; Lucini et al., 2000). The cyclosporine-induced baroreflex dysfunction

### Table 2

<table>
<thead>
<tr>
<th>Group</th>
<th>Phenylephrine (μg/kg, i.v.)</th>
<th>0.5</th>
<th>1.0</th>
<th>2.0</th>
<th>4.0</th>
<th>8.0</th>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sham</td>
<td>ΔMAP</td>
<td>7.7 ± 1.0</td>
<td>17.8 ± 2.3</td>
<td>27.8 ± 2.3</td>
<td>41.1 ± 2.4</td>
<td>52.1 ± 1.6</td>
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<td>ΔHR</td>
<td>-10.8 ± 3.4</td>
<td>-24.2 ± 3.3</td>
<td>-35.4 ± 3.4</td>
<td>-53.5 ± 3.8</td>
<td>-81.9 ± 5.5</td>
</tr>
<tr>
<td>CAS</td>
<td>ΔMAP</td>
<td>7.2 ± 1.8</td>
<td>15.0 ± 2.1</td>
<td>23.1 ± 2.9</td>
<td>35.7 ± 3.8</td>
<td>49.7 ± 4.5</td>
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<tr>
<td></td>
<td>ΔHR</td>
<td>-5.0 ± 1.5</td>
<td>-13.9 ± 2.3*</td>
<td>-21.5 ± 2.4*</td>
<td>-31.1 ± 3.0*</td>
<td>-42.3 ± 3.7*</td>
</tr>
<tr>
<td>CAS + T</td>
<td>ΔMAP</td>
<td>7.0 ± 1.3</td>
<td>12.3 ± 1.7</td>
<td>24.0 ± 3.0</td>
<td>33.3 ± 3.8</td>
<td>46.0 ± 4.2</td>
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<tr>
<td></td>
<td>ΔHR</td>
<td>-9.0 ± 1.6</td>
<td>-20.0 ± 2.8</td>
<td>-28.0 ± 2.7</td>
<td>-45.0 ± 4.2</td>
<td>-71.2 ± 7.8</td>
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<tr>
<td>CyA</td>
<td>ΔMAP</td>
<td>4.9 ± 1.0</td>
<td>9.5 ± 1.4*</td>
<td>14.5 ± 1.7*</td>
<td>24.0 ± 2.0*</td>
<td>39.1 ± 2.4*</td>
</tr>
<tr>
<td></td>
<td>ΔHR</td>
<td>-2.5 ± 1.4</td>
<td>-6.5 ± 1.7*</td>
<td>-14.1 ± 2.8*</td>
<td>-22.5 ± 3.0*</td>
<td>-38.4 ± 3.2*</td>
</tr>
<tr>
<td>CAS + CyA</td>
<td>ΔMAP</td>
<td>9.4 ± 4.6</td>
<td>9.4 ± 1.4*</td>
<td>13.1 ± 1.6*</td>
<td>19.1 ± 2.5*</td>
<td>32.6 ± 3.9*</td>
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<tr>
<td></td>
<td>ΔHR</td>
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<td>-7.5 ± 2.0*</td>
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<td>-16.7 ± 2.7*</td>
<td>-26.3 ± 3.4*</td>
</tr>
<tr>
<td>CAS + CyA + T</td>
<td>ΔMAP</td>
<td>5.4 ± 1.1</td>
<td>8.4 ± 1.7*</td>
<td>12.9 ± 1.8*</td>
<td>22.1 ± 2.5*</td>
<td>35.3 ± 3.1*</td>
</tr>
<tr>
<td></td>
<td>ΔHR</td>
<td>-6.9 ± 2.2</td>
<td>-10.8 ± 2.8*</td>
<td>-15.0 ± 2.9*</td>
<td>-20.9 ± 3.7*</td>
<td>-27.7 ± 3.3*</td>
</tr>
</tbody>
</table>

*P < 0.05 compared with sham-operated values.
has been implicated in the pressor effect that develops during cyclosporine therapy (Gerhardt et al., 1999; Lucini et al., 2000). A similar correlation between hypertension and baroreflex impairment has been reported in humans (Goldstein, 1983) and experimental models of hypertension (Gordon and Mark, 1983; Abdel-Rahman and Wooles, 1987). The exact mechanism that underlies baroreflex impairment by cyclosporine is not clear. One possible explanation may involve the ability of cyclosporine to produce testicular dysfunction and lower plasma testosterone levels (Krueger et al., 1991; Bowman et al., 1997). Testosterone has been shown in a recent study from our laboratory to facilitate baroreflex responsiveness via enhancing cardiac vagal activity (El-Mas et al., 2001). The present study tested the hypothesis that cyclosporine impairs baroreflex function via reducing plasma levels of testosterone and inhibiting its facilitatory effect on cardiac vagal control. The effects of short-term cyclosporine administration on plasma testosterone levels and reflex bradycardic responses that developed secondary to baroreceptor loading by phenylephrine were evaluated in sham-operated rats as well as in castrated rats with and without testosterone replacement. Baroreflex curves relating increases in MAP evoked by phenylephrine to the associated decreases in HR were constructed, and the slopes were taken as a measure of BRS (El-Mas and Abdel-Rahman, 1992, 1998; El-Mas et al., 2001). The relative contributions of the sympathetic and parasympathetic components to the reflex bradycardic responses were also investigated.

The present findings that castration impairs arterial baroreceptor control of reflex bradycardia and that testosterone replacement restores BRS to sham-operated levels support our previous findings (El-Mas et al., 2001) and suggest that testosterone exerts a favorable effect on baroreceptor function. The current study presents evidence that implicates the male gonadal hormone testosterone in the depressant effect of cyclosporine on baroreflexes. This notion is supported by two observations. First, short-term castration or cyclosporine treatment caused remarkable reductions in plasma testosterone levels and similar attenuation of the baroreceptor control of reflex bradycardia, as suggested by the significant reduction in the slope of the regression line (BRS), relating peripherally mediated elevations in mean arterial pressure to the associated decreases in heart rate. Second, the ability of cyclosporine to impair reflex bradycardia was abolished when administered to testosterone-depleted (i.e., castrated) rats. Comparison of the slopes of the regression lines revealed that BRS values in castrated, cyclosporine-treated, and CAS + CyA rats were not statistically different. Taken together, the findings that cyclosporine lowered plasma testosterone levels and attenuated reflex bradycardia in sham-operated but not in castrated rats establish the first experimental evidence that inhibition of testosterone-mediated baroreflex facilitation may account, at

<table>
<thead>
<tr>
<th>Group</th>
<th>Atropine % Reduction</th>
<th>Propranolol % Reduction</th>
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</thead>
<tbody>
<tr>
<td>sham</td>
<td>n=7, 57.67 ± 4.37</td>
<td>n=6, 22.35 ± 10.55</td>
</tr>
<tr>
<td>CAS</td>
<td>n=7, 30.34 ± 9.98*</td>
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</tr>
<tr>
<td>CyA</td>
<td>n=8, 40.15 ± 7.99*</td>
<td>n=8, 10.54 ± 10.19</td>
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<tr>
<td>CAS + CyA</td>
<td>n=5, 23.10 ± 7.33*</td>
<td>n=7, 7.05 ± 16.54</td>
</tr>
<tr>
<td>CAS + CyA + T</td>
<td>n=6, 25.61 ± 13.32*</td>
<td>n=7, 12.03 ± 10.11</td>
</tr>
</tbody>
</table>

* P < 0.05 compared with sham-operated values.

Fig. 3. Effects of CyA on heart rate responses (panel B) and BRS (panel C) values evoked by similar increases in mean arterial pressure (panel A) in conscious, freely moving, sham-operated, CAS rats and in CAS + T rats. Values are expressed as mean ± S.E.M. of the pooled data obtained prior to atropine or propranolol administration. * and #, P < 0.05 compared with sham-operated and CAS + T rat values, respectively.
least partly, for the deleterious effect of cyclosporine on baroreflex function.

We have recently shown that the facilitatory effect of testosterone on baroreceptor control of reflex bradycardia is mediated via enhancement of cardiac vagal activity (El-Mas et al., 2001). The present study, therefore, attempted to investigate whether cyclosporine impairs baroreflexes through interfering with the modulatory effect of testosterone on vagal function. This objective was accomplished by evaluating the effect of selective muscarinic or β-adrenergic blockade by atropine and propranolol, respectively, on reflex bradycardic responses. Similar to earlier reports (Glick and Braunwald, 1965; Coleman, 1980), the present results obtained from sham-operated rats showed that the reflex bradycardic responses were mediated predominantly via increased cardiac vagal activity, because atropine caused approximately 60% reduction in BRS compared with only 20% in the case of propranolol. The effects of castration, cyclosporine, and their combination on the vagal contribution to reflex bradycardia paralleled their effects on BRS. The ability of atropine to reduce the predominantly vagally mediated reflex bradycardic responses was similarly attenuated in castrated rats and cyclosporine-treated rats. These findings suggest that testosterone depletion evoked surgically (by castration) or chemically (by cyclosporine) produced comparable impairment of the

Fig. 4. Effect of muscarinic blockade by atropine (1 mg/kg, i.v.) on baroreflex curves relating decreases in heart rate to phenylephrine-induced elevations in mean arterial pressure in conscious, freely moving, sham-operated, CAS and CAS + T rats in the presence and absence of CyA. Insets show the slopes (BRS, beats per minute per mm Hg) of the baroreflex curves. Values are expressed as mean ± S.E.M. *, P < 0.05 compared with the “before” values.
vagal component. The view that the inhibitory effect of cyclosporine on vagal activity is testosterone-related gains further support from the observation that treatment of castrated rats with cyclosporine caused no additional decline in the overall contribution of the vagal component to reflex bradycardia. In effect, the reduction in BRS evoked by atropine in CAS + CyA rats was not statistically different from that of castrated rats. Collectively, these findings may plausibly suggest that cyclosporine counteracts testosterone-mediated baroreflex enhancement via compromising its facilitatory effect on the cardiac vagal control.

It is important, however, before this conclusion is accepted, to comment on two potential limitations. The first relates to the possibility that the lack of an action of cyclosporine on BRS in castrated rats may be accounted for by the presence of a significantly lower baseline BRS in these rats as compared with sham-operated rats. It could be argued, therefore, that the BRS in castrated rats may have reached its nadir so that cyclosporine cannot depress it further. However, this issue may be addressed by the present finding that muscarinic blockade by atropine produced a significant attenuation of the BRS in castrated rats. The susceptibility of the remaining baroreceptor activity in castrated rats to additional attenuation by atropine rules out the possibility that its low levels can account...
for the absence of cyclosporine effect on BRS in these rats. The second limitation pertains to the finding that cyclo-
sporine significantly reduced BRS in testosterone-replaced castrated rats, which have restored physiological levels of the hormone. This finding should not be interpreted to argue against a modulatory role for testosterone in cyclo-
sporine-baroreflex interaction. Instead, it may suggest that the inhibitory effects of cyclosporine on the baroreflex and vagal responses to testosterone are not due to inhibition of the testicular release of the hormone per se but due to the interaction of cyclosporine with specific receptors or sites within the baroreflex arc that mediate the hormone effect on baroreflex function. Possible targets for such interaction are the nucleus ambiguus and the dorsal motor nucleus of the vagus, brainstem areas in which androgen receptors have been identified (Sheridan and Weakley, 1982; Freeman, 1988) and are known to play a crucial role in the control of central vagal discharges (Ciriello and Calaresu, 1979; Van Giersbergen et al., 1992). More studies are needed, however, to determine the exact mecha-
nism(s) involved in the interaction between cyclosporine and gonadal hormones on baroreflex function.

The present finding that cyclosporine reduced the pressor responsiveness to phenylephrine deserves a comment. Whereas this finding is consistent with the reports that cyclo-
sporine decreases pressor responses were considered (see Fig. 3). It is conceivable, therefore, to propose that the altered \( \alpha_1 \)-adrenergceptor responsiveness has no impact on the cyclosporine-induced impairment of baroreflex function. In summary, the present study sought evidence to implicate androgens in the depressant effect of the immunosup-
pressant drug cyclosporine on baroreceptor control of HR in conscious rats. Short-term castration impaired, whereas tes-
tosterone replacement restored, BRS to sham-operated levels, suggesting a favorable effect for testosterone on barore-
flexes. Cyclosporine decreased plasma testosterone levels and reduced BRS to levels similar to those of castrated rats. Muscarinic blockade by atropine remarkably reduced BRS, an effect that was comparably attenuated by castration, cy-
clorsporine, or their combination. It is concluded that cy-
clorsporine impairs baroreceptor control of reflex bradycardia via reducing plasma testosterone levels and interrupting the facilitatory effect of the hormone on the autonomic (vagal) control of the heart. The finding that CyA significantly re-
duced BRS in testosterone-replaced castrated rats, i.e., in the presence of physiological levels of the hormone, infers that CyA interferes with baroreflex and vagal functions through altering the interaction of testosterone at specific target sites controlling these functions.

Acknowledgments

We thank Novartia Pharm, AG (Basel, Switzerland) for generously supplying cyclosporine A.

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