Short-term fluoxetine treatment enhances baroreflex control of sympathetic nervous system activity after hindlimb unloading

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Moffitt, Julia A., and Alan Kim Johnson. Short-term fluoxetine treatment enhances baroreflex control of sympathetic nervous system activity after hindlimb unloading. Am J Physiol Regul Integr Comp Physiol 286: R584–R590, 2004; 10.1152/ajpregu.00223.2002.—Data in humans indicate that individuals with orthostatic hypotension that are refractory to other traditional forms of therapy are responsive to selective serotonin reuptake inhibitor (SSRI) treatment. We tested the hypothesis that SSRI administration would help correct the attenuated baroreflex control of sympathetic nervous system activity in the hindlimb-unloaded (HU) rat model of cardiovascular deconditioning. An initial study was conducted to determine the time course of effects of fluoxetine (Flu) administration on baroreflex control of lumbar sympathetic nerve activity (LSNA) in conscious, chronically instrumented rats. Animals received either vehicle (Veh, sterile water) or 10 mg/kg Flu for 1, 4, or 16 days of treatment. Data indicate that while 1-day and 16-day Flu administration did not affect baroreflex function, baroreflex control of LSNA was enhanced after 4-day (short term) Flu administration. HU rats were then treated with Flu for 4 days and compared with HU rats receiving Veh and to casted control rats maintained in the normal posture that received either Veh or short-term Flu treatment. Similar to pilot data, short-term Flu treatment enhanced baroreflex control of LSNA in both HU rats and control rats. These data taken together indicate that baroreflex control of sympathetic nervous system activity is a possible mechanism responsible for the successful treatment of orthostatic intolerance with Flu.

serotonin; microgravity; spaceflight

The neurotransmitter 5-hydroxytryptamine (5-HT; serotonin) plays an integral role in mediating a number of physiological processes, including behavior and cardiovascular function (16, 26). Indeed, the class of drugs known as selective serotonin reuptake inhibitors (SSRIs) is very successful in the treatment of psychological depression and is currently among the most widely prescribed medications. Interestingly, although administration of an SSRI causes an immediate increase in synaptic release of 5-HT, antidepressant effects are not experienced until approximately 3–4 wk of chronic administration in humans (26, 33) and 2 wk in rat models of affective disorder (8). Unlike tricyclic antidepressants, SSRIs have been shown to elicit fewer negative side effects (27). They have been demonstrated to exert a protective effect against myocardial infarction (30) and have been shown to be an effective therapy for dysautonomia and reflex syncope syndromes.

In the current studies, we wanted to determine if the SSRI fluoxetine (Flu) would ameliorate baroreflex dysfunction in the HU rat model of cardiovascular deconditioning. Due to the unknown time course of autonomic effects associated with SSRI treatment, we first conducted an initial study to determine if significant effects on the sympathetic nervous system could be elicited in rats. We then determined whether these effects required acute or chronic SSRI administration. We hypothe-
METHODS

Animals

Male Sprague-Dawley rats (n = 33) obtained from Harlan (Indianapolis, IN) were used for all experimental procedures. Animals were housed individually in environmentally controlled conditions and maintained on a 12:12-h light-dark cycle and given food and water ad libitum. All experiments were conducted in accordance with the National Institutes of Health Guide for the Care and Use of Laboratory Animals and were approved by the Institutional Animal Care and Use Committee of the University of Iowa.

Time Course of Flu

Four groups of rats were used to evaluate the time course of the effects of Flu administration on baroreflex control of HR and sympathetic nervous system activity in a pilot study. Rats received injections of sterile water [vehicle (Veh); 1 ml/kg ip; n = 3] for a period of 16 days or received chronic injections of the SSRI Flu for 16 days (Flu-D16; 10 mg/kg ip; n = 3), a short-term course of treatment for 4 days (Flu-D4; 10 mg/kg ip; n = 3), or an acute treatment of Flu 1 day before baroreflex testing (Flu-D1; 10 mg/kg ip; n = 3). This dose of Flu has been well documented as an appropriate dose to produce maximal selective inhibition of 5-HT reuptake in vivo both acutely and chronically (13). Animals were weighed and given injections at approximately the same time each day (1600–1700). The drug fluoxetine hydrochloride (Flu) was obtained from Sigma (St. Louis, MO). Although the sample size for each group in this study was small, we repeated the primary finding (4-day enhancement) in the subsequent study that focused on the effects of SSRI administration after hindlimb unloading.

SSRI Treatment and Hindlimb Unloading

Male Sprague-Dawley rats (n = 21) obtained from Harlan were randomly assigned to HU or casted control (CC, see below) groups. Both groups were then subdivided into CC and HU groups that received either Veh or Flu treatment. Thus this study contained four experimental groups: casted-control vehicle (CC-Veh; n = 5), hindlimb unloading-vehicle (HU-Veh; n = 6), casted-control fluoxetine (CC-Flu; n = 5), and hindlimb unloading-fluoxetine (HU-Flu; n = 5). Rats in the Flu-treated groups received Flu injections (10 mg/kg ip) on the last 4 days of the CC or HU protocol. This time period was chosen based on pilot data from the time-course experiments that demonstrated that baroreflex function was enhanced in response to this treatment regimen. The remaining groups of CC and HU rats received injections of sterile water (1 ml/kg) on the last 4 days of the protocol. HU rats were acclimated to the unloading procedure by temporarily suspending the hindlimbs 2 h/day for 2 consecutive days before the HU procedure. The hindlimbs of HU rats were then elevated with a harness attached to the proximal two-thirds of the tail by modifying a technique previously described (14). Briefly, two hooks were attached to the tail with moleskin adhesive material. A curved rigid support made of lightweight plastic (X-lite splint, AOA/Kirschner Medical) was placed beneath the tail to allow adequate blood flow. The hooks were connected by a wire to a swivel apparatus at the top of the cage, and the hindlimbs were elevated so there was no contact with supportive surfaces. Rats were maintained in a suspension angle of approximately 30–35°. A small thoracic cast applied to reduce lordosis and help prevent the rats from reaching the tail apparatus. CC rats had the thoracic cast applied (n = 10) but were maintained in a normal cage environment. Animals remained in the HU or CC conditions for a total of 14 days. This time period was chosen because 1) Spacelab Life Sciences missions have been of similar duration, 2) humans subjected to bed rest exhibit stable changes within this time period (9), and 3) this duration of hindlimb unloading provides stable changes in muscle weight and strength in rats (7, 14, 19, 23). Body weights were recorded before and after the control or hindlimb unloading period. During the unloading protocol, the rats were closely monitored several times daily for adequate food and water intake, grooming behavior, and urination and defecation. All rats were housed individually in a temperature-controlled (69–72°F) atmosphere with a 12:12-h light-dark cycle and given rat chow (Purina 5008 rodent chow) and water ad libitum. On the 13th day of the protocol, animals were removed from unloading or normal cage activity conditions to undergo surgical procedures. HU or CC animals were returned to the suspension apparatus or to their cage, respectively, as soon as they had adequately recovered from anesthesia (<1 h). Recovery from anesthesia was determined when the animal was alert and responsive.

Surgical Procedures

Surgical procedures for the placement of femoral catheters and lumbar sympathetic recording electrodes were conducted under halothane anesthesia, using aseptic surgical technique. Polylethylene (PE 10 fused to PE 50) catheters were inserted into the aorta and abdominal vena cava via the left femoral artery and vein for measurement of arterial pressure and administration of vasoactive drugs, respectively.

With the use of a midline abdominal incision, a branch of the lumbar sympathetic chain was dissected free. A bipolar Teflon-insulated silver wire electrode (Medwire, 0.005-in. diameter, 36 gauge) threaded through Silastic tubing (0.25-in. ID) was placed around the isolated nerve. The nerve-electrode complex was covered with a polyvinylsiloxane gel (Coltene President, Mahwah, NJ), which was allowed to harden before closure. A ground wire was sewn to the abdominal wall, and incision sites were sutured closed. Both the catheters and the lumbar sympathetic recording electrodes were tunneled subcutaneously and exteriorized at the dorsal cervical region. Catheters were filled with 10 U/ml of heparinized saline and capped with an airtight plug until the experiment. Animals were given subcutaneous fluids and butorphanol (Stadol, 3 mg/kg) for postoperative analgesia. After immediate recovery from anesthesia, animals were returned to their cages for 24 h.

Experimental Procedures

Unrestrained animals were placed in an experimental cage that was in a Faraday cage to help reduce electrical noise. The arterial catheter was connected to a pressure transducer for recording of arterial pressure. Mean arterial pressure (MAP) was derived electronically using a low-pass filter. HR was determined by measuring the number of heart beats triggered from the arterial pressure pulse. Lumbar sympathetic nerve activity (LSNA) was amplified 2,000 times using a Grass preamplifier (PS11) and filtered using a high-pass frequency level of 100 Hz and a low-pass frequency level of 3 kHz. Action potentials were monitored using a Tektronix oscilloscope and a custom-made audio monitor (Bioengineering, Univ. of Iowa). Nerve activity was rectified and integrated using an RMS converter with a time constant of 28 ms. The rectified, integrated signal was then electronically averaged, and this mean signal was used as the relative measure of LSNA. Background noise was determined at the end of the experiment when sympathetic nerve activity was eliminated by an intravenous bolus dose of the ganglionic-blocking agent chlorisondamine (5 mg/kg). This level of background noise was then subtracted from the recorded LSNA before data manipulations. Data signals were gathered and analyzed with a PowerLab (ADInstruments) data-acquisition system.
Baroreflex Assessment Protocol

Baseline hemodynamic parameters were recorded for 20–40 min before experimental manipulations to ensure stabilization of MAP, HR, and LSNA. After the collection of baseline parameters, arterial baroreflex curves were generated by producing ramp changes in arterial pressure over approximately 2–3 min. Initially, MAP was increased to 170–180 mmHg by infusion of the α1-adrenergic receptor agonist phenylephrine (PE) at increasing rates (2–25 μg·kg⁻¹·min⁻¹). MAP, HR, and LSNA were allowed to return within 10% of baseline values before proceeding with the experimental protocol. Arterial pressure was then decreased to 50–60 mmHg by infusion of the vasodilator sodium nitroprusside (NTP) at sequentially increasing rates (10–100 μg·kg⁻¹·min⁻¹). The rate of change of arterial pressure was held constant by observing the recorded pressure change and varying the rate of infusion to produce a smooth ramp change in pressure. Care was taken to keep the rate of change of arterial pressure similar in all animals at approximately 1–2 mmHg/s. Volumes infused did not exceed 100 μl. Baroreceptors were always activated first (PE infusion) before unloading (NTP infusion), to minimize any potential effects of reflexly released humoral agents, such as vasopressin or ANG II, on baroreflex function.

Data Analysis

Lumbar sympathetic nerve responses were expressed as a percentage of baseline or control LSNA before experimental interventions. Baseline or control LSNA was considered to be 100%. This analysis allows for direct evaluation of the animal’s ability to reflexly increase or decrease LSNA relative to its basal level.

For baroreflex analysis, HR and LSNA were determined at different levels of MAP during PE and NTP infusions. Data relating changes in HR or LSNA to MAP were fit to a sigmoid logistic function (15) using a standard software package (Sigma Plot, Jandel Scientific, San Rafael, CA). The equation used for this mathematical model is

\[
\text{LSNA or HR} = \left( \frac{P_1 - P_3}{1 + \exp\left[\frac{P_3(MAP - P_1)}{P_2}\right]}\right) + P_3
\]

Parameters (P₁–P₄), which are used to describe basic baroreflex function, were generated from data fit to the logistic function. These parameters were 1) the maximum LSNA or HR during decreases in arterial pressure (P₁), 2) the coefficient used to calculate the gain as a function of pressure (P₂), 3) the inflection point or MAP at the midpoint of the curve (P₃), and 4) the minimum LSNA or HR at an increased arterial pressure (P₄). In addition, the gain (G) of the reflex at each level of arterial pressure was calculated for the entire baroreflex curve using the following equation

\[
G_{\text{MAP}} = \frac{(P_1 - P_3)P_2\exp(P_2(G_{\text{MAP}} - P_3)\left[1 + \exp(P_2(G_{\text{MAP}} - P_3))\right]^2}{(P_1 - P_3)^2\exp(P_2(G_{\text{MAP}} - P_3)\left[1 + \exp(P_2(G_{\text{MAP}} - P_3))\right]^2}
\]

For each individual animal’s fit curve, the four parameters (P₁–P₄) and gain were derived. These parameters and the gain of the baroreflex curve were averaged within a group. In evaluating the time course of Flu treatment, the average parameters were statistically compared between groups using one-way ANOVA. Statistical comparison of baroreflex parameters after Flu or Veh treatment in CC and HU rats was performed using two-way ANOVA. Body weight before and after the CC or hindlimb unloading procedures was evaluated using two-way ANOVA for repeated measures. The mean parameters were used to generate an average baroreflex curve for each group.

RESULTS

Time Course of Flu Pilot Study

Data from the pilot study examining the time course of effects after Flu treatment are presented in Table 1. While there were no significant differences in resting hemodynamic parameters among the four treatment groups, body weight was significantly lower in animals receiving a 16-day treatment regimen of Flu.

Baroreflex Parameters

There were no changes in baroreflex control of HR among any of the three treatment groups compared with the Veh-treated rats as evidenced by no significant differences in any of the parameters describing the baroreflex curves (Table 1). While acute (Flu-D1) and chronic (Flu-D16) Flu treatment did not alter baroreflex control of LSNA, short-term treatment of Flu (Flu-D4) elicited a significant enhancement in baroreflex control of LSNA as evidenced by a significantly greater maximal baroreflex parameter compared with the Veh group (Table 1). Although maximal gain was not significantly enhanced in this group, a definite trend is evident (P < 0.07).

Flu Treatment in HU Rats

Baseline parameters. Baseline hemodynamic parameters as well as body weights and soleus muscle weights among the four groups are shown in Table 2. Although there were no significant differences in resting MAP among the groups, resting HR was significantly elevated in both groups of HU rats. Both HU-Flu and HU-Veh groups demonstrated a significant reduction in soleus muscle weight and soleus-to-body weight ratio compared with rats in the CC-Veh and CC-Flu groups. Both CC groups experienced a significant increase in body weight throughout the 14-day protocol.

Baroreflex control of HR. Baroreflex control of HR after short-term treatment with Flu in HU rats is illustrated in Fig. 1. Neither hindlimb unloading nor short-term Flu altered the HR response to changes in MAP. This is further evidenced by no significant differences among any of the baroreflex parameters for HR in the four groups shown in Table 3.

Table 1. Baseline and baroreflex parameters after 1-, 4-, and 16-day fluoxetine treatment in rats

<table>
<thead>
<tr>
<th></th>
<th>MAP, mmHg</th>
<th>HR, beats/min</th>
<th>Body Weight, g</th>
<th>Maximum, beats/min</th>
<th>Midpoint, mmHg</th>
<th>Minimum, beats/min</th>
<th>Peak gain, beats/min⁻¹·mmHg⁻¹</th>
<th>%LSNA</th>
<th>Maximum, mmHg</th>
<th>Midpoint, mmHg</th>
<th>Minimum, mmHg</th>
<th>%LSNA</th>
<th>Peak gain, mmHg</th>
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<tbody>
<tr>
<td></td>
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<td></td>
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<tr>
<td>Veh</td>
<td>3</td>
<td>112±3</td>
<td>353±9</td>
<td>358±14</td>
<td>529±14</td>
<td>105±7</td>
<td>246±22</td>
<td>−2.5±0.1</td>
<td>320±7.7</td>
<td>99±5.6</td>
<td>52±16</td>
<td>−61±1.2</td>
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</tr>
<tr>
<td>Flu-D1</td>
<td>3</td>
<td>117±2</td>
<td>344±37</td>
<td>340±20</td>
<td>492±31</td>
<td>121±7</td>
<td>246±21</td>
<td>−3.1±0.3</td>
<td>310±15</td>
<td>108±13</td>
<td>43±6.8</td>
<td>−61±0.9</td>
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<tr>
<td>Flu-D4</td>
<td>3</td>
<td>111±3</td>
<td>357±13</td>
<td>335±8</td>
<td>460±37</td>
<td>121±6</td>
<td>230±23</td>
<td>−3.3±0.2</td>
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<td>113±4</td>
<td>367±20</td>
<td>298±13*</td>
<td>468±17</td>
<td>125±2</td>
<td>226±21</td>
<td>−2.4±0.3</td>
<td>310±7.8</td>
<td>99±1.8</td>
<td>28±6.7</td>
<td>−5.5±1.5</td>
<td></td>
</tr>
</tbody>
</table>

Values are means ± SE. Flu, fluoxetine; D1, 1-day treatment; D4, 4-day treatment; D16, 16-day treatment; MAP, mean arterial pressure; HR, heart rate; LSNA, lumbar sympathetic nerve activity. *P < 0.05 vs. vehicle (Veh).
crease in MAP compared with CC-Veh rats. In addition, the
mediated increases in LSNA in response to a de-
fl
LSNA after short-term treatment with Flu in HU rats are
control of sympathetic nervous system activity,
fl
barore
fl
ex control of HR by either hindlimb unloading or Flu treatment.

Data on the effects of the time course of Flu treatment on
sympathetic baroreceptor responses indicate that while acute (1
day) and chronic (16 day) Flu treatment did not affect barore-
ex control of LSNA, an intermediate short-term treatment (4
days) resulted in a significant enhancement in baroreflex con-
fl
in a marked reduction of the
nding that prolonged and not short-term SSRI
fi
ring activity of dorsal raphe
R587
Table 2. Baseline parameters after fluoxetine treatment in hindlimb unloaded rats

<table>
<thead>
<tr>
<th></th>
<th>MAP, mmHg</th>
<th>HR, beats/</th>
<th>Body Weight, g</th>
<th>Soleus Weight, mg</th>
<th>Soleus/Body Wt, mg/g</th>
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</thead>
<tbody>
<tr>
<td>n</td>
<td></td>
<td>min</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>CC-Veh</td>
<td>5</td>
<td>116±2</td>
<td>376±7</td>
<td>313±8</td>
<td>140±5</td>
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<td>HU-Veh</td>
<td>6</td>
<td>124±1</td>
<td>433±57</td>
<td>327±6</td>
<td>230±7</td>
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<tr>
<td>CC-Flu</td>
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<td>119±2</td>
<td>369±8</td>
<td>317±13</td>
<td>116±4</td>
</tr>
<tr>
<td>HU-Flu</td>
<td>5</td>
<td>119±2</td>
<td>397±8†</td>
<td>323±9</td>
<td>297±7</td>
</tr>
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</table>

Values are means ± SE. HU, hindlimb unloaded. *P < 0.05 vs. D1; †P < 0.05 vs. casted control (CC).

Baroreflex control of LSNA. Baroreflex-mediated changes in
LSNA after short-term treatment with Flu in HU rats are
illustrated in Fig. 2. HU-Veh rats demonstrated attenuation of
baroreflex mediated increases in LSNA in response to a de-
crease in MAP compared with CC-Veh rats. In addition, the
maximal parameter for baroreflex control of LSNA and the
maximal gain was significantly lower in HU-Veh rats com-
pared with that of CC-Veh rats (Table 3). There was a main
group effect for a significantly lower minimal parameter for
HU rats compared with CC rats in their respective drug
treatment group. This effect was present for maximal gain as
well (Table 3). Confirming results from the pilot study (Table
1), short-term Flu treatment (i.e., 4 days) in CC-Flu rats
resulted in a significant enhancement in maximal LSNA
baroreflex responses compared with CC-Veh rats (Fig. 2, Table
3). Data further indicate that this short-term Flu treatment
resulted in a normalization of the attenuation in baroreflex
control of LSNA in HU rats (Fig. 2). This is evidenced by there
being no significant difference between the baroreflex param-
eters for LSNA in HU-Flu rats compared with CC-Veh rats as
shown in Fig. 2 and Table 3.

DISCUSSION

The results of the present study indicate that short-term

treatment with the SSRI Flu in rats results in an enhancement in
baroreflex control of sympathetic nervous system activity,
and this effect serves to correct the experimentally induced
attenuation in baroreflex function in the HU rat model of
cardiovascular deconditioning.

Fig. 1. Mean baroreflex curves describing reflex control of heart rate (HR) after either vehicle (Veh) or short-term fluoxetine (Flu) treatment in casted control (CC) and hindlimb unloaded (HU) rats. There was no significant effect on baroreflex control of HR by either hindlimb unloading or Flu treatment.

Considering that it is necessary to administer Flu chronically
to alleviate the psychological symptoms associated with de-
pression, we originally hypothesized that only chronic (i.e., 16
day) treatment with Flu would result in an enhancement in
baroreflex control of sympathetic nervous system activity.
However, data indicate that only a 4-day treatment with Flu
produced a significant enhancement in baroreflex function
(Fig. 2, Tables 1 and 3). Although these results are interesting
and somewhat surprising, there are several possibilities that
might explain these effects. First, it is possible that while
behavioral effects require chronic SSRI treatment, cardiovas-
cular effects can be elicited in response to a short-term treat-
ment regimen. Studies investigating the cardiovascular benefits
of SSRI treatment in humans have primarily evaluated these
responses after chronic treatment (5, 11, 27, 29, 30). However,
recently Shores et al. (32) cited a reduction in plasma NE
appearance rates after short-term treatment with the SSRI
sertraline in healthy subjects. The effects after chronic treat-
ment were not evaluated in this (32) study.

It is also possible that the enhancement in sympathetic
baroreflex function is due to the specific adaptations in neuro-
transmission that occur at the short-term time point of admin-
istration. Data indicate that short-term SSRI treatment results
in a marked reduction of the firing activity of dorsal raphe
serotonergic neurons that is partially recovered by 7 days of
treatment and completely recovered by 14 days (6). This is in
contrast to the finding that prolonged and not short-term SSRI
administration results in a marked decrease in locus ceruleus
NE neuronal activity (34). This imbalance in raphe 5-HT
neuronal activity vs. locus ceruleus NE neuronal activity has
been cited as a possible reason why anxiety and panic disorder
symptoms are often exaggerated after short-term SSRI treat-
ment and then diminish altogether after chronic treatment (34).
Considering these data, it is tempting to speculate that perhaps
short-term SSRI treatment results in an overactivation of sym-
pathetic reflexes due to this imbalance in 5-HT/NE neuronal
firing. This would explain both the augmentation in anxiety
and panic symptoms and the transient enhancement in barore-
excontrol of LSNA after short-term SSRI treatment. None-
Table 3. Curve parameters describing baroreflex control of HR and LSNA after 4-day fluoxetine treatment in hindlimb unloaded rats

<table>
<thead>
<tr>
<th>HR Baroreflex Parameters</th>
<th>LSNA Baroreflex Parameters</th>
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<tr>
<td></td>
<td>Maximum, %LSNA</td>
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<td>Minimum, %LSNA</td>
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<td>Peak gain, beats/min mmHg^{-1}</td>
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<table>
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<tr>
<th></th>
<th>n</th>
<th>Maximum, beats/min</th>
<th>Midpoint, mmHg</th>
<th>Minimum, beats/min</th>
<th>Midpoint, mmHg</th>
<th>Peak gain, beats/min mmHg^{-1}</th>
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<tr>
<td>CC-Veh</td>
<td>5</td>
<td>500±12</td>
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<td>221±27</td>
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<td>-0.5±0.4</td>
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<td>HU-Veh</td>
<td>6</td>
<td>516±4</td>
<td>137±2</td>
<td>280±9</td>
<td>-4.0±0.4</td>
<td>-0.3±0.2</td>
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<tr>
<td>CC-Flu</td>
<td>5</td>
<td>512±2</td>
<td>122±4</td>
<td>276±9</td>
<td>-3.4±0.4</td>
<td>-0.3±0.2</td>
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<tr>
<td>HU-Flu</td>
<td>5</td>
<td>480±11</td>
<td>117±4</td>
<td>286±7</td>
<td>-2.7±0.2</td>
<td>-0.3±0.2</td>
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<td>CC-Veh</td>
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<td>107±3</td>
<td>17±7</td>
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<td>HU-Veh</td>
<td>6</td>
<td>117±4</td>
<td>-9±4*</td>
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<td>CC-Flu</td>
<td>5</td>
<td>110±3</td>
<td>23±10</td>
<td>12.6±1.3</td>
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<td>HU-Flu</td>
<td>5</td>
<td>106±4</td>
<td>-14±2*</td>
<td>-6.0±0.6*</td>
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Values are means ± SE. *P < 0.05 vs. CC; †P < 0.05 vs. Veh in respective group.

theless, it is interesting why enhancement in sympathetic baroreflex responses does not persist after chronic treatment.

A third possibility for this effect is that the plasticity and adaptive ability of the arterial baroreflex prevents such an enhancement in baroreflex control of sympathetic nervous system activity to persist in an animal with normal baroreflex function. This would prevent any deleterious cardiovascular effects such as hypertension from developing. However, clinical data indicate that overdosage of SSRIs often produces hypertension (26).

Because group sizes were small in our pilot study in which we studied the time course of SSRI treatment effects on baroreflex function, we felt that it was important to systematically replicate the 4-day data demonstrating the enhancement in baroreflex control of LSNA. We did this in conjunction with the hindlimb unloading study. The results indicate that the baroreflex control of LSNA is enhanced at 4 days in both CC and HU rats receiving Flu compared with their respective Veh treatment groups. Although the effect of 4 days of Flu treatment in the HU-Flu group (vs. their respective control or HU-Veh) was similar to the Flu-D4 group (vs. their respective control or Veh), it would not be appropriate to combine the data from the two studies since control animals in the hindlimb unloading study received a thoracic cast, while those in the time course (pilot) study did not. Nevertheless, the systematic replication of the findings of the two studies increases the degree of confidence in the reliability of the finding that 4 days of treatment with Flu enhances baroreflex control of LSNA. In addition, the fact that this effect helped normalize attenuated baroreflex control of LSNA after HU adds additional support for this conclusion.

Baroreflex data in untreated HU and CC rats are very similar to previous reports (12, 20) and have been essentially replicated in the present study. Comparable to the previous results, HU rats exhibited no significant alterations in baroreflex control of HR (Fig. 1; Table 3) but had a significant attenuation in baroreflex-mediated increases in LSNA compared with control rats (Fig. 2, Table 3). This attenuation in LSNA reflex responses was essentially offset through short-term Flu treatment as there were no significant differences in baroreflex control of LSNA in HU-Flu compared with CC-Veh rats (Fig. 2, Table 3). Because CC-Flu rats also demonstrated a significant enhancement in baroreflex control of LSNA similar to data shown in Fig. 2 and Table 3, there was no significant statistical interaction between the factors of drug treatment and deconditioning status. This may argue against any underlying functional change in the serotonergic system accompanying cardiovascular deconditioning as a mechanism accounting for the attenuation in baroreflex control of LSNA. However, to more conclusively eliminate a role of the serotonergic system controlling baroreflex function in HU rats, further investigation is necessary.

Data in humans indicate that individuals with orthostatic hypotension that are refractory to other traditional forms of therapy are often responsive to SSRI treatment (11, 29). The HU rat model of cardiovascular deconditioning is an animal model commonly used to study mechanisms responsible for the orthostatic intolerance commonly experienced after bed rest and spaceflight. Considering that baroreflex-mediated changes in sympathetic nervous system activity are of primary importance when compensating for changes in arterial blood pressure during an orthostatic challenge (3, 28), we wanted to determine if SSRI treatment may enhance baroreflex function and thus improve orthostatic intolerance. Results indicate that short-term Flu administration resulted in a significant enhancement in baroreflex control of LSNA as well as a correction in attenuated baroreflex function in HU rats (Fig. 2, Tables 1 and 3). Thus enhancement in baroreflex control of sympathetic nervous system activity is a possible mechanism responsible for the successful treatment of orthostatic intolerance in humans after SSRI administration.

The specific mechanism by which Flu treatment works to restore attenuated baroreflex function is not clear. It is possible
that the attenuation in baroreflex gain after hindlimb unloading in rats is due to autonomic imbalance and short-term SSRI treatment helps reduce sympathetic tone and thus restore autonomic balance and improve baroreflex function. This possibility is supported by several observations. First, previous data indicate that there is a high degree of correlation between a reduction in autonomic balance as measured through a decrease in HR variability and the attenuation of baroreflex gain (17). Second, hindlimb unloading in rats is associated with a resting tachycardia and reduction in the sympathetic baroreflex gain as documented in the current and previous studies (12, 20, 22). Data from the current study indicate that Flu normalized the attenuated sympathetic baroreflex gain (Fig. 2) and partially normalized the resting tachycardia associated with hindlimb unloading (Table 2). Previous studies indicate that SSRI treatment may restore autonomic imbalance after long-term treatment in posttraumatic stress disorder patients (5). Plasma NE appearance rates are reduced after short-term Flu treatment in healthy humans (32). These findings taken together suggest an SSRI-mediated restoration in autonomic balance may be a mechanism responsible for normalization of attenuated sympathetic baroreflex function after hindlimb unloading.

Individuals with a history of psychological depression are at a much greater risk for developing cardiovascular disease than patients with no prior history of depressive disorders (24). Major depression essentially doubles the risk that patients with newly diagnosed coronary artery disease will experience an adverse cardiovascular event within a year after the diagnosis (4). Increased sympathetic nervous system activity has often been cited as a mechanism in the link between psychological depression and cardiovascular disease (24). Recent data indicate that in rats exposed to the chronic mild stress rodent model of depression there was augmented cardiac sympathetic tone (10). Data from the current study suggest that SSRI administration reduces the basal sympathetic tone to stabilize or normalize baroreflex control of sympathetic nervous activity. Data in humans indicate that this could be a possibility since Flu administration in depressed patients with heart disease resulted in a significant decrease in resting HR (27). In addition SSRI treatment in posttraumatic stress disorder patients normalizes the reduction in HR variability, indicating a possible reduction in an elevated basal sympathetic tone (5).

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