Role of the Exercise Pressor Reflex in Rats With Dilated Cardiomyopathy

Scott A. Smith, PhD; Pradeep P.A. Mammen, MD; Jere H. Mitchell, MD; Mary G. Garry, PhD

Background—In heart failure, there is a sympathetically mediated hyperkinetic cardiovascular response to exercise that limits tolerance to physical activity. Alterations in skeletal muscle morphology and metabolism have led to the hypothesis that the exercise pressor reflex (EPR) becomes hyperactive after the development of cardiomyopathy and contributes to the exaggerated circulatory response elicited.

Methods and Results—To test this hypothesis, Sprague-Dawley rats were divided into the following groups: control, sham, and dilated cardiomyopathy (DCM, induced by ischemic injury). Using transthoracic echocardiography, left ventricular fractional shortening was 47 ± 2%, 44 ± 1%, and 24 ± 2% in control, sham, and DCM rats, respectively. Activation of the EPR by electrically induced static muscle contraction resulted in significantly larger increases in mean arterial pressure and heart rate in DCM animals (32 ± 2 mm Hg, 13 ± 1 bpm) compared with control (20 ± 1 mm Hg, 8 ± 1 bpm) and sham (20 ± 2 mm Hg, 8 ± 1 bpm) rats. Comparable results were obtained with selective stimulation of the mechanically sensitive component of the EPR by passive muscle stretch. The augmentations in EPR and mechanoreflex activity in DCM occurred progressively over a 10-week period, becoming greater as the severity of left ventricular dysfunction increased.

Conclusions—In DCM, the potentiated cardiovascular response to static muscle contraction is mediated, in part, by an exaggerated EPR. The muscle mechanoreflex contributes significantly to the EPR dysfunction that develops. (Circulation. 2003;108:1126-1132.)

Key Words: afferent ■ heart failure ■ hemodynamics ■ nervous system, autonomic ■ exercise

Exercise intolerance is a hallmark feature of chronic heart failure (CHF). Muscle atrophy, decreased peripheral blood flow, fiber-type transformation, and reduced oxidative capacity are important factors limiting exercise performance in CHF.1–6 In addition, cardiovascular regulation during physical activity is clearly altered with CHF, because studies using dynamic and static forms of exercise have demonstrated augmentations in sympathetic nerve activity, vascular resistance, heart rate (HR), and blood pressure.7–15

Two distinct neural control mechanisms are activated by exercise: central command and the exercise pressor reflex (EPR). Central command is a mechanism whereby signals from a central site responsible for recruiting motor units activate cardiovascular control areas in the brain stem.16,17 The EPR is a mechanism whereby signals from skeletal muscle group III (predominately mechanically sensitive) and group IV (predominately metabolically sensitive) afferents likewise evoke increases in blood pressure and HR via coordinated changes in autonomic outflow.18–20

Because of the peripheral skeletal myopathy that develops in heart failure,4 the EPR has been implicated as a possible mechanism by which circulatory control is dysregulated in cardiomyopathic individuals, thereby contributing to exercise intolerance.21 Because the metabolic component of the reflex may be blunted in CHF,10,22 it has been hypothesized that the mechanical component of the reflex preferentially drives the EPR hyperactivity that develops.11,23

The contribution of neurally mediated peripheral mechanisms to the evolution of reduced exercise capacity in heart failure is poorly understood. The purpose of this investigation was to use a novel rat model24 to determine whether EPR-mediated changes in circulatory hemodynamics are altered with dilated cardiomyopathy (DCM) in the absence of central command. Furthermore, we sought to dissect the contribution of the skeletal muscle mechanoreflex to the manifestation of cardiovascular dysregulation within this model.

Methods

Experiments were performed in 50 age-matched male Sprague-Dawley rats (Harlan, Indianapolis, Ind). The procedures outlined were approved by the Institutional Animal Care and Research Advisory Committee of the University of Texas Southwestern Medical Center.

Received December 23, 2002; de novo received March 18, 2003; accepted May 1, 2003.

From the Departments of Internal Medicine (S.A.S., P.P.A.M., J.H.M., M.G.G.) and Physiology (J.H.M.), Harry S. Moss Heart Center, and Department of Health Care Sciences (S.A.S.), University of Texas Southwestern Medical Center, Dallas, Tex.

Correspondence to Mary G. Garry, PhD, Department of Internal Medicine, University of Texas Southwestern Medical Center, 5323 Harry Hines Blvd, Dallas, TX 75390-9174. E-mail mary.garry@utsouthwestern.edu

© 2003 American Heart Association, Inc.

Circulation is available at http://www.circulationaha.org

DOI: 10.1161/01.CIR.0000084538.40542.56
Model of Dilated Cardiomyopathy

Animals were induced with isoflurane gas anesthesia (2% to 5% in 100% oxygen), intubated, and ventilated. A thoracotomy was performed, exposing the heart, and the left anterior descending coronary artery was ligated to produce DCM. Buprenorphine (20 µg/kg) was administered for postoperative analgesia. Sham operations were performed without ligation of the coronary artery. Under isoflurane anesthesia, transthoracic echocardiography (Sonos 5500, Agilent Technologies) was performed 6 to 10 weeks after surgery. On completion of physiological experimentation, the heart was excised, weighed, post-fixed, and processed for histological examination. In addition, the lungs and tibia were removed and weighed or measured for length, respectively. Excluding thoracotomy, these procedures were also performed in age-matched control animals.

Acute Experimental Preparation

Rats were initially anesthetized with isoflurane gas and instrumented as previously described. Briefly, animals were intubated for mechanical ventilation and cannulated with jugular venous and carotid arterial catheters. A laminectomy exposing the lower lumbar portion of the spinal cord (L2 to L 6 ) was performed, and stimulating electrodes were placed around the cut peripheral ends of the L 4 and L 5 ventral rootlets. The calcaneal bone of the right hindlimb was cut, and the Achilles tendon was connected to a force transducer (FT-10, Grass Instruments). Subsequently, animals were rendered insentient by precollicular decerebration. After forebrain transection and aspiration, gas anesthesia was discontinued.

Protocol: Physiological Experimentation

Electrically induced static contraction of the triceps surae was used to activate both mechanically and metabolically sensitive skeletal muscle afferents at maximal and submaximal intensities. Using constant current stimulation (1 to 3 times motor threshold, 0.1-ms pulse duration, 40 Hz), 30-second contractions were produced by excitation of the L 4 and L 5 ventral roots with the peak arterial pressure, force development, and HR responses recorded. Preferential activation of mechanically sensitive somatosensory fibers was achieved by passively stretching the hindlimb muscles using a rack and pinion system (Harvard Apparatus). At the conclusion of each experiment, the neuromuscular blocking agent vecuronium bromide (1 mg/mL) was administered intravenously and stimulation of the ventral roots was repeated at 3, 5, and 10 times motor threshold.

Data Acquisition and Statistical Analysis

Cardiovascular and contractile force data were acquired and analyzed using hardware and software for the CED micro 1401 system (Cambridge Electronic Design). Baseline values were determined using 30 seconds of data before a given maneuver. The peak response was defined as the greatest change from baseline elicited during the 30-second execution of a contraction or stretch. When measuring blood pressure, the rate of the response was defined as the change in mean arterial pressure (MAP) over time from the initiation of the response to the peak of the response. On all data sets, statistics were performed using linear regression analysis, Student t test, or ANOVA, with Student Newman-Keuls or Dunnett post hoc test used as appropriate.

Results

Characterization of the Model of Cardiomyopathy

Significant changes in cardiac and skeletal muscle morphology, histology, and function resulted from the induction of ischemic injury. Morphometric data for control, sham, and DCM rats are presented in Table 1. Body mass of DCM rats was not statistically different from that of sham or control animals. Ratios of heart mass to body mass and heart mass to tibial length were significantly greater in DCM animals. In addition, ratios of lung mass to body mass were larger in DCM than in sham rats.

Coronary artery ligation caused an increase in cardiac fibrosis and a corresponding loss of myocardium in the direct region of ligation as well as outside the zone of ligation (Figure 1A). Left ventricular chamber diameter was increased 2-fold in ligated hearts compared with normal and sham hearts. Dilation of the right ventricle was also evident. Using transthoracic echocardiography (Figures 1B and 1C), DCM rats exhibited significant left ventricular systolic dysfunction as characterized by marked reductions in percent fractional shortening commensurate to elevations in left ventricular end systolic (LVESD) and left ventricular end diastolic (LVEDD) dimensions. Only animals exhibiting a greater than 25% reduction in ventricular function compared with sham and control animals were considered cardiomyopathic. Despite this qualification, DCM rats were not decompensated hemodynamically, as evidenced by resting HR and MAP values comparable to control and sham animals (Table 2).

The Circulatory Response to Activation of the Exercise Pressor Reflex Is Exaggerated in Cardiomyopathic Rats

At maximal intensity, electrically induced static exercise evoked elevations in MAP and HR in control and sham rats that were significantly less than those expressed in DCM animals (Figure 2A). The rate of the MAP response to static contraction was significantly greater (*P<0.05) in DCM (5.1±0.4 mm Hg/sec) than in control (3.0±0.3 mm Hg/sec) and sham (2.9±0.2 mm Hg/sec). In each group, graded contractions at submaximal work loads elicited pressor responses that were linearly related to force development (*P<0.001, Figure 2B). These responses were augmented in DCM rats over a wide range of stimulus intensities. In all animals, neuromuscular blockade eliminated circulatory alterations to ventral root stimulation, indicating responses were attributable exclusively to muscle contraction. These responses have also been shown to be abolished by transection of the L 4 through L 6 dorsal roots in the rat, confirming the intramuscular origin of the reflex. The same response patterns were obtained by passively stretching skeletal muscle at levels of tension equal to those evoked during muscle contraction (Figures 2C and 2D). Within groups, the magnitudes of the responses obtained during stretch were not significantly different from those elicited by static exercise.
Time Course for the Development of Pressor Reflex Dysfunction

After coronary artery ligation, functional abnormalities in pressor reflex circulatory control were evident in DCM rats 6 weeks after surgery (Figure 3). By 8 weeks, muscle contraction in DCM induced significantly larger increases in MAP and HR than observed in control and sham animals. These exaggerations in responsiveness reached a plateau 10 weeks after ligation. A similar cardiovascular profile developed in response to passive muscle stretch.

The Magnitude of the Pressor Response Is Related to the Severity of Ventricular Systolic Dysfunction

As presented in Figure 4A, fractional shortening (FS) was inversely related to the MAP response to muscle contraction in a linear fashion \( P<0.001 \). Abnormalities in pressor reflex function were evident in DCM animals, with reductions in FS of only 25% to 35% (Figure 4B) becoming maximal at decreases of 45% to 55%. Greater impairment of left ventricular function (ie, FS <20%) did not additionally augment the pressor response to exercise. A comparable response profile was observed by passive stretch of the skeletal muscle (Figures 4C and 4D).

Discussion

Alterations in skeletal muscle morphology and function, including atrophy,2,4 changes in fiber type composition,2,3,25,26 decreased oxidative capacity,2,25,27,28 abnormal high-energy phosphate handling,29 and exercise-induced hyperkalemia,30 have been demonstrated in heart failure. Given these changes, it has been hypothesized that neural reflexes originating within skeletal muscle (ie, the EPR) potentially contribute to the aberrant cardiovascular response to physical activity noted with the pathogenesis of CHF. Using a novel decerebrate rat model,24 the present investigation demonstrated that static muscle contraction elicited exaggerations in EPR activity in animals with dilated cardiomyopathy. This potentiation in pressor reflex control was progressive and inversely related to left ventricular systolic function. Furthermore, there was a significant increase in the rate of the pressor response. Given the shorter activation latencies reported for mechanically sensitive group III compared with metabolically sensitive group IV afferents,31 this finding suggests the response to contraction in DCM was dominated by the mechanoreflex. In support of this conclusion, selective stimulation of mechanically sensitive muscle afferents (by passive stretch) also elicited an augmented circulatory response in DCM animals.

| TABLE 2. Baseline Hemodynamics Before Contraction and Stretch Maneuvers |
|-----------------------------|-----------------------------|-----------------------------|
|                             | Control (n=20)              | Sham (n=15)                 | DCM (n=15)                  |
|                             | Contraction                | Stretch                     | Contraction                | Stretch                     | Contraction                | Stretch                     |
| MAP, mm Hg                  | 102±6                      | 105±6                       | 104±10                     | 108±11                      | 105±6                      | 104±7                       |
| HR, bpm                     | 375±10                     | 382±10                      | 375±21                     | 374±20                      | 379±7                      | 374±8                       |

Values are mean±SEM.
Finally, these physiological abnormalities were observed in an animal model in which the influence of central command was absent. Similar findings have been reported in humans, as low levels of involuntary biceps contraction, a strategy used to selectively engage the mechanoreflex, has been shown to enhance renal vasoconstriction in CHF patients.11 Taken together, these findings suggest that the EPR dysfunction observed in cardiomyopathy is mediated, in part, by the muscle mechanoreflex.

The mechanisms by which the mechanoreflex causes augmentations in EPR function remain obscure. Likely candidates include increased mechanoreceptor density or enhanced receptor responsiveness to mechanical distortion. In support of the latter, it has been demonstrated recently that the circulatory response mediated by the EPR is dependent on the magnitude of sensory input rather than the modality of afferent fiber activation (ie, mechanical versus metabolic stimuli).32 Given the polymorphic nature of the group III skeletal muscle afferents associated with the mechanically sensitive component of the EPR,31 it is also likely that a small proportion of the exaggerated cardiovascular response to static contraction in DCM is driven by direct activation of these neurons by exercise-induced metabolites. Likewise, during contraction it is also plausible that mechanoreceptors are sensitized to mechanical stimulation by alterations in the chemical milieu of the muscle interstitium.33 In support of this concept, impaired removal of exercise-induced metabolites, which may serve to sensitize these afferent neurons, is likely to occur in heart failure.5,9

Perspectives
The physiological events mediating the alterations in EPR function in heart failure are unknown. As discussed, the mechanisms by which the mechanoreflex becomes exaggerated are presently unclear. Furthermore, a great deal of controversy exists regarding the contribution of the metabolic component of the EPR, because its activity has been reported to be both enhanced14,15,21,34 and reduced10,22 in response to exercise in CHF patients. As stated, it is plausible that initial reductions in peripheral blood flow result in an abnormal accumulation of exercise-induced metabolites in the muscle interstitium.5,9 As a result, mechanically sensitive neurons
may become sensitized to physical distortion, whereas metaboreceptors undergo a period of hyperexcitability. Prolonged exposure to these metabolites may eventually serve to desensitize or downregulate metabolically sensitive neurons, culminating in a decrease in their activity. In response to a blunting of the metaboreflex, molecular adaptations may begin within mechanically sensitive afferents to compensate for the functional loss of these fibers. As the disease progresses, edema-related limb congestion may serve to additionally sensitize mechanoreceptors by chronic mechanical distention such that the rate and magnitude of their activation are exaggerated. If this scenario proves valid, there may be periods of hyperexcitability for both metabolically and mechanically sensitive afferents during the course of heart failure that explain some of the discrepancies reported in previous human studies. In addition, other factors, including modifications in muscle metabolism and fiber type composition (eg, reductions in the percentage of oxidative fibers and increases in the percentage of glycolytic fibers), may facilitate these disease-induced alterations in reflex function. For example, because the EPR-mediated circulatory response to activation of glycolytic muscle has been shown to be greater than the response elicited from oxidative muscle, fiber-type transformations could profoundly affect the physiological expression of the pressor reflex.

Clinical Relevance
In the rat model used for this investigation, the magnitude of EPR dysfunction increased as decrements in left ventricular performance became more pronounced. Furthermore, these changes occurred progressively over time. As in human heart failure, it is feasible that these alterations in EPR activity correlate well with the development of exercise intolerance. If so, this model may prove useful in determining the chronology of pharmacological and molecular modifications to the EPR (in relation to ventricular function) that mediate reductions in exercise capacity using techniques presently not available to human experimentation. Such studies maintain the potential of identifying novel therapeutic targets aimed at ameliorating disorders in EPR function and improving exercise capacity in CHF.

The potential benefits of increasing exercise tolerance in heart failure patients are numerous. For example, physical training in human CHF has been shown to improve oxygen uptake, limb blood flow, and skeletal muscle capillary to fiber ratio. In rat models of cardiomyopathy, training has been reported to enhance chronotropic function, reverse detrimental variations in cardiac myosin gene expression and isozyme composition, attenuate ventricular dilation, and diminish wall tension within the heart. Increases in oxidative enzyme activity and improvements in exercise-induced metabolic profiles within skeletal muscle have also been described. If effective treatment of EPR dysfunction in heart failure proves a viable therapy for potentiating exercise tolerance, resultant augmentations in exercise capacity would enhance a patient’s ability to tolerate physical training, potentially increasing the benefits derived from its prescription.

Summary
In conclusion, this investigation provides evidence that abnormalities in EPR function develop with the pathogenesis of cardiomyopathy. Specifically, it has been demonstrated that (1) the EPR is overactive in rats with dilated cardiomyopathy,
(2) the development of abnormal EPR activity occurs progressively, (3) the magnitude of the cardiovascular response mediated by the EPR is greater as the severity of left ventricular dysfunction is increased, (4) mechanosensitive afferents that respond to passive stretch are capable of driving the exaggerated EPR in the absence of metaboreceptor input, and (5) augmentations in EPR and mechanoreflex activity in dilated cardiomyopathy can be expressed in the absence of central command. Additional dissection of the physiological and molecular correlates that accompany this EPR dysfunction will yield insights into the basic anomalies that lead to exercise intolerance in heart failure. Through such insights, these studies have the potential of leading to novel therapeutics targeted at improving exercise capacity in heart failure.

Acknowledgments
This research was supported by grants from the American Heart Association, Texas Affiliate (0160075Y to Dr Garry) and the National Institutes of Health (HL06296 to Drs Mitchell and Garry; HL10473 to Dr Smith). Dr Mammen is a Pfizer postdoctoral fellow in cardiovascular medicine. The authors thank Margaret Robledo, Martha Romero, and Julius Lamar Jr for their expert technical assistance.

References


Role of the Exercise Pressor Reflex in Rats With Dilated Cardiomyopathy
Scott A. Smith, Pradeep P.A. Mammen, Jere H. Mitchell and Mary G. Garry

Circulation. 2003;108:1126-1132; originally published online August 18, 2003;
doi: 10.1161/01.CIR.0000084538.40542.56
Circulation is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 2003 American Heart Association, Inc. All rights reserved.
Print ISSN: 0009-7322. Online ISSN: 1524-4539

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://circ.ahajournals.org/content/108/9/1126

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in Circulation can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to Circulation is online at:
http://circ.ahajournals.org//subscriptions/