Original Contributions

Normotensive Blacks Have Heightened Sympathetic Response to Cold Pressor Test

David A. Calhoun, Muthoka L. Mutinga, Angela S. Collins, J. Michael Wyss, Suzanne Oparil

The purpose of this study was to compare sympathetic nerve activity responses to the cold pressor test in black and white normotensive subjects. We recorded muscle sympathetic nerve activity (microneurography of the peroneal nerve), arterial blood pressure, and heart rate in 9 normotensive American blacks (24±2 years, mean±SEM) and 10 normotensive American whites (28±2 years) at rest and during hand immersion in ice water (cold pressor test). Body weight was not different in the two groups (72.4±3.7 versus 74.1±3.8 kg, black versus white subjects). During supine rest, mean arterial pressure (92±2 versus 93±3 mm Hg, black versus white), heart rate (66±4 versus 62±3 beats per minute, black versus white), and muscle sympathetic nerve burst frequency (12±2 versus 17±3 bursts per minute, black versus white) were not different in the two groups. During the cold pressor test, mean arterial pressure, heart rate, and muscle sympathetic nerve activity increased from supine rest in both groups. The magnitudes of increases in mean arterial pressure and total minute muscle sympathetic nerve activity were significantly greater in blacks than whites (33.5±3 versus 22.4±3 mm Hg and 416±24% versus 243±31% of control, respectively, black versus white, P<.05). The increases in heart rate were not significantly different for the two groups. These data suggest that the enhanced pressor response to cold stress observed in normotensive blacks is attributable to greater increases in peripheral sympathetic nerve activity. This heightened sympathetic response to stress may predispose blacks to the development of hypertension. (Hypertension. 1993;22:801-805.)

KEY WORDS • blacks • sympathetic nervous system • ethnic differences

A dolescent and young adult American blacks consistently have been shown to have greater pressor responses to certain laboratory stressors than age-matched American whites.1-5 This heightened pressor response to stress may be important in the pathogenesis of hypertension, because it characterizes the early stages of hypertension.6-9 The mechanism of the heightened pressor response to stress in blacks remains speculative. The greater stress-induced increase in blood pressure observed in blacks is accompanied by greater increases in peripheral sympathetic nervous system reactivity. However, indirect assessments of sympathetic nervous system activity, such as measurements of venous plasma norepinephrine levels, have not suggested significant racial differences in sympathetic nervous system reactivity.10-12 Microneurographic measurement of efferent sympathetic nerve traffic to skeletal muscle is now commonly used to evaluate peripheral sympathetic nervous system activity in humans.12 Because muscle sympathetic nervous system activity (MSNA) is directly measured from peripheral nerves, microneurography is more sensitive and specific than traditional indirect assessments of sympathetic nervous system activity.13 With the use of microneurography, small but significant perturbations in sympathetic nervous system activity can be demonstrated that may not be seen with conventional assessments of sympathetic nervous system activity, such as measurement of venous plasma norepinephrine levels.14

The primary purpose of the present study was to determine if the greater pressor response to cold stress manifested by blacks is related to greater sympathetic neural reactivity. A secondary aim was to determine if resting levels of sympathetic nervous activity are different in normotensive blacks and whites. To address these aims, we made microneurographic measurements of MSNA and recorded arterial blood pressure and heart rate in age- and weight-matched normotensive black and white subjects during supine rest and in response to the cold pressor test.

Methods

Subjects

Nineteen healthy volunteer subjects participated in this study after providing written informed consent. Subjects were normotensive, free of known cardiopulmonary and/or neuropathic disease, and were not taking medications known to affect cardiovascular or sympathetic nerve function. Nine subjects (5 men, 4 women) were American blacks, and 10 subjects (7 men, 3 women) were American whites. Race was determined by self-report. Eight of the black subjects and 8 of the white subjects were students. The level of completed

Received December 21, 1992; accepted in revised form May 5, 1993.

From the Vascular Biology and Hypertension Program, Division of Cardiovascular Disease, Departments of Medicine and Cell Biology, University of Alabama at Birmingham.

Correspondence to David A. Calhoun, MD, Vascular Biology and Hypertension Program, 520 ZRB, UAB Station, University of Alabama at Birmingham, Birmingham, AL 35294.
edcation was similar in the two groups (12.9 ±0.4 versus 13.6 ±0.4 years, black versus white). Seven of the black and 1 of the white subjects reported a family history of hypertension. All experimental procedures and protocols performed by these subjects had received prior approval from the Institutional Committee for Research on Human Subjects of the University of Alabama at Birmingham.

**Experimental Measurements**

Muscle sympathetic nerve activity. Multiunit recordings of MSNA were obtained from the right peroneal nerve at the fibular head using microneurography according to the technique of Vallbo et al. The neural activity was amplified, filtered (bandwidth, 700 to 2000 Hz), full-wave rectified, and integrated (time constant, 0.1 second) to obtain a mean voltage display of sympathetic activity. A recording of efferent sympathetic activity was accepted when the neurograms revealed spontaneous, pulse-synchronous bursts of neural activity, with the largest bursts showing a minimal signal-to-noise ratio of 3:1. The presence of muscle efferent sympathetic activity was documented by a biphasic response to the Valsalva maneuver and the absence of response to arousal stimuli, such as a skin pinch.

Arterial blood pressure and heart rate. Arterial blood pressure was recorded continuously in the left hand by the Finapres device (Finapres 2300, Ohmeda, Louisville, Colo.). Accuracy of the Finapres was confirmed by conventional mercury sphygmomanometry in the left arm (brachial artery) of each subject. Heart rate was determined from electrocardiographic tracings.

Breathing. Qualitative measurements of the rate and depth of breathing were obtained from a pneumograph (Stethograph, Harvard Apparatus, South Natick, Mass) positioned around the abdomen. The pneumograph recordings were made to ensure that abnormal breathing maneuvers (ie, Valsalva) that are known to influence MSNA were not performed by the subjects during the experimental protocols.

Cold pressor test. A cold pressor test was performed by immersing the subject's right hand to the wrist in ice water (1°C). The cold pressor test has been shown to significantly increase MSNA.

Twenty-four-hour urinary sodium and potassium measurements. All subjects were asked to return a 24-hour urine collection. Adequacy of the urine collection was determined according to predicted creatinine excretion per kilogram per 24 hours. Sodium and potassium concentrations were determined by flame photometry.

**Experimental Protocol**

Subjects were instrumented when supine in a quiet, semi-dark room. After an acceptable signal was obtained, baseline MSNA was recorded for at least 10 minutes to ensure stability of the neurogram. This was followed by a 5-minute control period. The cold pressor test was then performed for 2 minutes. Throughout the experimental session, subjects were reminded to remain relaxed and breathe normally; the pneumograph recordings were examined to exclude any abnormal respiratory maneuvers such as the Valsalva strain. All variables were recorded continuously on an electrostatic recorder (WindoGraf, Gould Instruments, Cleveland, Ohio) for subsequent analysis.

**Data Analysis**

Sympathetic bursts were determined by inspection of the filtered voltage neurograms. A deflection on the mean voltage display was counted as a burst if it had a minimal signal-to-noise ratio of 2:1 and was pulse-synchronous (with an interburst interval equal to or a multiple of the RR interval). Nerve traffic was expressed as the number of bursts of sympathetic activity per minute and as total minute activity (calculated as the product of bursts per minute and average burst amplitude per minute and presented in arbitrary units). Because burst amplitude is influenced by the proximity of the recording electrode to the sympathetic neurons and the gain of the amplifier, total minute activity cannot be used for intersubject (or intergroup) comparisons of absolute muscle sympathetic activity. However, it can be used to quantify changes in MSNA in response to perturbation within a subject. Nerve recordings were analyzed with the investigator blinded to the identity of the subjects. Values for all variables were averaged for each of the 5-minute control periods. The final 1-minute recording of the cold pressor test was used to determine changes from control.

Age, weight, and urinary electrolytes for the two groups were compared by t test. Hemodynamic and MSNA values during the control period and changes from control during the cold pressor test for the two subject groups were first compared by one-way analysis of variance (ANOVA). The effects of gender and race comparisons of absolute muscle sympathetic activity. However, it can be used to quantify changes in MSNA in response to perturbation within a subject. Nerve recordings were analyzed with the investigator blinded to the identity of the subjects. Values for all variables were averaged for each of the 5-minute control periods. The final 1-minute recording of the cold pressor test was used to determine changes from control.

**Results**

Baseline Characteristics

Age and body weight were not different in the two groups (24 ±2 versus 28±2 years and 72±3.7 versus 74.1±3.8 kg, black versus white subjects). Group data during supine rest (control period) before cold stress are shown in the Table; there were no differences between the two groups for any variable. Burst frequency at rest was not significantly different in the black versus white subjects but tended to be lower in blacks (12±2 versus 17±3 bursts per minute, black versus white subjects).
O

white). Systolic, diastolic, and mean arterial pressures and heart rate at rest were not different in the two groups.

**Cold Pressor Test**

Hand immersion in ice water produced increases in systolic, diastolic, and mean arterial pressures in both groups (Fig 1). The magnitude of the cold-induced changes was significantly greater in the black subjects than in the white subjects for each of the blood pressure parameters. The increases in systolic, diastolic, and mean arterial blood pressures were 33.5±3, 27.8±2, and 29.6±2 mm Hg, respectively, for the black subjects versus 22.4±3, 17.4±3, and 19.5±3 mm Hg, respectively, for the white subjects. Burst frequency and total minute nerve activity increased above control levels in response to hand immersion in both groups (Figs 2 and 3). The magnitudes of change were significantly greater in the black subjects. The increases in burst frequency were 24.9±3 bursts per minute in blacks versus 15.6±3 bursts per minute in whites (P<.05). Total minute nerve activity increased by 416±24% of control in blacks versus 243±31% of control in whites (P<.05). Heart rate increased above control levels in response to hand immersion in both groups (Fig 2), but the magnitude of the change was not different in the black and white subjects. Responses to cold stress were not significantly affected by gender in either blacks or whites.

**Urinary Sodium and Potassium Excretion**

Seven of the black subjects and eight of the white subjects returned adequate 24-hour urine collections. The sodium (177±11 versus 179±17 mmol/24 h, black versus white) and potassium (64±7 versus 69±7 mmol/24 h, black versus white) excretions per 24 hours were similar in the black and white subjects.

**Discussion**

This study provides the first direct comparison of sympathetic nerve activity in black and white subjects. The principal new finding is that the magnitude of the sympathetic neural response to the cold pressor test is significantly greater in normotensive American blacks than in normotensive American whites. Sympathetic nerve activity during supine rest tended to be lower in blacks but was not statistically different from whites. A heightened sympathetic response to stress may contrib-
ute to the higher incidence of hypertension in American blacks compared with whites.

Hypertension is more prevalent, more severe, and more often complicated by end-organ damage in American blacks than among whites. The reasons for the increased prevalence and severity of hypertension in blacks are unknown, but heightened sympathetic nervous system reactivity is hypothesized to play an important role. This hypothesis is consistent with the broader hypothesis that proposes a neurogenic basis for hypertension in general. As stated by Folkow, repeated activation of the sympathetic nervous system through exposure to environmental stress induces vascular hypertrophy, which causes an increase in vascular resistance and a consequent elevation of blood pressure. Blacks, because they seem to react more vigorously to stress than whites, would be particularly prone to stress-induced increases in blood pressure.

A major weakness in the hypothesis that increased sympathetic reactivity predisposes blacks to developing hypertension has been the inability to demonstrate that sympathetic responses to stress are different in blacks versus whites. Racial comparisons of sympathetic nerve reactivity to stress have been infrequently reported and, when reported, have been generally limited to indirect assessments of sympathetic nerve activity, such as measurement of venous plasma norepinephrine levels. Although plasma norepinephrine concentrations have been shown to consistently reflect sympathetic nervous system activity during interventions or in disease states that produce large increases in sympathetic nerve activity, including upright tilting, exercise, and congestive heart failure, changes in norepinephrine levels often fail to identify smaller changes in sympathetic activity, as in response to the cold pressor test. The failure of venous plasma norepinephrine levels to accurately reflect small changes in sympathetic nerve activity is not surprising given that (1) circulating norepinephrine represents only a small portion of the norepinephrine released from sympathetic nerve terminals, (2) neural release and uptake of norepinephrine vary from person to person, and (3) regional levels of norepinephrine vary according to the contribution of different vascular beds. By directly measuring MSNA, microneurography overcomes many of the limitations of using plasma norepinephrine levels as an index of sympathetic nerve activity.

The results of the present study, which used the cold pressor test to stress subjects, are consistent with previous findings that young adult blacks have greater pressor responses to stress than whites. Other investigators have shown similar results with other types of stressors, including the cold face stimulus, mental stress, and physical exercise. Subjects prone to developing hypertension and subjects with borderline hypertension have also been shown to have heightened pressor responses to stress, suggesting that enhanced vascular reactivity may play an important role in the etiology of hypertension.

The novel observation of the present study is that the greater stress-induced pressor response observed in blacks is correlated to enhanced sympathetic reactivity. Previous investigators had shown that the greater increases in blood pressure manifested by blacks in response to stress occurred in association with greater increases in peripheral vascular resistance. However, the mechanism of the increased peripheral vascular resistance remained unknown. Measurement of venous plasma norepinephrine levels had not indicated any significant racial differences in sympathetic reactivity, suggesting that the greater increase in peripheral vascular resistance in blacks may be due to structural alterations in the vessel wall or to differences in the release of and/or sensitivity to humoral vaspressor factors. We conclude, however, that the greater pressor response to cold stress manifested by blacks is attributable, at least in part, to a greater increase in sympathetic nerve activity. Whether blacks also manifest heightened sympathetic reactivity to other types of stressors, such as mental stress or physical exercise, needs to be determined.

Several factors, such as socioeconomic status, dietary NaCl ingestion, gender, and family history of hypertension, are clearly related to the development of hypertension and perhaps to the vascular response to stress. In the present study, the subjects were predominantly students, with the overall educational level being similar in the two racial groups. Dietary sodium and potassium ingestion, as assessed by 24-hour urinary excretion, was similar in the black and white subjects. Interactions of gender and race were not significant. In the present study, most of the black subjects had a positive family history of hypertension, whereas most of the white subjects had a negative family history. Some investigators have found family history of hypertension to be related to stress responses, whereas other investigators have not. Studies are ongoing in this laboratory to determine if the greater sympathetic response to cold stress observed in blacks in the present study is related to a family history of hypertension.

Acknowledgments

This work was supported by grants HL-07457 and HL-37722 from the National Heart, Lung, and Blood Institute, National Institutes of Health, Bethesda, Md. Dr Calhoun is a recipient of a Physician-Scientist Award (HL-02568) from the National Heart, Lung, and Blood Institute and is supported by a Grant-in-Aid from the Alabama Affiliate of the American Heart Association.

References

Normotensive blacks have heightened sympathetic response to cold pressor test.
D A Calhoun, M L Mutinga, A S Collins, J M Wyss and S Oparil

Hypertension. 1993;22:801-805
doi: 10.1161/01.HYP.22.6.801

Hypertension is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 1993 American Heart Association, Inc. All rights reserved.
Print ISSN: 0194-911X. Online ISSN: 1524-4563

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://hyper.ahajournals.org/content/22/6/801

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in Hypertension 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 Hypertension is online at:
http://hyper.ahajournals.org/subscriptions/