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Research Article

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Sympathetic Activation in Exercise Is Not Dependent on Muscle Acidosis

Direct Evidence from Studies in Metabolic Myopathies

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Abstract

Muscle acidosis has been implicated as a major determinant of reflex sympathetic activation during exercise. To test this hypothesis we studied sympathetic exercise responses in metabolic myopathies in which muscle acidosis is impaired or augmented during exercise. As an index of reflex sympathetic activation to muscle, microneurographic measurements of muscle sympathetic nerve activity (MSNA) were obtained from the peroneal nerve. MSNA was measured during static handgrip exercise at 30% of maximal voluntary contraction force to exhaustion in patients in whom exercise-induced muscle acidosis is absent (seven myophosphorylase deficient patients; MD [McArdle's disease], and one patient with muscle phosphofructokinase deficiency [PFKD]), augmented (one patient with mitochondrial myopathy [MM]), or normal (five healthy controls). Muscle pH was monitored by ^{31}P-magnetic resonance spectroscopy during handgrip exercise in the five control subjects, four MD patients, and the MM and PFKD patients. With handgrip to exhaustion, the increase in MSNA over baseline (bursts per minute [bpm] and total activity [%]) was not impaired in patients with MD (17±2 bpm, 124±42%) or PFKD (65 bpm, 307%), and was not enhanced in the MM patient (24 bpm, 131%) compared with controls (17±4 bpm, 115±17%). Post-handgrip ischemia studied in one McArdle patient, caused sustained elevation of MSNA above basal suggesting a chemoreflex activation of MSNA. Handgrip exercise elicited an enhanced drop in muscle pH of 0.51 U in the MM patient compared with the decrease in controls of 0.13±0.02 U. In contrast, muscle pH increased with exercise in MD by 0.12±0.05 U and in PFKD by 0.01 U. In conclusion, patients with glycogenolytic, glycolytic, and oxidative phosphorylation defects show normal muscle sympathetic nerve responses to static exercise. These findings indicate that muscle acidosis is not a prerequisite for sympathetic activation in exercise. (*J. Clin. Invest.* 1998. 101:1654–1660.)

Key words: McArdle's disease • myophosphorylase deficiency • phosphofructokinase deficiency • mitochondrial myopathy • microneurography

Introduction

Increases in heart rate and blood pressure during exercise are regulated both by central and peripheral neural mechanisms (for review see reference 1). The central mechanism is attributed to a parallel central command stimulation of locomotion and the autonomic circuitry to the cardiovascular system from motor centers in the brain (2). The peripheral neural mechanism involves stimulation of mechanoreceptors in the contracting muscle and stimulation of chemosensitive nerve endings by metabolites accumulating in working muscle (for review see reference 3). These muscle afferents reflexly activate the autonomic exercise responses via unmyelinated and thinly myelinated nerve fibers. Although reflex regulation of autonomic adjustment to exercise by chemoreceptors was suggested as early as 1886 by Zuntz and Geppert (4), the chemical mediator(s) in muscle responsible for eliciting this reflex is still undefined. Acidification of muscle, caused by the glycolytic flux and subsequent lactate production that results from glycogen breakdown in muscle during exercise, has been suggested by several investigators to be a potent stimulator of the exercise reflex (5–7).

Microneurography of the peroneal nerve to study muscle sympathetic nerve activity (MSNA)[1] during moderate levels of static exercise is a well-established approach to investigate the function of chemosensitive muscle afferents in humans (8). Using this method, Pryor et al. (9) explored the MSNA response to static exercise in four patients with McArdle's disease, a condition in which muscle acidosis is absent in exercise because of myophosphorylase deficiency that blocks muscle glycogen degradation. They found that the increase in MSNA during exercise was abolished in these patients, suggesting an important role of pH for eliciting the exercise reflex (9). The findings, however, conflict with other studies in patients with glycolytic and glycogenolytic defects, which suggest a normal or even exaggerated sympathetic activation in dynamic exercise as indicated by enhanced blood pressure and catecholamine increases under these conditions (10–12).

The purpose of this study, therefore, was to further investigate the role of muscle pH in eliciting the exercise reflex by

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1. *Abbreviations used in this paper:* ^{31}P-MRS, ^{31}phosphorous magnetic resonance spectroscopy; MM, mitochondrial myopathy; MSNA, muscle sympathetic nerve activity; MVC, maximal voluntary contraction force; PFKD, phosphofructokinase deficiency.

studying muscle sympathetic nerve discharge in response to static handgrip exercise in patients with absent muscle acidosis (muscle phosphorylase and phosphofructokinase deficiencies), in a condition associated with enhanced muscle acidosis in exercise (mitochondrial myopathy), and in healthy subjects with a normal exercise-induced acidification of muscle.

Methods

Characteristics of the subjects. Four women and three men with McArdle's disease, ages 25–51 yr (mean: 35 yr), one 57 yr-old-man with muscle phosphofructokinase deficiency (PFKD), one 41 yr-old-man with mitochondrial myopathy (MM) and five sedentary, healthy volunteers (two women and three men), ages 22–50 yr (mean: 37 yr) participated in the study. No subject took any medication and all had normal 12-lead electrocardiograms. All patients had a clinical story of exercise intolerance. The diagnosis of myophosphorylase and muscle phosphofructokinase deficiency was confirmed biochemically by demonstration of absent enzyme activity in muscle biopsies. The MM patient had a 3,243 mutation of the mitochondrial DNA, a normal MRI of the brain, elevated plasma lactate levels at rest, myopathic findings on muscle biopsy with ragged-red and cytochrome oxidase negative fibers and abnormal mitochondria and no clinical, electromyographic or electroneurographic signs of peripheral nervous affection or encephalopathy. Patients with glycolytic and glycogenolytic defects all had a normal neurological examination.

The study was approved by the ethical committee for Copenhagen and Frederiksberg communities and the institutional review board of the University of Texas Southwestern Medical Center, and Presbyterian Hospital, Dallas, Texas. The subjects were informed of the nature and risks of the study, and gave written consent to participate.

Experimental protocol. Multiunit recordings of postganglionic MSNA were obtained with tungsten microelectrodes inserted selectively into muscle nerve fascicles of the peroneal nerve, using the microneurographic technique of Vallbo et al. (13). The neural signals were amplified, filtered (band width of 700–2,000 Hz), rectified, and integrated to obtain a mean voltage display of MSNA. Requirements for an acceptable recording have been described in detail previously (8, 14). Sympathetic bursts were identified by inspection of the mean voltage neurograms. Nerve traffic was expressed both as bursts per minute (an index of the frequency of sympathetic discharge), and as bursts per minute times mean burst amplitude, also named total activity, that was expressed as a percentage of the control value (an estimate of relative changes in integrated activity).

Handgrip exercise was performed with the nondominant arm. 2 h before data collection started, maximal voluntary contraction force (MVC) during handgrip was determined by having the subject squeeze the handgrip at maximal force for a few seconds during intense verbal encouragements from the investigator. A consistent MVC determined thrice was considered acceptable. All subjects were studied in the recumbent position. Simultaneous MSNA, blood pressure (Finapres; Ohmeda, Englewood, CO), heart rate (derived from the electrocardiogram), and force of contraction, using a handgrip dynamometer, were recorded before, during and after static handgrip to exhaustion at 30% of MVC.

A catheter was placed in a cubital vein of the arm to be exercised in five of the seven McArdle patients and in all other subjects to facilitate the sampling of blood for plasma lactate measurements in all subjects and for blood pH measurements in McArdle and control subjects. Plasma lactate was measured on a YSI 2300 STAT PLUS glucose/lactate analyzer, and blood pH was measured using an AVL Compact blood gas analyzer.

^{31}P-magnetic resonance spectroscopy (^{31}P-MRS) of the forearm flexor muscles was performed before and during static handgrip at 30% MVC to exhaustion as described previously (15), except that the antenna was a 35-mm diameter two-turn inductively driven radio frequency surface coil. Three free induction decays were summed for each spectrum, providing a time resolution of 15 s. Before peak integration, baseline correction was performed applying a 10-point semiautomatic cubic spline procedure. Peak integration was performed with a least square fitting routine, assuming Lorenzian line shape. pH_i was calculated from the chemical shift difference of the inorganic phosphate peak with respect to the phosphocreatine peak (16). Diprotonated phosphate levels in muscle at rest and during the last minute of exercise were calculated as described previously (17). Measurements were performed in four McArdle patients, the MM patient, the PFKD patient and the five healthy subjects. It was not considered necessary to test all McArdle patients with ^{31}P-MRS, because biochemically proven absent myophosphorylase is invariably associated with a slight muscle alkalosis in exercise as that shown in the four studied patients (18, 19).

Responses of MSNA to baroreceptor unloading during the Valsalva maneuver and stimulation of cutaneous afferents during the cold pressor test, where the hand is held in ice water for 2 min, were used as standard nonexercise stimuli after the exercise protocol to evaluate validity of the MSNA responses to exercise stimuli (13, 20).

Post-handgrip ischemia performed by inflating a cuff on the exercised arm to more than systolic pressure immediately after exercise has been used to demonstrate that increases in MSNA during static handgrip exercise in healthy humans is caused by a metaboreflex related to metabolic changes in the working muscle (13, 14). We aimed at testing MSNA during post-handgrip ischemia in the patients with glycogenolytic or glycolytic defects in this study to evaluate whether MSNA responses evoked by exercise were the result of a chemoreflex, mechanoreflex, or a centrally mediated sympathetic outflow. The hallmark of these metabolic myopathies, however, is exercise-induced muscle cramps and pain, and the diagnostic ischemic forearm exercise test has repeatedly evoked painful cramps and potential muscle injury in these patients. For this reason, post-handgrip ischemia can only be performed with caution in a selected number of these patients, who do not easily cramp during exercise. We were able to perform post-handgrip ischemia in one McArdle patient who had no muscle pain at exhaustion in the first handgrip protocol. The post-handgrip ischemia period was limited to 1 min to prevent potential muscle damage. Three healthy control subjects were also studied with post-handgrip ischemia. All subjects studied with post-handgrip ischemia were investigated 1 h after the nonischemic protocol had been completed.

Data analyses. Statistical analysis was performed with Page's test for ordered alternatives followed by multiple comparison with a control, or by Wilcoxon's ranked sum test. Values are expressed as means±SEM. $P < 0.05$ was considered significant.

Results

Baseline data (Table I). At rest, MSNA, heart rate, blood pressure, and muscle pH, assessed by ^{31}P-MRS, did not differ between groups. Plasma lactate was higher in the MM patient than in any other subject. Resting MSNA was assessed by analyzing the average activity in the 3 min preceding exercise or the nonexercise stimuli.

Exercise duration, force, and muscle pain (Table II). The average force development was similar in all groups. The mean duration of exercise was not significantly different between healthy subjects, MM, and McArdle patients. Mean duration of exercise was shorter in the PFKD patient compared with the average of the other groups. The PFKD patient and six of the seven McArdle patients experienced muscle stiffness and pain, but not overt contracture, at the end of exercise. In all these patients, the pain persisted with the same intensity in the first minute of recovery from exercise, assessed on a visual an-

Table I. MSNA, Muscle pH, Heart Rate, Mean Blood Pressure, and Venous Effluent Lactate and pH Responses to Static Handgrip Exercise at 30% MVC in Healthy Sedentary Subjects (Control) and in Patients with McArdle's Disease, PFKD, and MM

			Exercise		
	n	Rest	1.0 min	Exhaustion	Recovery
MSNA					
Bursts/min					
Control	5	18±3	23±4*	35±6*	17±4
McArdle	7	22±4	30±5*	38±5*	27±5
PFKD	1	33	68	100	31
MM	1	25	28	49	21
MSNA					
Total activity					
Control	5	—	18±15	115±17*	35±14
McArdle	7	—	75±47	124±42*	56±42
PFKD	1	—	151	307	−47
MM	1	—	1	131	−9
Muscle pH					
Control	5	7.01±0.01	—	6.88±0.02*	—
McArdle	4	7.03±0.004	—	7.15±0.06*	—
PFKD	1	7.00	—	7.01	—
MM	1	7.07	—	6.56	—
Heart rate; BPM					
Control	5	66±4	79±5*	95±3*	65±3
McArdle	7	70±6	85±6*	92±4*	73±6
PFKD	1	74	98	98	77
MM	1	64	94	101	70
Mean BP; mmHg					
Control	5	106±13	123±12*	140±15*	103±10
McArdle	7	90±7	111±8*	125±14*	104±7
PFKD	1	109	137	137	116
MM	1	78	96	99	78
Venous lactate; mM					
Control	5	1.0±0.1	—	4.4±0.6*	—
McArdle	5	1.2±0.1	—	1.0±0.1*	—
PFKD	1	1.4	—	1.1	—
MM	1	3.2	—	5.7	—
Venous pH					
Control	5	7.35±0.01	—	7.18±0.05*	7.28±0.02*
McArdle	5	7.36±0.01	—	7.38±0.02	7.37±0.01

Values are means±SEM. MSNA is expressed as bursts per minute (*BPM*) and percentage change in total MSNA activity from rest. *Change ($P < 0.05$) from rest. Recovery data are from the first minute after exercise. Total activity of MSNA is shown as percentage change from rest.

alogue scale. Typically the pain disappeared within 5–10 min after exercise.

Effect of exercise on muscle pH and diprotonated phosphate, and venous effluent blood pH and lactate levels (see Fig. 2; Table I). Peak decreases in muscle pH at exhaustion were 0.13±0.02 U in controls (range 0.06–0.20) and 0.51 U in the MM patient (see Table I for absolute values). In contrast, muscle pH increased with exercise by 0.12±0.05 U in McArdle patients (range 0.04–0.20) and was essentially unchanged in the PFKD patient (Table I).

The exercise-induced increase in diprotonated phosphate was similar in McArdle patients (from 1.0±0.1 to 3.3±0.7 mM) and controls (from 0.8±0.1 to 2.9±0.6 mM). The exercise-induced increase in diprotonated phosphate in the PFKD patient was the lowest of all subjects (from 1.0 to 1.7 mM) and the increase in the MM patient was higher than in any other subject (from 1.8 to 10.7 mM).

In accordance with the marked differences in muscle pH during exercise in the various conditions, venous effluent plasma lactate also differed substantially between groups. Plasma lactate decreased slightly with exercise in McArdle and PFKD patients, whereas plasma lactate increased substantially in healthy controls and the MM patient (Table I). In parallel with venous lactate and muscle pH findings in McArdle and control subjects, pH in venous effluent blood from the exercised arm decreased with exercise in healthy controls, but was essentially unchanged in McArdle patients (Table I). Changes in venous blood pH were not assessed in the MM and PFKD patients.

Effect of exercise on muscle sympathetic discharge and cardiovascular responses (Figs. 1–2; Table I). MSNA, expressed both as bursts per minute and as total activity, increased with handgrip exercise in all subjects. The increase in MSNA occurred after 45–60 s of exercise in all subjects, except the PFKD patient, whose MSNA increased after ~ 20 s of exercise. MSNA, expressed both as burst per minute and as percent change in total activity, was essentially unchanged from basal values in the first 30 s of exercise in the MM patient (Δ3 bpm, Δ1%), McArdle patients (Δ-3±2 bpm, Δ-13±6%), and healthy controls (Δ1±1 bpm, Δ7±6%). The maximal increases during exercise were comparable in the MM, McArdle, and healthy subjects. The PFKD patient had a MSNA response to exercise that was higher than in any of the other subjects. MSNA returned to basal levels in the first minute after exercise in all studied groups.

Heart rate and blood pressure increased significantly during static handgrip exercise in all subjects. Peak exercise heart rate and blood pressure responses, expressed both in absolute values and as change from basal, did not differ between groups.

Effect of post-handgrip ischemia on muscle sympathetic discharge and cardiovascular responses (Fig. 3). During post-handgrip ischemia in the McArdle patient, MSNA remained elevated in contrast to the prompt return of MSNA to basal levels during nonischemic recovery (Fig. 3). Similar findings were obtained in the three healthy control subjects during post-handgrip ischemia (Fig. 3). In contrast to nonischemic recovery af-

Table II. MVC and Duration of Static Handgrip Exercise at 30% MVC in Five Healthy Subjects (Control), in Seven Patients with Myophosphorylase Deficiency (McArdle), in One Patient with Muscle PFKD, and in One Patient with MM

	MVC (kg)	Exercise duration (s)
Control	40±5	191±25
McArdle	37±3	145±25
PFKD	40	102
MM	32	158

Values are means±SEM.

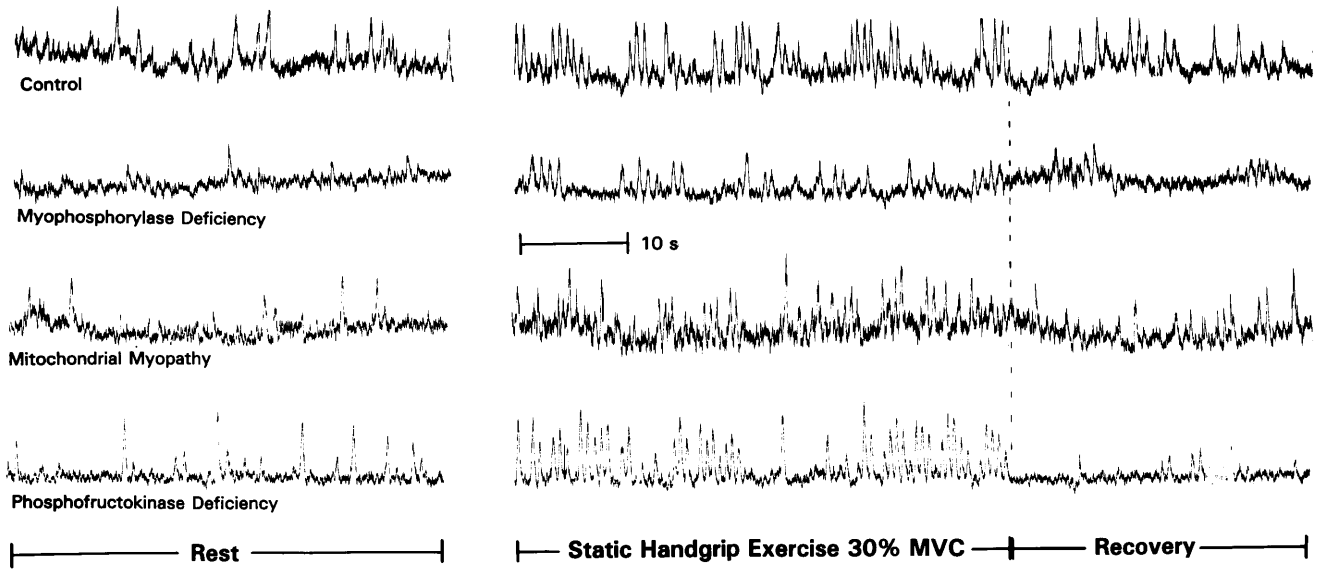

Figure 1. Integrated neurograms of muscle sympathetic nerve activity at rest, during the last 35 s of static handgrip exercise at 30% of maximal voluntary contraction force (*MVC*) to exhaustion and during recovery from exercise in a healthy subject (*Control*), a myophosphorylase-deficient patient, the MM patient, and the muscle PFKD patient.

ter handgrip exercise in McArdle patients, the exercise pressor response was sustained during post-handgrip ischemia in the patient. In fact, during post-handgrip ischemia in the McArdle patient, the pressure was 6 mmHg higher than at the end of exercise. In the controls, blood pressure dropped by 7±2 mmHg during post-handgrip ischemia compared with the pressure at the end of exercise, but was still 16±6 mmHg higher than basal pressure.

MSNA responses to the Valsalva maneuver and the cold pressor test. The Valsalva maneuver and the cold pressor test evoked comparably large increases in MSNA in all studied groups. Thus, the Valsalva maneuver evoked an increase in total MSNA of 227±99% in McArdle patients, 332±142% in healthy controls, 271% in the PFKD patient and 214% in the MM patient. Based on previous experience, the PFKD patient and two McArdle patients declined to perform the cold pressor test. In the other subjects, the cold pressor test evoked an increase in MSNA from rest to the last minute of hand in ice of 49±28% in McArdle patients, 74±42% in healthy controls, and 23% in the MM patient.

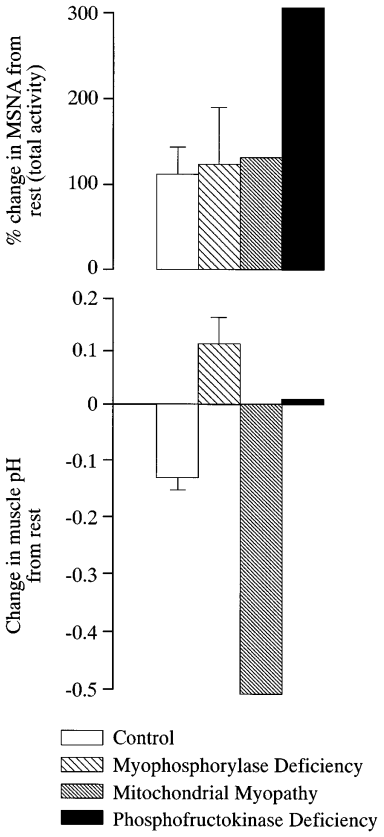

Figure 2. Changes from rest to exhaustion in MSNA and ^{31}P-MRS–assessed muscle pH in healthy subjects (*Control*) and in patients with McArdle's disease, MM and muscle PFKD. Values are mean±SEM. The exercise paradigm was handgrip exercise at 30% of maximal voluntary contraction force. For number of observations per condition see Table I.

Discussion

By using different inborn errors of skeletal muscle metabolism as experimental models, we have studied whether a decrease in muscle pH is a prerequisite for sympathetic activation during exercise. The principal new finding is that sympathetic activation during static exercise, as indicated by increases in muscle sympathetic nerve activity, heart rate, and blood pressure, is not impaired by absent muscle acidosis or augmented by enhanced muscle acidosis during exercise. The results indicate that activation of sympathetic nerve discharge during exercise is not dependent on a lowering of muscle pH related to glycogen degradation and lactate production in contracting muscle.

These findings challenge the concept of a close coupling between a lowering of muscle pH and sympathetic activation in exercise, a link that has been proposed by several reports in the last decade. The major findings have been: (*a*) human studies that have employed ^{31}P-MRS to identify a temporal correlation between the fall in muscle pH during static exercise and

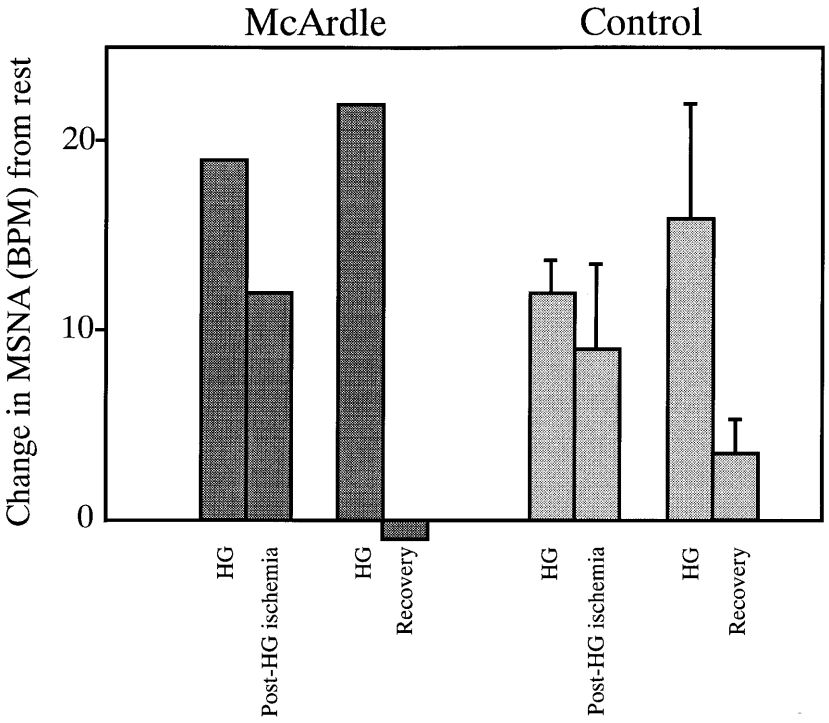

Figure 3. Changes in MSNA expressed as bursts per minute (*BPM*) from rest to handgrip exercise (*HG*) and from rest to post-handgrip ischemia or nonischemic recovery in one McArdle patient and three healthy control subjects. Data were collected in the last minute of exercise and the first minute after exercise. Values are means±SEM for controls.

sympathetic activation as indicated by increase in MSNA, blood pressure, and vasoconstriction (6, 7), and which have demonstrated attenuated sympathetic activation during exercise in conditions that lower lactate production, such as glycogen depletion, myophosphorylase deficiency, and dichloroacetate administration (9, 21, 22); and (*b*) animal studies that have identified a temporal correlation between the exercise pressor response and the increase in venous effluent H^+ from working muscle (23), and shown activation of group III and IV muscle afferents (24, 25) and enhanced exercise pressor reflex responses (5, 26) after intra-arterially administered lactic acid in skeletal muscle. The general acceptance of the pH-hypothesis as a possible cardiovascular regulatory mechanism during exercise is indicated by its inclusion in recent textbooks on cardiovascular homeostasis (27, 28).

However, besides the findings in this study, several previous observations have challenged the role of muscle pH as a sympathetic activator: (*a*) the fact that pH normally continues to drop after exercise in healthy subjects (29, 30) while MSNA, blood pressure, and heart rate return promptly to baseline; (*b*) the finding that factors related to training and not hydrogen ion concentration seems to correlate with MSNA increases in exercise (31) and that sympathetic exercise responses have been shown to correlate more closely to levels of diprotonated phosphate than pH in muscle (29); and finally, (*c*) that sympathetic activation as indicated by pressor and neuroendocrine responses, rather than being attenuated, is in fact exaggerated during dynamic exercise in glycolytic disorders that lack muscle acidosis in exercise (11, 12).

In particular, our study contradicts a study by Pryor et al. (9) who found an abolished exercise pressor response and an abolished increase in MSNA during static handgrip exercise in patients with McArdle's disease. The lack of sympathetic nerve responses to exercise was attributed to an absence of lactate production and muscle acidosis in McArdle patients (9). Two major differences between this investigation and the study by Pryor et al. may alone or in combination explain the differing results. First, the interpretation of sympathetic responses to exercise was confounded by a 60% higher resting sympathetic activity in McArdle patients compared with healthy controls in the study by Pryor et al. (9). In our study, resting sympathetic tone as indicated by MSNA, heart rate, and blood pressure was similar between groups. Second, extra care was exercised in the Pryor et al. study to protect McArdle patients from muscle contracture. This resulted in an unusually short duration of exercise (90 s) and the exclusion of post-exercise ischemia from the protocol. In contrast, patients exercised until exhaustion in our study. The cautious examination of the patients by Pryor et al. may have resulted in an underestimation of the MVC in McArdle patients. All of the confounding factors in the Pryor et al. study blunt sympathetic responses and, therefore, may explain the no-response nature of their study. No McArdle patient participated in both our study and the study by Pryor et al. Still, the possibility that responses may vary among McArdle patients seems unlikely since each of seven consecutively studied patients in our study demonstrated a normal sympathetic response to exercise. Extending the study to include a different glycolytic defect showed similar results—in fact the PFKD patient showed the largest MSNA increase of any subject.

It could be argued that the normal pressor response and exercise-induced increase in MSNA in our patients were related to a centrally mediated sympathetic outflow caused by a higher central command in this condition or elicited by a mechanoreflex. However, the time course of the MSNA response to exercise with a sympathetic activation occurring $\sim$ 45 s after onset of exercise closely resembles that characteristic of a chemoreflex (8, 13, 14, 27). In contrast, sympathetic

responses caused by a mechanoreflex (32) or central command (8, 33) occur at the onset of exercise. Furthermore, the sustained MSNA and pressor response during post-handgrip ischemia, which we demonstrated in the McArdle patient, clearly indicates that the increase in MSNA during exercise in McArdle patients is caused by a chemoreflex unrelated to muscle acidity.

The effect of muscle pain on MSNA is unknown, but our data suggest that MSNA is not affected by this type of pain. In fact, MSNA appears not to respond with an arousal response to pain stimuli in general as indicated by no response of MSNA to painful electrical skin stimulation (34), and the finding that MSNA may be inhibited by other painful stimuli (13). This study indicates that MSNA is not affected by muscle pain since MSNA returned promptly to basal levels after exercise in the PFKD and McArdle patients, although muscle pain in the first minute post-exercise was as intense as during exercise in these patients. Thus, interpretation of MSNA responses to exercise were not affected by differences in pain perception between groups in this study.

McArdle and PFKD patients have uniformly exaggerated sympathetic activity to the neuroendocrine and cardiovascular system during cycle exercise (11, 12). The hyperkinetic circulatory and endocrine exercise responses under these conditions of absent lactic acidosis are comparable to the circulatory and endocrine responses to cycle exercise in mitochondrial myopathy patients with severely enhanced exercise-induced muscle and systemic lactic acidosis (35). These findings along with those of this study, indicate that error signals unrelated to increases in $[H^+]$ are responsible for sympathetic activation in static as well as dynamic exercise.

The results in the patient with MM also challenge the role of muscle acidification in eliciting sympathetic activation during exercise. Despite an exaggerated muscle acidosis compared with controls, the MM patient showed normal increases in MSNA and a normal pressor response during exercise. Muscle pH in the MM patient stayed below 6.6 in the first minute after exercise, whereas MSNA and pressor responses returned promptly to basal levels. A similar continued, or more commonly, enhanced muscle acidosis after static exercise is seen in healthy subjects (29, 30), in whom MSNA and pressor responses also return to basal immediately after exercise. Key studies that have advanced the hypothesis of a close coupling between muscle pH and sympathetic outflow during static exercise, have failed to address the issue of rapidly decreasing MSNA, blood pressure, and heart rate during enhanced muscle acidosis in the immediate recovery after exercise (7, 9). This study has documented that the increase in MSNA during static exercise is normal, whether muscle pH decreases or increases abnormally in exercise, and that MSNA returns to basal levels immediately after exercise irrespective of whether muscle acidosis is absent or enhanced after exercise.

Diprotonated phosphate changes have been found to correlate better than pH with sympathetic activation during exercise, and has therefore been suggested as a possible mediator of the exercise reflex (17). Our finding of the lowest exercise-induced increase in diprotonated phosphate in the PFKD patient who had the highest MSNA increase of any subject, and the finding of the highest exercise-induced increase in diprotonated phosphate in the MM patient who had a normal MSNA response to exercise, do not suggest an important role for diprotonated phosphate as a mediator of the exercise reflex.

In contrast to patients with muscle phosphorylase or phosphofructokinase deficiencies, who have no abnormalities of peripheral nerves or the CNS, patients with mitochondrial myopathies often have such abnormalities (36). However, our patient with mitochondrial myopathy had no clinical, neurophysiologic or MRI signs of peripheral nerve or CNS affection, indicating that the studied patient was a valid model, like McArdle and PFKD patients, to evaluate the relationship between muscle pH and MSNA responses to exercise. This is further emphasized by the normal responses of the muscle sympathetic nerve response to standard nonexercise stimuli in all patients compared with healthy controls (13, 20).

Mitochondrial disorders often do not show abnormal decreases in intramuscular pH during exercise as assessed by ^{31}P-MRS (29, 30). This may be due to an adaptation in these patients to increased lactate production by increasing rates of acid extrusion from muscles (30). In line with this, we screened three other patients with mitochondrial myopathy (one patient with a 4,409 mtDNA mutation and two patients with large-scale single mtDNA deletions) before finding the presented MM patient. All these other MM patients had normal intramuscular pH and MSNA responses to exercise (data not shown). The small drop in muscle pH observed in healthy controls in this study is consistent with findings in other studies of healthy humans performing static handgrip exercise at 30% maximal voluntary force (7, 9, 31). The severe muscle acidosis with this type of exercise observed in the MM patient, may have been caused by a low buffering capacity associated with a high muscle lactate level at rest.

In summary, our findings suggest that a mediator other than muscle acidification is the principal activator of sympathetic activation during exercise. If autonomic adaptation to exercise is regulated by redundant mechanisms, this study can not preclude a role of muscle acidosis in this setting. However, this study provides direct evidence that muscle acidification is not a prerequisite for sympathetic activation in exercise.

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