Control of Skin Sympathetic Nerve Activity During Intermittent Static Handgrip Exercise

Urs A. Leuenberger, MD; Sogol Mostoufi-Moab, MD; Michael Herr, PhD; Kristen Gray, MS; Allen Kunselman, MA; Lawrence I. Sinoway, MD

Background—Exercise activates the sympathetic nervous system as a function of the type and intensity of exercise and of the target organ studied. Although central command and activity of metabolically sensitive afferents from exercising muscle are the principal determinants of sympathetic outflow directed to skeletal muscle, the mechanisms that govern sympathetic outflow directed to skin are less clear.

Methods and Results—We measured skin sympathetic nerve activity (SSNA) during intermittent static handgrip (SHG; at 45% of maximal voluntary contraction; four 5-second contractions per minute for 3 minutes), during unrestricted forearm perfusion (control), during stimulation of forearm mechanoreceptors with venous congestion, and during ischemia produced by forearm circulatory arrest. Under all 3 conditions, SSNA increased within 1 to 2 seconds of the onset of handgrip. During ischemia but not during venous congestion, SSNA increased more compared with control (P<0.05) and remained elevated when forearm ischemia was maintained after handgrip exercise (posthandgrip circulatory arrest). In addition, simulated handgrip and intermittent forearm compression produced by a pneumatic cuff also evoked brief increases of SSNA.

Conclusions—In addition to central neural factors, afferent input from exercising muscle plays an important role in modulating sympathetic outflow directed to skin. (Circulation. 2003;108:2329-2335.)

Key Words: exercise ■ nervous system, sympathetic ■ reflex ■ physiology

Exercise activates the sympathetic nervous system as a function of the type of exercise, its intensity, and the target organ studied.1,2 Exercise-induced sympathetic nervous system activation is thought to redistribute blood flow toward exercising muscle and to play a role in thermoregulation.1,2 New neurophysiological techniques allow measurements of sympathetic nerve traffic in peripheral nerves in humans.3 Two types of efferent sympathetic nerve traffic have been identified. First, muscle sympathetic nerve activity (MSNA) is thought to represent vasoconstrictor nerve traffic directed to skeletal muscle.3 MSNA is controlled principally by the arterial baroreflex and is modulated by afferent signals from metabolically sensitive nerve endings in skeletal muscle, from arterial chemoreceptors, and by “central command.”3,2,4–6 Second, skin sympathetic nerve activity (SSNA) is targeted to the skin vasculature and to sweat glands.3 SSNA is less tightly linked to baroreflex control but responds to central neural stimuli and arousal.7 Previous studies suggest that SSNA is increased by central command and by input from mechanoreceptors but not from metabolically sensitive skeletal muscle afferents.7,8 In addition, whereas MSNA typically rises in a time-dependent fashion and after the onset of exercise,9,10 it has been suggested that SSNA may increase immediately before the onset of muscle contraction.7 However, in recent studies performed in our laboratory, exercise-induced increases of SSNA consistently followed the onset of muscle contraction.11,12 Furthermore, during prolonged rhythmic handgrip exercise in heart failure, SSNA rose and remained elevated during posthandgrip circulatory arrest (PHG-CA).12 Because this maneuver isolates the effects of metabolic byproducts of muscle contraction on metabolically sensitive muscle afferents, under certain conditions SSNA may be modulated by the muscle metaboreflex.12

In this study, we sought to further examine the mechanisms that govern SSNA during exercise. Therefore, we tested the effects of intermittent static handgrip (SHG) on SSNA during unrestricted forearm perfusion, during stimulation of muscle mechanoreceptors achieved by forearm venous congestion (VC), and during stimulation of muscle metaboreceptors resulting from forearm ischemia. Our previous work suggests that VC induced by inflating an upper-arm cuff to 90 mm Hg doubles the MSNA response to handgrip exercise.11 Our hypotheses were, first, that SSNA increased not before but immediately after the onset of muscle contraction. Second,
we postulated that stimulation of mechanoreceptors in the forearm would enhance the SSNA response to exercise. Last, we postulated that muscle contraction under ischemic conditions would also stimulate SSNA. Our results support the first and last hypotheses but not the second.

**Methods**

**Subjects**

We studied 17 healthy volunteers (2 female, 15 male; age 26±1 years) who were on no medications. The studies were performed after an overnight fasting period. Caffeine-containing products were held for at least 12 hours. The study protocol was approved by the Institutional Review Board, and each participant signed written informed consent. All studies were performed in a temperature-controlled (21°C) human research laboratory with the subjects in the supine position.

**Hemodynamic Measurements**

Mean arterial blood pressure (MAP) was determined with an automatic sphygmomanometer (Dinamap, Critikon), and heart rate (HR) was determined via ECG as described previously.12

**Microneurography**

SSNA was determined via peroneal microneurography3 as described previously.11,12 Briefly, a tungsten microelectrode was inserted into the peroneal nerve below the fibular head to record activity in efferent sympathetic fascicles carrying vasoconstrictor and sudomotor nerve traffic to the lower leg.3 The nerve traffic signal was filtered, amplified, rectified, and integrated and recorded electronically (see below). We considered an SSNA signal as acceptable when (1) weak electrical stimulation of the electrode produced paresthesias but no muscle twitches, (2) skin stroking within the receptive field of the impaled nerve elicited an afferent response, (3) a loud noise resulted in discharges of SSNA (arousal), and (4) the signal-to-noise ratio was >3:1. SSNA was expressed as bursts/min and total amplitude/min (U/min).

**Skin Blood Flow**

Skin blood flow was determined with a laser Doppler flow probe positioned within the receptive field represented by the SSNA signal and was expressed as relative units.6

**Intermittent SHG Exercise**

Intermittent SHG exercise was performed in the nondominant arm with a Stoelting handgrip dynamometer. A pneumatic cuff was mounted on the upper arm to produce forearm VC or ischemia (see below). The maximum voluntary contraction was determined. We chose a workload of 45% of maximum voluntary contraction at a rate of 4 contractions per minute (5 seconds contraction, 10 seconds relaxation) in our protocol because in pilot studies this workload produced prompt discharges of SSNA and could be maintained easily for 3 minutes. Because we were concerned that verbal commands could elicit arousal-related discharges of SSNA,13 we used a light-emitting diode (LED) as the cue for onset and offset of handgrip. The subjects were instructed to perform handgrip whenever the indicator light was on. The force of contraction was maintained at the desired level via visual feedback from a force indicator. The subject's effort was rated according to the Borg scale.14 To reduce anticipatory effects, before data collection the subjects were familiarized with all experimental procedures.

**Experimental Protocols**

The subjects performed one or several of the protocols described below. Thirty minutes was allowed between protocols to allow blood pressure, HR, and SSNA to return to baseline values.

![Diagram of experimental protocols](http://circ.ahajournals.org/)

**Figure 1.** Schematic of experimental protocols 1 to 3. VC indicates forearm VC (cuff pressure, 90 mm Hg); CA, forearm ischemia induced by forearm CA (cuff pressure, 250 mm Hg); and SHG, intermittent SHG.

**Protocol 1 (Control)**

After baseline measurements over a period of 5 minutes, the subjects performed intermittent SHG at 45% of maximum voluntary contraction (5 seconds contraction, 10 seconds relaxation) for 3 minutes. Just before the end of the last contraction, PHG-CA was produced for 1 minute by inflating the arm cuff to 250 mm Hg. The cuff was then deflated, and recovery measurements were made over a period of 4 minutes.

**Protocol 2 (VC)**

After baseline measurements for 5 minutes, VC of the forearm was produced by inflating the arm cuff to 90 mm Hg.11 This procedure has been shown to markedly enhance the reflex responses to static exercise, probably via sensitization of mechanically sensitive muscle afferents. After 5 minutes of VC, intermittent SHG was performed for 3 minutes. Just before the end of the last contraction, the cuff pressure was increased to 250 mm Hg for 1 minute (PHG-CA), after which the cuff was deflated, and recovery data were obtained for 4 minutes.

**Protocol 3 (Forearm Ischemia)**

After baseline measurements for 5 minutes, forearm ischemia was produced via forearm CA by inflating the arm cuff to 250 mm Hg. After 5 minutes of ischemia, intermittent SHG was performed for 3 minutes. After exercise, ischemia was maintained for 1 minute (PHG-CA). Recovery measurements were obtained for 4 minutes. Figure 1 shows a schematic of protocols 1 to 3.

**Protocol 4 (Virtual Handgrip)**

In these experiments, the subjects were instructed to "pretend" to exercise in synchrony with the LED signal without actual forearm contraction. The LED was activated and deactivated as above (5 seconds on, 10 seconds off) for 3 minutes.

**Protocol 5 (Intermittent Forearm Compression)**

In this protocol, a pneumatic cuff was placed around the forearm. After baseline measurements, the forearm cuff was inflated rhythmically (5 seconds on, 10 seconds off) to 50 mm Hg for 3 minutes.

**Signal Averaging and Integration**

To assess the timing of SSNA within the contraction-relaxation cycle, we time-averaged and integrated SSNA throughout the 3-minute exercise period (12 contraction-relaxation cycles) by a computerized method. For this purpose, the outputs from the LED, the dynamometer, and SSNA were digitized and recorded electronically (PowerLab software, ADInstruments).

The signals were coalesced and processed as described previously.15,16 A buffer holding 1 complete contraction-relaxation cycle was initialized for each averaged signal. The onset of the LED signal identified the onset of each contraction-relaxation cycle and its corresponding SSNA data point. The 12 initial SSNA data points were averaged and saved in the SSNA averaging buffer. Similarly, the SSNA and hand dynamometer data points were averaged for each time point in the contraction-relaxation cycle. The DC baseline level of the averaged SSNA signal was then removed by subtracting its minimum value from all values in the signal.
Numerical integration of averaged and baseline-adjusted SSNA was performed by use of a trapezoidal integration algorithm. Each SSNA integral for 1 contraction-relaxation cycle was normalized, and the fraction of integrated SSNA during each phase of the contraction-relaxation cycle was determined.

Data Analysis and Statistics

We used an ANOVA for repeated measures to compare the responses of blood pressure, HR, and SSNA under the 3 main conditions (control, VC, CA). Comparisons between conditions were performed by a 2-way ANOVA. Post hoc comparisons were performed by use of Dunnett’s test. Where appropriate, pairwise comparisons were performed by paired t test. All statistical analyses were performed with the SAS statistical package (SAS Institute). A value of \( P<0.05 \) was considered statistically significant. All data are presented as mean±SEM.

**Results**

Effects of Intermittent SHG on MAP and HR

The MAP and HR data are shown in the Table. Under control conditions (n=10) and during VC (n=10), intermittent SHG led to small time-dependent increases in MAP and HR that resolved during PHG-CA. Substantial increases of MAP and HR were noted when SHG was performed during ischemia (CA; n=9). The increase of MAP was largely maintained during PHG-CA.

Effects of Forearm VC or Forearm Ischemia on the SSNA Responses to Intermittent SHG

Recordings of the responses to intermittent SHG are shown in Figures 2 and 3, and the group data are shown in Figure 4 and

| Effects of Intermittent SHG and PHG-CA on MAP, HR, and SSNA |
|-------------------|-----------------|----------|----------|----------|----------|----------|
|                   | Baseline        | VC or CA | G1       | G2       | G3       | PHG-CA   | R         | F         | P         |
| MAP, mm Hg        |                 |          |          |          |          |          |           |           |           |
| Control (n=10)    | 91±3            | ...      | 93±3     | 96±3*    | 96±4*    | 92±4     | 91±4      | 3.74      | 0.0064    |
| VC (n=10)         | 92±2            | 92±2     | 96±2     | 98±3*    | 101±3*   | 94±3     | 93±2      | 4.41      | 0.0011    |
| CA (n=9)          | 92±2            | 94±2     | 103±3*   | 110±3*   | 118±4*   | 111±5*   | 93±3      | 27.74     | 0.0001    |
| HR, bpm           |                 |          |          |          |          |          |           |           |           |
| Control           | 57±2            | ...      | 60±2     | 62±2*    | 62±3*    | 57±2     | 58±2      | 7.35      | 0.0001    |
| VC                | 58±2            | 58±2     | 63±3*    | 65±3*    | 61±2     | 58±2     | 57±2      | 8.13      | 0.0001    |
| CA                | 56±2            | 56±2     | 64±3*    | 66±4*    | 70±4*    | 62±3     | 56±2      | 13.91     | 0.0001    |
| SSNA, U/min       |                 |          |          |          |          |          |           |           |           |
| Control           | 93±14           | ...      | 159±28*  | 128±18   | 130±17   | 98±18    | 80±8      | 6.27      | 0.0002    |
| VC                | 111±18          | 130±25   | 209±33*  | 171±35*  | 155±34   | 87±22    | 107±18    | 8.23      | 0.0001    |
| CA                | 77±13           | 109±15   | 163±30*  | 142±21   | 235±42*  | 166±39*  | 69±14     | 6.75      | 0.0001    |
| SSNA, bursts/min  |                 |          |          |          |          |          |           |           |           |
| Control           | 12.4±1.9        | ...      | 15.5±2.1 | 15.8±2.3 | 16.0±2.0 | 13.2±2.6 | 11.6±1.4  | 3.36      | 0.0115    |
| VC                | 13.9±2.0        | 14.7±2.2 | 18.9±2.2*| 15.9±2.1 | 15.1±2.3 | 10.7±2.3 | 14.6±2.5  | 4.16      | 0.0017    |
| CA                | 10.8±2.1        | 13.1±1.9 | 18.2±1.6*| 15.2±2.3 | 21.6±2.6*| 17.3±3.3*| 9.0±2.0   | 6.42      | 0.0001    |

G1–3 indicate minutes 1–3 of intermittent static handgrip; R, recovery.

\( *P<0.05 \) vs baseline.

**Figure 2.** Recording from 1 subject showing blood pressure (BP), SSNA, muscle tension, and LED signal during intermittent SHG in freely perfused condition (Control).
Before exercise, VC and CA had no significant effects on SSNA. In all 3 conditions, the onset of SHG produced a prompt initial increase of SSNA. During control (n=10) and VC (n=10), SSNA decreased progressively during minutes 2 and 3 of SHG and returned to baseline during PHG-CA. In contrast, during ischemic handgrip (CA; n=9), SSNA was highest during minute 3 and remained elevated during PHG-CA. The subjective effort (Borg scale) at end exercise was 9.0±0.6 for control and was higher (P<0.05) at 13.4±1.2 for VC and at 16.4±1.5 for CA, respectively.

Signal Averaging and Integration
SSNA signal averaging demonstrated that under all 3 conditions, sympathetic discharges were closely linked to the onset of handgrip and typically occurred 1 to 2 seconds after but not before the onset of contraction (Figure 5). Overall, during exercise under control, VC and CA conditions 44±2%, 49±3%, and 45±1% of SSNA, respectively, occurred during the 5-second contractions, whereas the balance occurred during the 10-second relaxation periods (P=NS between conditions). Comparison of the first 2 versus the last 2 contraction-relaxation cycles demonstrated habituation of these responses, which reached statistical significance for VC and CA (Figure 6).

Effects of Virtual Handgrip (n=6)
Virtual handgrip produced a prompt and transient increase in SSNA. During minute 1 of simulated exercise, SSNA increased from 110±29 to 185±37 U/min (P<0.05) but returned to 115±23 and 109±29 U/min during minutes 2 and 3 (P=NS). Signal averaging and integration of SSNA demonstrated marked habituation from the first to the last cycle (data not shown). A recording of SSNA during simulated handgrip from 1 subject is shown in Figure 7.
Figure 6. Graph showing integrated SSNA during 5-second contraction phase of first and last 2 contraction-relaxation cycles under 3 main experimental conditions. If no synchronization occurs between SSNA and handgrip, integrated SSNA should be uniformly spread over 15-second interval so that 5-second contraction and 10-second relaxation phases should contribute 1/3 (dotted line) and 2/3 of total integrated SSNA, respectively. *P<0.05.

Effects of Forearm Compression (n=4)

Intermittent forearm compression produced a prompt and transient increase in SSNA with a magnitude and temporal pattern similar to that of virtual grip. During minute 1 of intermittent forearm compression, SSNA increased from 93±23 to 132±32 U/min (P=0.057), remained elevated during minute 2 at 142±27 U/min (P<0.05), and decreased to 111±22 U/min during minute 3 (P=NS). Signal averaging and integration of SSNA demonstrated marked habituation from the first to the last cycle (data not shown).

Onset Latency for SSNA During Intermittent SHG (n=9)

The onset latency for SSNA relative to the onset of handgrip contraction was 0.60±0.05 second for control exercise, 0.59±0.06 second for VC, and 0.69±0.05 second for CA, respectively (P=NS).

Skin Blood Flow Data (n=6)

Compared with baseline, skin blood flow did not change significantly during handgrip exercise (control, 1.28±0.54 to 1.31±0.56 U; VC, 1.04±0.43 to 1.20±0.54 U; CA, 0.89±0.50 to 0.97±0.52 U; P=NS for all). Similarly, skin vascular resistance did not change significantly during handgrip exercise.

Discussion

Our studies produced 3 major findings. First, intermittent SHG triggers discharges of SSNA within 1 to 2 seconds after the onset of but not before muscle contraction. During continued exercise, SSNA decreased toward baseline despite increasing effort. Second, handgrip exercise performed during VC, a procedure that is thought to sensitize mechanically sensitive afferents, produced no significant augmentation of the SSNA responses. Third, intermittent SHG performed during forearm ischemia evoked a progressive increase in SSNA that was sustained during PHG-CA. These data suggest that input from metabolically sensitive nerve endings is capable of modulating sympathetic outflow to the skin. In contrast, with the handgrip paradigm used here, we found no significant role of the mechanoreflex in controlling sympathetic activity directed to skin. These findings emphasize the complexity of sympathetic neural control during exercise.

A unique feature of our study was that we modulated experimentally the activity of afferent input from metabolically and mechanically sensitive muscle afferents and of central neural factors during handgrip exercise. The substantial pressor response noted during ischemic exercise (CA) and during PHG-CA is clear evidence that the muscle metaboreflex was activated. Conversely, the prompt (transient) increase of SSNA noted by simulated exercise (“virtual” handgrip) demonstrates that central cortical events alone can activate SSNA. One consistent finding of our study was that SHG resulted in prompt increases of SSNA. Under all experimental conditions, the very first handgrip contraction was followed within 1 to 2 seconds by discharges of SSNA. In no subject did we observe an increase in SSNA above baseline before the development of muscle tension. This is in contrast to a report by Vissing et al in which SSNA increased immediately before the onset of muscle contraction. We believe this discrepancy may be a result of methodological differences. To reduce the possibility that sympathetic discharges preceding the onset of exercise were caused by a nonspecific arousal effect, we used visual rather than verbal instructions to precisely signal the onset and offset of handgrip. Although after the first contraction, all further handgrip contractions occurred at regular intervals and were therefore anticipated by the subjects, reproducible SSNA discharges preceding muscle contraction were not noted.

It has been postulated that discharges of SSNA evoked by exercise are principally a result of central command. Central command refers to a signal emanating from the rostral brain that projects to autonomic circuits in the brainstem and is part of the generalized activation of motor and sympathetic neurons at the onset of exercise. Several of our observations support a role of central command in initiating the SSNA response to exercise. For example, the latency period of SSNA discharges from the onset of muscle contraction is consistent with a central origin of this signal and is similar to that noted when the cerebral
cortex was stimulated with a magnet.19 Furthermore, prompt and transient discharges of SSNA with a similar latency period also occurred in our virtual handgrip trials when afferent input from exercising muscle was absent. The transient discharges of SSNA during intermittent forearm compression suggest that afferent input from the arm, presumably relayed via mechanically sensitive afferents in skeletal muscle or in skin, can also contribute to exercise-induced discharges of SSNA. Interestingly, in both cases (virtual handgrip and intermittent forearm compression), the responses were substantially attenuated when the stimulus was continued. It is well known that arousal responses diminish over time with repeated presentation of the stimulus,13,20 a phenomenon referred to as “habituation.”

Two observations strongly suggest that input from metabolically sensitive afferents from exercising muscle is a major modulator of SSNA. First, during intense metabolic stimulation produced by exercise during forearm ischemia (CA), SSNA increased progressively and remained elevated after exercise as long as the forearm was kept ischemic (PHG-CA). Because this maneuver traps metabolites accumulated in the forearm during exercise, this response is probably mediated by metaboreflex activation. Second, whereas early during exercise, sympathetic discharges occurred disproportionately during the 5-second handgrip rather than during the 10-second relaxation phase, toward the end of ischemic exercise, SSNA was almost evenly distributed throughout the exercise cycle. Therefore, skin sympathetic activation occurred largely at a time when central command and mechanoreceptors would be expected to be inactive.

Our finding of elevated SSNA during PHG-CA differs from reports by Visser and Saito.7,8 There are 2 potential explanations for this discrepancy. First, it is possible that our exercise paradigm provided a greater metabolic stimulus than those reported previously. Second, the “metaboreflex response” of SSNA in our study may have been facilitated by the intermittent nature of handgrip. It is notable that we previously reported a PHG-CA response for SSNA in patients with heart failure in whom profound muscle acidosis is common even during intermittent exercise.12

Although during ischemic exercise SSNA rose progressively, in both nonischemic conditions (control and VC), SSNA decreased over time despite increasing effort and approached baseline during the period of PHG-CA. The lack of a progressive rise of SSNA during intermittent SHG is similar to that during rhythmic handgrip exercise performed for 30 minutes.12 Because SSNA was highest during the initial exercise cycles and did not rise progressively despite increasing effort, central command alone cannot explain the pattern of SSNA during exercise in our study.

Although VC, a maneuver that increases the MSNA response to handgrip by sensitizing mechanically sensitive afferents in the forearm,11 did not significantly affect magnitude and time course of SSNA during handgrip exercise, a potential modulating role for mechanoreceptors cannot be excluded. For example, our signal-averaged data revealed prominent transient discharges of SSNA that occur characteristically within 1 to 2 seconds of development of muscle tension but also with external forearm compression. This efferent discharge pattern is consistent with that of group III afferents in response to electrical stimulation.21 It should also be considered that afferent nerve endings in skeletal muscle may have overlapping properties, ie, some mechanically sensitive afferents may also respond to chemical stimuli and vice versa.21

Although our handgrip paradigm produced clear effects on SSNA, their functional consequences are not clear. In the thermoneutral environment in which the study was performed, we noted no effect on forearm skin blood flow. However, we cannot exclude a possible effect on sudomotor activity that could play a role in thermoregulation during intense exercise.

In conclusion, our data demonstrate that intermittent SHG exercise leads to a prompt activation of sympathetic nerve traffic directed to skin. This neural signal does not precede but rather closely follows the development of muscle tension. Brief transient discharges of sympathetic nerve traffic directed to skin can also be elicited by an arousal-like effect of virtual handgrip or by mechanical stimuli independent of muscle contraction. Ischemic handgrip exercise is associated with a substantial pressor response and a parallel increase of SSNA, suggesting that stimulation of metabolically sensitive muscle afferents is capable of modulating sympathetic nerve activity directed to skin. The functional role of sympathetic activation directed to the skin during intense forearm exercise remains unclear.

Acknowledgments

This study was supported by American Heart Association grant 9950426N (Dr Leuenberger) and National Institutes of Health (NIH) grants R01-HL-12227 and K24-HL-04011 (Dr Sinoway) and NIH/National Center for Research Resources grant M01-RR-10732. The authors thank Jennie Stoner for her expert secretarial assistance.

References


Control of Skin Sympathetic Nerve Activity During Intermittent Static Handgrip Exercise
Urs A. Leuenberger, Sogol Mostoufi-Moab, Michael Herr, Kristen Gray, Allen Kunselman and Lawrence I. Sinoway

_Circulation_. 2003;108:2329-2335; originally published online November 3, 2003;
doi: 10.1161/01.CIR.0000093280.40118.30
_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/108/19/2329

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/