Exercise training enhances baroreflex control of heart rate by a vagal mechanism in rabbits with heart failure

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Received 16 January 2002; accepted in final form 4 February 2002

Exercise training enhances baroreflex control of heart rate by a vagal mechanism in rabbits with heart failure. J Appl Physiol 92: 2403–2408, 2002; 10.1152/japplphysiol.00039.2002.—Moderate exercise training (Ex) enhances work capacity and quality of life in patients with chronic heart failure (CHF). We investigated the autonomic components of resting heart rate (HR) and the baroreflex control of HR in conscious, instrumented rabbits with pacing-induced CHF after Ex. Sham and CHF rabbits were exercise trained for 4 wk at 15–18 m/min, 6 days/wk. Arterial pressure and HR were recorded before and after metoprolol (1 mg/kg iv) or after atropine (0.2 mg/kg iv). Mean arterial pressure was altered by infusions of sodium nitroprusside and phenylephrine. The data were fit to a sigmoid (logistic) function. Baseline HRs were 266.5 ± 8.4 and 232.1 ± 1.6 beats/min in CHF and CHF Ex rabbits, respectively (P < 0.05). In the unblocked state, CHF rabbits had a significantly depressed peak baroreflex slope (1.7 ± 0.3 vs. 5.6 ± 0.7 beats·min⁻¹·mmHg⁻¹; P < 0.001) and HR range (128.6 ± 34.5 vs. 253.2 ± 20.3 beats/min; P < 0.05) compared with normal subjects. Ex increased baroreflex slope to 4.9 ± 0.3 from 1.7 ± 0.3 beats·min⁻¹·mmHg⁻¹ in unblocked rabbits (P < 0.001 compared with CHF non-Ex). Ex did not alter baroreflex function in sham animals. After metoprolol, baroreflex slope was significantly increased in CHF Ex rabbits (1.5 ± 0.2 vs. 3.0 ± 0.2 beats·min⁻¹·mmHg⁻¹; P < 0.05). After atropine, there was no significant change in baroreflex slope or HR range between CHF Ex and CHF rabbits. These data support the view that enhancement of baroreflex control of HR after Ex is due to an augmentation of vagal tone.

A large amount of data exists on cardiac autonomic regulation after Ex in normal humans and animals. Much less is known concerning these adjustments in the CHF state. It is widely accepted that Ex results in a reduction in resting cardiac sympathetic tone and an increase in vagal tone (9, 11, 36). If this scenario existed in the CHF state, it would be beneficial to patients, because it should decrease resting heart rate (HR) and myocardial oxygen consumption. In addition, augmentation of the arterial baroreflex control of HR will enhance blood pressure regulation and may protect against arrhythmic episodes, such as sudden death in the CHF state.

The aim of the present study was to determine the changes in cardiac vagal and sympathetic tone in an animal model of CHF after an Ex program. We hypothesized that Ex in CHF would result in a decrease in sympathetic tone, an increase in vagal tone, and an augmentation in BRS.

MATERIALS AND METHODS

Experiments were carried out in 26 male New Zealand White rabbits weighing between 2.5 and 3.5 kg. All experiments were reviewed and approved by the University of Nebraska Medical Center Institutional Animal Care and Use Committee. All experiments conformed to the guidelines for care and use of laboratory animals of the American Physiological Society and the National Institutes of Health.

Surgical instrumentation. Rabbits were instrumented as described earlier (23). In brief, with sterile technique, a left thoracotomy was performed through the fourth intercostal space. After the pericardium was opened, a pair of 2-mm

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piezoelectric crystals (Sonometerics) was sutured across the left ventricle from anterior to posterior near the base of the ventricle. A platinum wire pacing electrode of our own design was secured to the apical surface of the left ventricle. All wires were tunneled beneath the skin and exited in the midscapular area. The animals were treated with antibiotics (Baytril, 2.3 mg/kg im twice per day for 3 postoperative days) and allowed to recover for ~2 wk before being used in any experiment. A few days before the experiment, a Tygon catheter was implanted into the left carotid artery and jugular vein to record arterial and central venous pressures (CVPs) and to administer drugs during the experiment. Catheters were filled with heparin (1,000 U/ml) and sealed until the day of the experiment.

Induction of heart failure. After recovery from surgery, the heart failure groups were paced at rates between 320 and 340 beats/min by using a small, light-weight pacing unit of our own design. The pacing rate was then adjusted and monitored by the cardiac dimension tracings. In general, each rabbit was paced at 320 beats/min for the first week to determine whether it would tolerate this protocol. After the first week, the pacing rate was increased to 340 beats/min and left at this rate for the remainder of the protocol. The rabbits were continually paced for ~4 wk (CHF Ex and CHF non-Ex groups), at which time a second surgery was performed to implant the vascular catheters. Recordings of cardiac dimension and HR were taken twice per week to monitor the degree of cardiac dilation during the pacing protocol. The rabbits were placed into a Plexiglas box to limit movement, and the pacemaker was turned off for 30 min before any data were recorded.

Ex protocol. Rabbits were trained to run on a motor-driven exercise wheel of our own design. As described previously (23), the rabbits were acclimated to the wheel before the initial surgery. After the recovery from surgery, rabbits were exercised for a total of 40 min/day, 6 days/wk. A warm-up period of 5 min at 5 m/min was followed by peak exercise (15–18 m/min) for 30 min. This was followed by a cool down of an additional 5 min at 5 m/min.

Experimental protocol. Four groups of animals were studied. Group 1 (n = 6) was a sham (nonpaced) group that was instrumented in an identical fashion to all other groups and was not exercise trained. Group 2 (n = 7) was a sham group that was exercise trained. Group 3 (n = 6) was a CHF group that was not exercise trained. Finally, group 4 (n = 6) was a CHF group that was exercise trained. On the day of the experiment, the rabbit was placed in the Plexiglas box, and the pacemaker was turned off for 30 min. Baseline recordings of arterial pressure, cardiac diameter, and HR were taken for several minutes. All hemodynamic measurements were recorded on a computer with the Powerlab data-acquisition system and software (AD Instruments, Milford, MA). The baseline data for HR, arterial pressure, and CVP are similar to those reported by our laboratory in a previous study (23), because many of the animals studied in the baseline state were included in that study.

Arterial baroreflex control of HR was determined in response to an injection of sodium nitroprusside (100 μg/kg) and phenylephrine (PE; 30 μg/kg iv). Injections were made in random order and were given in the same volume to each rabbit. After the control curve, a second curve was constructed ~10 min after administration of 0.2 mg/kg iv of atropine methylbromide. A new set of baseline recordings was taken before the baroreflex curve was constructed after atropine. On another day, the same procedure was carried out, but the control curve was followed by another curve constructed 10 min after administration of 1 mg/kg iv of metoprolol bitartrate. These doses blocked the chronotropic effect of a 1 μg/kg dose of acetylcholine and a 0.3 μg/kg dose of isoproterenol, respectively. Atropine and metoprolol studies were randomized as to which was done first.

Arterial baroreflex. Arterial baroreflex curves were constructed as previously described by our laboratory (23, 24). In brief, several points for HR were taken during the fall or rise in arterial pressure after the administration of sodium nitroprusside and PE, respectively. Data points were obtained at ~2-s intervals. The logistic regression curve, as described by Kent et al. (21), was fit to the data points by using the following equation

$$HR = A/(1 + \exp[B(MAP - C)]) + D$$  (1)

where A is HR range, B is the slope coefficient, C is the pressure at the midpoint of the range, D is minimum HR (21), and MAP is mean arterial pressure. The peak slope (or maximum gain) was determined by taking the first derivative of the baroreflex curve as described by

$$\text{Slope} = (A \times B \times \exp[B(MAP - C)])^2 / ((1 + \exp[B(MAP - C)])^2)$$  (2)

The mean value of each curve parameter was used to derive a composite curve for each group of rabbits before and after each intervention.

Statistical analysis. Data are expressed as means ± SE. The differences between groups were determined by a one-way ANOVA with the Newman-Keuls test used for post hoc analysis when comparing baseline hemodynamic data. A two-way ANOVA was used when comparing group and drug effects. A P value of <0.05 was considered statistically significant.

RESULTS

Table 1 contains the baseline hemodynamic values in the four groups of rabbits. Although there were no significant differences in MAP, resting HR was significantly higher in CHF rabbits compared with the sham group. Ex significantly lowered HR in the CHF group. However, Ex did not significantly lower HR in the sham group, even though the average decrease in this group was ~16 beats/min. Compared with sham animals, the CVP was significantly increased in both CHF groups.

To further document the CHF state of groups 3 and 4 and to show that Ex did not improve myocardial function in CHF, the data in Fig. 1 illustrate that chronic pacing increased both end-diastolic and end-systolic diameter and reduced the velocity of shorten-

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<th>Table 1. Baseline hemodynamics in sham, CHF, sham exercise trained, and CHF exercise trained rabbits</th>
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Values are means ± SE. MAP, mean arterial pressure; HR, heart rate; CVP, central venous pressure; CHF, chronic heart failure; Ex, exercise trained. Data include some animals from a previous study (23). * P < 0.05 compared with normal; † P < 0.05 compared with CHF.
ing. The cardiac dilation and reduction in shortening were not altered by Ex in CHF. Furthermore, Ex had no effect on cardiac dimensions in the sham animals.

Figure 2 illustrates a recording of hemodynamics in a CHF rabbit and in a CHF Ex rabbit. The response to PE administration is shown. PE evoked an increase in arterial pressure in both rabbits; however, a more profound bradycardia was evoked in the CHF Ex rabbit. Figure 3 shows the mean resting HR changes in each group after either atropine or metoprolol administration. As shown, the decrease in HR after metoprolol was not different among any of the groups. However, atropine significantly increased HR in both of the Ex groups.

Arterial baroreflex curves were constructed on each rabbit before and after blockade of either sympathetic or vagal efferent pathways. Figure 4 shows composite baroreflex curves before any autonomic blockade. BRS was significantly reduced in CHF rabbits compared with any of the other groups (1.7 ± 0.3 vs. 5.6 ± 0.7 beats·min⁻¹·mmHg⁻¹ in CHF and normal groups, respectively; *P < 0.05). BRS did not differ between sham Ex and sham groups. The improvement in baroreflex function (BRS and HR range) in the CHF Ex group (BRS = 4.9 ± 0.3 beats·min⁻¹·mmHg⁻¹) was largely due to an enhancement in the minimum HR after administration of PE (minimum HR: normal, non-Ex 115.4 ± 12.1 beats/min; normal Ex 105.2 ± 5.2 beats/min; CHF non-Ex 216.8 ± 8.6 beats/min, CHF Ex 152.7 ± 19.5 beats/min; *P < 0.05).

Figure 5 shows the corresponding baroreflex HR range in each group of rabbits in the unblocked state and after administration of either metoprolol or atropine. Compared with the sham group, HR range was significantly lower in the unblocked state and after metoprolol or atropine in the CHF group. HR range did not differ in the sham Ex group compared with the sham group in the unblocked state or after either metoprolol or atropine. However, Ex increased HR range in the CHF group in the unblocked state and after administration of metoprolol, whereas there was no augmentation after administration of atropine in this group. Figure 6 shows the mean data for the peak baroreflex slopes in the four groups of rabbits. The peak slope was significantly decreased in the CHF non-Ex group in all three states. Ex had no effect on

Fig. 1. Cardiac dimensions [end diastolic (ED; A) and end systolic (ES; B)] and the velocity of cardiac shortening [dD/dtmax, where D is minimum heart rate (HR); C] for all 4 groups of rabbits. Despite exercise training (Ex), chronic heart failure (CHF) rabbits still exhibited significant cardiac dilation and a reduction in dD/dtmax. Values are means ± SE. Pre, before experiment; Post, after experiment. *P < 0.05 compared with the prepared state. Some baseline data were previously published (23).

Fig. 2. Original hemodynamic recordings from a CHF rabbit (A) and a CHF Ex rabbit (B). Arrow, injection of phenylephrine (10 μg/kg) was given intravenously. MAP, mean arterial pressure; bpm, beats/min.

Fig. 3. The percent change (delta) in resting HR in each group of rabbits after autonomic blockade with either metoprolol or atropine. Ex enhanced the atropine responses but had no effect on the metoprolol responses. Values are means ± SE. *P < 0.05 compared with the respective non-Ex group.

Fig. 4. Original baroreflex HR range in each group of rabbits in the unblocked state and after administration of either metoprolol or atropine. Compared with the sham group, HR range was significantly lower in the unblocked state and after metoprolol or atropine in the CHF group. HR range did not differ in the sham Ex group compared with the sham group in the unblocked state or after either metoprolol or atropine. However, Ex increased HR range in the CHF group in the unblocked state and after administration of metoprolol, whereas there was no augmentation after administration of atropine in this group. Figure 6 shows the mean data for the peak baroreflex slopes in the four groups of rabbits. The peak slope was significantly decreased in the CHF non-Ex group in all three states. Ex had no effect on
baroreflex slope in normal animals but increased baroreflex slope in the CHF Ex group in the unblocked state and after metoprolol but not after atropine.

DISCUSSION

The present study documents that Ex enhances arterial baroreflex function in the CHF state, confirming our laboratory’s previous study in the same model of CHF (23). The enhancement in baroreflex control of HR after Ex appears to be mediated primarily by enhancement of vagal outflow to the sinoatrial (SA) node. In addition to a significant contribution of the vagus-to-baroreflex function, there also appears to be an important contribution of the vagus to resting HR, because there were significantly greater increases in HR after atropine administration in both normal and CHF rabbits after 1 mo of Ex compared with the non-Ex groups (Fig. 3).

The lack of a sympathetic component to the enhanced baroreflex function is curious, because our laboratory has previously shown that resting renal sympathetic nerve activity (RSNA) is significantly decreased in CHF Ex rabbits (23). This observation suggests a differential control of sympathetic function to the heart vs. the peripheral circulation after Ex in CHF rabbits. Autonomic regulation of cardiovascular function after Ex is controversial. In a study carried out in normal humans who were subjected to 12 wk (3 times/wk) of treadmill or leg-cycle exercise, no change in muscle sympathetic nerve activity was noted after baroreflex testing (34). Similarly, in a study carried out in normal rabbits that were exercise trained for 8 wk at a rate significantly higher than that of the rabbits in the present study, DiCarlo and Bishop (12) showed decreases in BRS for both HR and RSNA. On the other hand, studies carried out in hypertensive rats showed improvement in baroreflex function after Ex (6, 22).

Unfortunately, the autonomic components of the HR responses were not investigated in that study. Similarly, it was found that endurance Ex in normal humans depressed the baroreflex control of HR (35). Williamson and Raven (40) showed that the carotid-cardiac baroreflex gain was not altered in Ex subjects, but there was a leftward shift of the R-wave-R-wave interval-carotid sinus pressure relationship, consistent with the decrease in resting HR in trained subjects. All of these studies were carried out in normal humans or animals. In contrast, studies by Iellamo et al. (18), which were carried out in patients with established coronary artery disease, showed increases in BRS after a 2- to 4-wk Ex regimen. Furthermore, studies by Coats et al. (8) and by Tyni-Lenne et al. (37) both point

![Fig. 4. Composite arterial baroreflex curves from each group of rabbits. MAP is plotted against HR (A) and against the slope of the baroreflex curves (B). A: *minimum HR is significantly higher ($P < 0.05$) in the CHF group compared with all other groups. B: *peak baroreflex slope is significantly less in the CHF group compared with all other groups, $P < 0.05$. Some of the animals in this figure were included in a previous study from this laboratory (23).](image)

![Fig. 5. Baroreflex HR range in each group of rabbits in the unblocked state and after metoprolol or atropine. Values are means ± SE. *Significantly ($P < 0.05$) different from the normal group. †Significantly ($P < 0.05$) different from the non-Ex group.](image)

![Fig. 6. Mean data for the maximum baroreflex slopes in each group of rabbits. Baroreflex sensitivity was depressed in CHF rabbits and was normalized in CHF Ex rabbits. Whereas there was a significant increase after metoprolol in the CHF Ex group, there was no difference after atropine administration. *Significantly different ($P < 0.05$) from the sham group. †Significantly different ($P < 0.05$) from the CHF group.](image)
to a role for Ex in reducing sympathetic tone and increasing vagal tone in patients with CHF.

Ex has become a therapeutic modality in some patients with CHF. Whereas there have been reports that Ex enhances quality of life indexes in patients with CHF (25, 39), only recently has it been shown that Ex may increase survival in this disease state (4, 29). Whereas the study by Belardinelli et al. (4) does not definitively show enhanced survival in these patients (7), it does suggest that, in a controlled environment, Ex may have a beneficial effect on morbidity and mortality in patients with CHF. The mechanism(s) for the apparent salutary effects of Ex in the CHF is not clear. Ex increases oxygen consumption in patients with CHF, minimizes ventricular wall stress (10), and improves endothelial function (3, 16, 38) and skeletal muscle metabolic function (27). Although these changes are undoubtedly important for the beneficial effects of Ex in CHF, there are additional neurohumoral changes that may be equally important (5). A previous study from this laboratory has shown that Ex improves baroreflex function, reduces resting RSNA, and decreases circulating ANG II (23). Whereas ANG II has been shown to reduce BRS (30, 32), existing data suggest that ANG II has no specific effects on the SA nodal response to vagal stimulation (26). However, ANG II has been shown to be associated with the attenuated vagal baroreflex response in rats with chronic myocardial infarctions (15) and in humans subjected to face immersion (2). Furthermore, ANG II acting in the nucleus tractus solitarii and perhaps other areas of the brain stem may influence the vagal component of the arterial baroreflex (19). In a study by Reid and Chou (31), it was shown that, in conscious rabbits, atropine reduced the baroreflex response to PE, but propanolol had little effect. These data suggest that the effects of ANG II are mediated by a vagal mechanism.

Two additional mechanisms may be of relevance in explaining the results of the present study. First, in normal rabbits, DiCarlo and co-workers (12–14) have shown that Ex enhances the input from cardiac afferents, which, in turn, depresses arterial baroreflex function. These conclusions were based on functional changes after afferent blockade. However, in a more recent study from their laboratory (33), direct recordings were made from vagal afferents of Ex and sedentary rabbits. This study showed that vagal afferent function was not altered by Ex. The differences in the effects of Ex on baroreflex function between these studies and the sham animals in the present study may be due to the fact that the rabbits in the studies of DiCarlo and co-workers were exercised to a higher level than those in our study. It was not possible to exercise the CHF rabbits at this higher level. Second, it is possible that Ex alters baroreflex function in a fundamentally different way in CHF animals compared with normal animals that have lower levels of resting neurohormones.

It is also possible that Ex upregulates SA nodal muscarinic receptor density or activity. However, a study by Negrao et al. (28) would argue against this idea. In this study, electrical stimulation of the vagus resulted in smaller bradycardic effects in Ex rats compared with sedentary rats. However, sympathetic tone was reduced in Ex rats. These authors suggested that Ex causes an intrinsic change in pacemaker cells to induce a resting bradycardia.

In summary, the results of the present study support the hypothesis that Ex enhances baroreflex control of HR in animals with CHF. This enhanced bradycardia is mediated primarily by an increased influence of the vagus nerve on HR with minimal contribution of cardiac sympathetic innervation. The mechanism(s) for this Ex effect in CHF is not clear; however, based on previous studies from this and other laboratories, it may involve alterations in central vagal outflow. It may be speculated from these data that Ex improves sympathovagal balance and, therefore, may be protective against the generation of cardiac arrhythmias in the CHF state.

The authors thank Johnnie F. Hackley and Pamela Curry for expert technical assistance. These studies were supported by National Heart, Lung, and Blood Institute Grant PO-1 HL-62222.

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