

Carotid baroreceptor stimulation alters cutaneous vascular conductance during whole-body heating in humans

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Prior studies investigating carotid baroreflex control of the cutaneous vasculature have yielded mixed findings. However, previously used methodological and analytical techniques may limit the ability to detect carotid baroreflex-mediated changes in cutaneous vascular conductance (CVC). The aim of this study was to test the hypothesis that dynamic carotid baroreceptor stimulation (i.e. 5 s trials) using neck pressure (NP, simulated carotid hypotension) and neck suction (NS, simulated carotid hypertension) will decrease and increase CVC, respectively, during normothermic and whole-body heating conditions in resting humans. Data were obtained from nine subjects (age, 31 ± 2 year). The ratio of forearm skin blood flux (laser-Doppler flowmetry) and arterial blood pressure (Finapres) was used as an index of CVC. Multiple 5 s trials of NP ($+40_{\text{Torr}}$) and NS (-60_{Torr}), as well as breath-hold/airflow control trials, were applied during end-expiratory breath-holds while subjects were normothermic and heat stressed (change in core temperature $\sim 0.75^{\circ}\text{C}$). CVC responses to each NP and NS trial were averaged into 1 s intervals during the following periods: 3 s prestimulus, 5 s during stimulus, and 5 s poststimulus. Peak CVC responses (3 s average) to NP and NS were compared to prestimulus values using paired *t* test. During normothermia, NP decreased CVC by 0.032 ± 0.007 arbitrary units (a.u.) mmHg^{-1} ; ($P < 0.05$); however, breath-hold/airflow control trials resulted in similar decreases in CVC. NS did not change CVC ($\Delta = 0.002 \pm 0.005$ a.u. mmHg^{-1} ; $P = 0.63$). During whole-body heating, NP decreased CVC (by 0.16 ± 0.04 a.u. mmHg^{-1} ; $P < 0.05$), whereas NS increased CVC by 0.07 ± 0.03 a.u. mmHg^{-1} ; ($P < 0.05$). Furthermore, these changes were greater than, or directionally different from, the breath-hold/airflow control trials. These findings indicate that carotid baroreceptor stimulation elicits dynamic changes in CVC and that these changes are more apparent during whole-body heating.

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In humans, whole-body heat stress redistributes upwards of 50% of cardiac output to the cutaneous vasculature (Rowell *et al.* 1969). In addition, orthostatic tolerance is reduced during whole-body heat stress under a variety of gravitational stressors (Lind *et al.* 1968; Allan & Crossley, 1972; Wilson *et al.* 2002). During the aforementioned heat stress condition, the cutaneous vasculature represents the primary contributing vascular bed to total peripheral resistance and would, therefore theoretically, be an important target for baroreflex control of arterial blood pressure. While the arterial baroreflex is the primary short-term regulator of arterial blood pressure, studies investigating arterial baroreflex control of the cutaneous vasculature during both normothermic and heat stress conditions have yielded mixed findings.

In 1970, Beiser *et al.* reported a decrease in cutaneous vascular resistance (the reciprocal of conductance) during normothermic conditions in response to loading the carotid baroreceptors using the neck chamber technique (Beiser *et al.* 1970). However, their use of strain-gauge photoplethysmography, which does not discriminate between the blood flow responses in the skin and the underlying skeletal muscle, as well as the nature of the analysis used to determine baroreflex control of the cutaneous vasculature (i.e. subtraction of the response between a control forearm and an arm whose cutaneous vasculature was precontracted using iontophoresis of adrenaline (epinephrine)) may have limited such interpretation. Furthermore, Crandall *et al.* (1996) did not observe changes in cutaneous vascular

conductance (CVC) in response to carotid baroreceptor unloading using the neck chamber technique during both normothermic and whole-body heating conditions. Furthermore, findings by Wallin *et al.* (1975) and Wilson *et al.* (2001) have indicated an absence of isolated carotid baroreflex and simultaneous carotid and aortic baroreflex control of skin sympathetic nerve activity (sSNA), respectively. Wallin *et al.* (1975) measured the integrated sSNA response to direct, electrical stimulation of the carotid sinus nerve (simulated hypertension) during normothermia, and observed no reproducible changes in sSNA. However, Wallin *et al.* (1975) did not measure skin blood flow responses, and therefore interpretation of those findings as it pertains to carotid baroreflex control of the cutaneous vasculature remains limited. Wilson *et al.* (2001) reported that arterial baroreceptor stimulation with bolus sodium nitroprusside followed by bolus phenylephrine infusions (i.e. the modified Oxford technique), as well as sustained reductions in blood pressure with steady-state nitroprusside infusion, did not significantly change integrated sSNA. Due to the direct effect of the infused drugs on the cutaneous vasculature, Wilson *et al.* (2001) only examined the integrated neural response to this pharmacological perturbation, and thus did not assess arterial baroreflex control of the cutaneous vasculature (i.e. skin blood flow, or CVC). Furthermore, changes in CVC have not always been accompanied by changes in sSNA (Cui *et al.* 2004), further complicating the interpretation of such findings regarding arterial baroreflex control of the cutaneous vasculature. Finally, any potential dynamic response to carotid baroreceptor stimulation (i.e. of the order of seconds) was not assessed in the aforementioned study by Crandall *et al.* (1996). Despite the lack of a change in mean CVC in that study, it is possible that the carotid baroreceptor stimulation used by Crandall *et al.* (i.e. repeated 500 ms pulses) resulted in dynamic fluctuations in CVC that were perhaps masked due to averaging of the responses over a 1 min period, and/or the closed-loop nature of the stimulus (Crandall *et al.* 1996).

In the current investigation, we used the neck chamber technique to selectively load and unload the carotid baroreceptors to test the hypothesis that open-loop carotid baroreceptor stimulation with 5 s trials of neck pressure (NP) and neck suction (NS) will dynamically modulate CVC during normothermia and whole-body heating in resting humans.

Methods

Subjects

Nine subjects (4 men and 5 women) voluntarily participated in the present investigation (age, 31 ± 2 years; height, 166 ± 11 cm; weight, 69 ± 1 kg; mean \pm s.e.m.).

All procedures conformed to the standards set by the Declaration of Helsinki. Each subject signed an informed consent that was approved by the institutional review boards of the University of Texas South-western Medical Center and Presbyterian Hospital of Dallas. Prior to participation, all subjects were familiarized with the testing protocols. Subjects were healthy, non-smokers, free of known cardiovascular and respiratory diseases, and were not using prescription or over-the-counter medications. Subjects were advised to not consume alcohol for 24 h before any of the scheduled experiments. Subjects were also asked to refrain from the consumption of caffeinated beverages from 12 h before the scheduled experiments.

Instrumentation

Subjects were dressed in a tube-lined perfusion suit enabling the control of skin and core temperature (T_{core}) via changing the temperature of the water perfusing the suit. T_{core} was measured using a telemetry temperature pill swallowed by subjects at least 1 h before any data collection (normothermic T_{core} data were identified prior to heat stress which was ~ 2 h post-ingestion). Whole-body skin temperature (T_{sk}) was measured from the electrical average of six thermocouples (Taylor *et al.* 1989) fixed to the skin with porous adhesive tape. A separate thermocouple was fixed to the skin of the neck under the neck chamber to measure neck skin temperature (T_{neck}) during the baroreceptor perturbation trials. Non-invasive measures of arterial blood pressure were taken continuously using finger cuff photoplethysmography (Finapres, Ohmeda, Louisville, CO, USA). Arterial blood pressure was also determined by auscultation of the brachial artery (SunTech, Meical Instruments, Raleigh, NC, USA). Diastolic blood pressures determined by auscultation were used to calibrate diastolic blood pressures determined by finger cuff photoplethysmography. This was done by determining the average diastolic blood pressure over the ~ 30 s period during which the auscultation-derived diastolic pressures were collected, and correcting for the offset between the two measures (i.e. adjusting the finger diastolic pressures to the auscultation-derived pressures). Heart rate was collected from an electrocardiogram (ECG) signal (Agilent, Munich, Germany) interfaced with a cardiometer (1000 Hz sampling rate, CWE, Ardmore, PA, USA). Two laser Doppler flowmeter probes (Perimed, North Royalton, OH, USA) were placed on dorsal forearm skin midway between the wrist and the elbow. The values from these flow probes were averaged, with the resulting value used for the ensuing data and statistical analysis. Subjects were fitted with a malleable lead neck chamber covering the anterior two-thirds of the neck for baroreflex testing.

Experimental protocol

During normothermic testing, water at 34°C was perfused through the tube-lined suit for approximately 30 min before baroreceptor perturbations. Four trials of NP followed by four trials of NS were performed in randomized order for 16 total trials during normothermia (i.e. 4 NP, 4 NS, 4 NP, 4 NS, or 4 NS, 4 NP, 4 NS, 4 NP). A minimum of 45 s was allowed to pass between each trial to enable physiological variables to return to prestimulus values. Following approximately 30 min of baroreflex testing during normothermia, whole-body heating was performed by perfusing water at 44–48°C through the tube-lined suit. After an increase in T_{core} of approximately 0.7°C was observed, the temperature of the water was decreased (42–44°C) to attenuate further increases in T_{core} . After 5–10 min at these reduced water temperatures, baroreflex testing was repeated similar to normothermia. Under both thermal conditions, following baroreflex testing, neck chamber control trials were also performed to determine the effect of baroreceptor-independent stimulation associated with the application of the neck chamber technique and breath holding (see below).

Measurements and procedures

Testing was performed with subjects in the supine position. Cardiovascular variables were monitored and recorded on a computer equipped with customized software (Necsuc3) that collects and records data triggered from the R-wave of the ECG, as well as a second computer equipped with an online data acquisition program (Biopac, CA, USA). Carotid baroreflex control of mean arterial pressure (MAP) and CVC was assessed by applying single 5 s pulses of NP (+40_{Torr}) and NS (−60_{Torr}) as described by Potts *et al.* (1993). Briefly, NP and NS trials were performed during an approximately 15 s breath-hold at normal end-expiration, in order to minimize the respiratory modulation of HR and MAP (Eckberg *et al.* 1980). Peak responses for MAP were determined as the greatest change over a 3 s period in response to the application of NP or NS and these peak changes from repeated trials were averaged to provide a mean response for each subject. CVC was calculated using the following formula:

$$\begin{aligned} \text{CVC (arbitrary units (a.u.) mmHg}^{-1}) \\ = \text{skin blood flux (a.u.)/MAP (mmHg)} \end{aligned}$$

To determine the effect of the breath-hold, as well as changes in T_{neck} associated with the application of NP and NS, breath-hold/airflow control trials were also performed. During these control trials, the chamber was removed from the subject's neck and the subject held his/her breath for a similar duration, relative to the experimental trial, while the flow of air, normally used to generate chamber pressure (i.e. before the chamber was

removed) was directed on the neck. The flow to the neck was adjusted in an effort to achieve similar changes in T_{neck} to those observed during actual trials of NP and NS. Care was taken to prevent air flow above the neck (i.e. facial region).

Data analysis

Steady-state haemodynamic and thermal variables were determined using 1 min averages during normothermic and heat-stress conditions. Responses to carotid baroreceptor stimulation were determined using two different analyses. For both analyses, data from each trial were averaged in 1 s intervals, beginning with 3 s prestimulus followed by 5 s during the carotid stimulation and 5 s following carotid stimulation (i.e. 13 s total per trial).

The traditional approach was used by determining the 3 s interval representing the largest change in CVC relative to the respective prestimulus value for each separate trial of NP and NS. The average of these peak responses was then determined for both NP and NS for each subject within each thermal condition.

A more conservative approach was performed by first averaging the second-by-second responses to all NP, as well as to all NS trials for each subject within each thermal condition. After all NP and NS trials were averaged, the 3 s interval of the *averaged* data with the largest change in CVC was then compared to the 3 s prestimulus period. This analysis resulted in slightly smaller average responses to NP and NS during both thermal conditions relative to the traditional approach. However, peak changes determined using this more conservative analysis minimized the influence of baroreflex-independent fluctuations in skin blood flow on the peak responses.

Breath-hold/airflow control trials were analysed using the more conservative approach (described above). Using the second-by-second analysis, peak CVC responses observed during NP and NS trials (conservative analysis) were then compared to breath-hold/airflow control trials at similar time points for each subject. Therefore, the comparison between the control trials and 'actual' trials was based on the time of the peak CVC response, as determined in relation to an individual's 'conservative' analysis. That is, if an individual's peak response to NP, as determined using conservative analysis, occurred during seconds 6 to 8, the comparison to the breath-hold/airflow control trials was made during those same time points. However, the same subject's peak response to NS may have occurred during seconds 5 to 7. Therefore, the comparison to the control trials was made during those time points. Breath-hold/airflow control trials were performed on eight subjects during normothermia and seven subjects during whole-body heating. The averaged peak change (3 s) in

Table 1. Cardiovascular and thermal variables

	Normothermia	Whole-body heating
HR (beats min ⁻¹)	60.9 ± 4.4	76.3 ± 5.3*
SBP (mmHg)	114.2 ± 3.2	116.1 ± 4.3
DBP (mmHg)	67.8 ± 2.3	63.7 ± 1.9*
MAP (mmHg)	83.3 ± 2.4	81.2 ± 2.5
<i>T</i> _{core} (°C)	37.0 ± 0.1	37.8 ± 0.1*
<i>T</i> _{sk} (°C)	34.7 ± 0.1	37.3 ± 0.2*
Skin blood flux (a.u.)	18.7 ± 1.4	66.8 ± 11.9*
CVC (a.u. mmHg ⁻¹)	0.23 ± 0.02	0.81 ± 0.14*

HR, heart rate; SBP, systolic blood pressure; DBP, diastolic blood pressure; MAP, mean arterial pressure; *T*_{core}, core temperature; *T*_{sk}, skin temperature; CVC, cutaneous vascular conductance; *significantly different from normothermia ($P < 0.05$)

*T*_{neck} in response to NP and NS during normothermic conditions were ~0.9°C and 0.4°C, respectively, while the breath-hold/airflow control trials applied during normothermic conditions changed *T*_{neck} by ~0.9°C. The averaged peak change (3 s) in *T*_{neck} in response to NP and NS during whole-body heating were ~1.5°C and 1.0°C, respectively, while the breath-hold/airflow control trials applied during whole-body heating changed *T*_{neck} by ~1.6°C.

Statistical analysis

Comparisons of steady-state physiological variables between normothermia and whole-body heating were made using paired *t* tests. Comparisons of CVC in response to baroreflex testing were made within each thermal

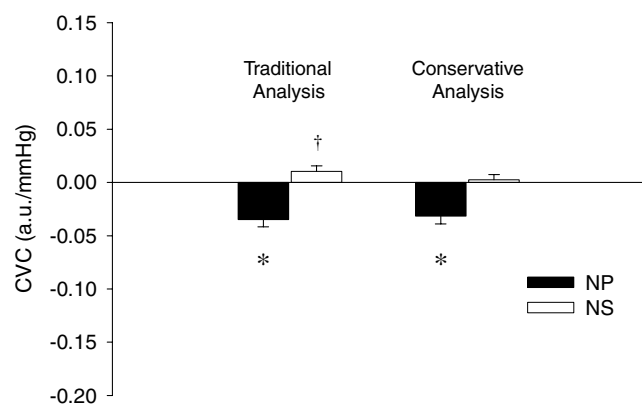


Figure 1. Carotid baroreflex-mediated changes in cutaneous vascular conductance (CVC) in response to neck pressure (NP, filled bars) and neck suction (NS, open bars) during normothermia

Data are presented as a change from baseline (0.23 a.u. mmHg⁻¹). Traditional analysis and conservative analysis (see Methods) of the same data are presented as means ± s.e.m. *Significantly different from prestimulus ($P < 0.05$). †Compared to prestimulus ($P = 0.079$). $n = 9$

condition using paired *t* tests. Statistical significance was set at $P < 0.05$.

Results

Cardiovascular and thermal variables during normothermia and whole-body heating

Steady-state cardiovascular and thermal data during normothermia and whole-body heating are presented in Table 1. Skin blood flux increased from 18.7 ± 1.4 a.u. during normothermia to 66.8 ± 11.9 a.u. during whole-body heating ($P < 0.05$), resulting in an approximate four-fold increase in CVC (0.81 ± 0.14 a.u. mmHg⁻¹) compared to normothermia (0.23 ± 0.02 a.u. mmHg⁻¹; $P < 0.05$).

Carotid baroreceptor stimulation during normothermia

Traditional analysis. When the peak responses to each trial of NP (+40_{Torr}) were determined for each subject, NP decreased CVC by 0.035 ± 0.007 a.u. mmHg⁻¹ from baseline ($P < 0.05$) (see Fig. 1). This equated to a 16.9 ± 2.6% decrease in CVC from baseline. The peak responses to each separate trial of NS (-60_{Torr}) tended to increase CVC 0.010 ± 0.005 a.u. mmHg⁻¹ from baseline ($P = 0.079$), resulting in 5.1 ± 2.8% increase from baseline.

Conservative analysis. When all trials to NP (+40_{Torr}) within a subject were first averaged, the application of NP decreased CVC by 0.032 ± 0.007 a.u. mmHg⁻¹ from baseline ($P < 0.05$). This equated to a 15.3 ± 2.9% reduction in CVC. However, using this analytical method for NS, CVC did not change (change (Δ) = 0.002 ± 0.005 a.u. mmHg⁻¹) from baseline ($P = 0.63$; see Fig. 1).

Breath-hold/airflow control trial analysis. At the time of the peak CVC response during NP trials, the breath-hold/airflow control trial reduced CVC by 0.030 ± 0.007 a.u. mmHg⁻¹ ($P < 0.05$) from prestimulus values. Peak decreases in CVC in response to actual NP trials from baseline ($\Delta = -0.031 \pm 0.008$ a.u. mmHg⁻¹) were not different from changes in CVC observed during the breath-hold/airflow control trials ($\Delta = -0.030 \pm 0.007$ a.u. mmHg⁻¹; $P > 0.05$).

At the time of the peak CVC response during NS trials, breath-hold/airflow control trials tended to decrease CVC by 0.013 ± 0.007 a.u. mmHg⁻¹ ($P = 0.11$) from prestimulus values. The difference in the change in CVC between NS and breath-hold/airflow control trials

($\Delta = 0.013 \pm 0.006$ a.u. mmHg^{-1}) tended to be different ($P = 0.068$), see Fig. 2.

Carotid baroreceptor stimulation during whole-body heating

Traditional analysis. When peak responses to each separate trial of NP ($+40_{\text{Torr}}$) were determined for each subject, NP decreased CVC by 0.168 ± 0.028 a.u. mmHg^{-1} from baseline ($P < 0.05$). This response equated to a $15.8 \pm 3.4\%$ reduction in CVC. Figure 3 depicts the CVC response to separate trials of NP, as well as the averaged response (see Conservative analysis below) in one subject. The peak responses to each separate trial of NS (-60_{Torr}) increased CVC by 0.097 ± 0.030 a.u. mmHg^{-1} from baseline ($P < 0.05$). This response equated to a $9.0 \pm 3.0\%$ increase in CVC (see Fig. 4).

Conservative analysis. When all trials to NP ($+40_{\text{Torr}}$) within a subject were first averaged before identifying the peak response, NP decreased CVC by 0.145 ± 0.030 a.u. mmHg^{-1} from baseline ($P < 0.05$). This response equated to a $13.7 \pm 3.8\%$ reduction in CVC. Using this analytical technique for NS, this perturbation increased CVC ($\Delta = 0.071 \pm 0.027$ a.u. mmHg^{-1} from baseline, $P < 0.05$). This response equated to a $6.7 \pm 2.7\%$ increase from baseline CVC (see Fig. 4).

Breath-hold/airflow control trial analysis. During whole-body heating, at the time of the peak CVC response during NP, CVC was not significantly changed during breath-hold/airflow control trials

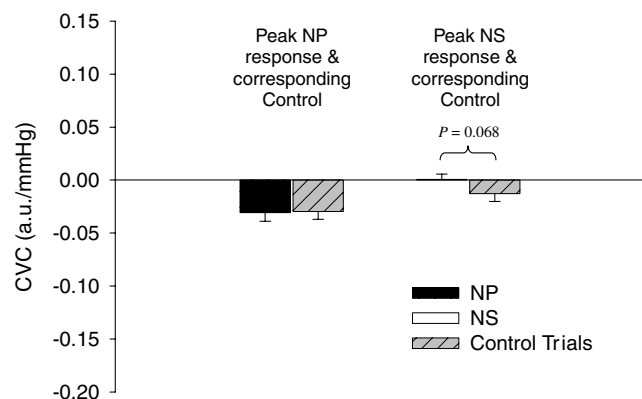


Figure 2. Comparison of neck pressure (NP) and neck suction (NS) trials to breath-hold/airflow control trials (Control Trials) during normothermia

Peak changes in cutaneous vascular conductance (CVC) in response to NP and NS were compared to the respective control trials from data obtained at the same time relative to the carotid stimulus. Data are presented as a change from baseline (0.23 a.u. mmHg^{-1}). $n = 8$. Error bars are s.e.m.

($\Delta = -0.056 \pm 0.039$ a.u. mmHg^{-1} , $P = 0.20$) compared to prestimulus values. Furthermore, the peak decrease in CVC in response to NP ($\Delta = -0.129 \pm 0.029$ a.u. mmHg^{-1}) was greater than this non-significant change in CVC ($\Delta = -0.056 \pm 0.039$ a.u. mmHg^{-1}) during the breath-hold/airflow control trial ($P < 0.05$), see Fig. 5.

At the time of the peak CVC response during the NS trials, CVC was likewise unchanged during breath-hold/airflow control trials ($\Delta = -0.030 \pm 0.039$ a.u. mmHg^{-1} , $P = 0.47$) compared to prestimulus values. Peak increases in CVC to NS ($\Delta = 0.073 \pm 0.031$ a.u. mmHg^{-1}) tended to be greater than decreases in CVC ($\Delta = -0.030 \pm 0.039$ a.u. mmHg^{-1} ; $P = 0.083$) during the breath-hold control trials (note the directional difference).

Discussion

The primary finding of this investigation is that carotid baroreceptor stimulation elicits dynamic changes in CVC during whole-body heating, despite previous contrary findings. In contrast, carotid baroreflex-mediated changes in CVC were less clear under normothermic conditions. The application of either NP or NS reduced T_{neck} during both thermal conditions, and because changes in T_{neck}

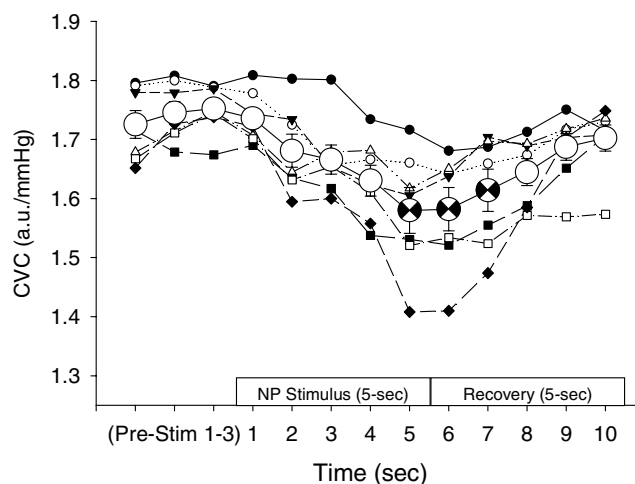


Figure 3. Representative data from one subject during whole-body heating in response to neck pressure (NP)

Cutaneous vascular conductance (CVC) responses to individual trials of NP are represented by the multiple data indicated with smaller symbols. Peak changes from these data were used for the traditional analysis. The averaged CVC response for these individual trials is indicated by the larger, open circle symbols (\pm s.e.m.). The averaged peak data are indicated by the partially filled, larger circles (occurring at seconds 5, 6 and 7); these are the data used for the conservative analysis. The x-axis indicates the 3 s during prestimulus (Pre-Stim 1–3), followed by the 5 s of the neck chamber stimulus and the 5 s of recovery.

may have contributed to the CVC response to the neck chamber stimulation, breath-hold/airflow control trials were also performed. These control trials resulted in slight reductions in CVC during both thermal conditions. While these reductions in CVC may partially account for changes in CVC observed during normothermia in response to NP, decreases in CVC in response to the breath-hold/airflow control trial were markedly smaller than that observed in response to NP during whole-body heating, as well as directionally different from NS. Taken together, these findings indicate that the carotid baroreflex exhibits an efferent limb governing the cutaneous vasculature, although this was only consistently observed while subjects were heat stressed.

Heat stress redistributes cardiac output to the cutaneous vasculature and it has been demonstrated that this redistribution can surpass 50% of the prevailing cardiac output (Rowell *et al.* 1969). Therefore, during heat stress, the cutaneous vasculature has the potential to become an important target for arterial blood pressure control. Despite this, prior evidence suggests that the carotid baroreflex does not control the cutaneous vasculature (Wallin *et al.* 1975; Crandall *et al.* 1996). This lack of control has been proposed as a potential rationale for the reduction in arterial baroreflex control of blood pressure observed during heat stress (Crandall, 2000), as well as the reduced orthostatic tolerance to a number of gravitational stressors (Lind *et al.* 1968; Allan & Crossley, 1972; Wilson *et al.* 2002) observed during heat stress.

In the current investigation, carotid baroreceptor unloading with NP decreased CVC while subjects were

normothermic. However, it is important to note that the application of the breath-hold/airflow control trial resulted in similar decreases in CVC during normothermic conditions. Therefore, despite different responses between NP and NS observed during normothermia (i.e. decrease *versus* no change in CVC, respectively), the interpretation of such findings remains limited. During whole-body heating, NP decreased CVC by $\sim 14\text{--}16\%$ (depending on the analysis used), while NS increased CVC by $\sim 7\text{--}9\%$. These changes were significantly different from those of breath-hold/airflow control trials, indicating carotid baroreflex modulation of the cutaneous vasculature. These findings are in contrast to the findings of Crandall *et al.* (1996), who evaluated carotid baroreflex control of the cutaneous vasculature during normothermic and heat stress conditions using 500 ms pulses of NP during each cardiac cycle over 3 min, and did not observe an effect of NP on CVC. However, the application of the baroreceptor stimulation, as well as the analysis of CVC, were markedly different between these two studies, and may explain the different interpretation of the respective findings. First, the baroreceptor stimulation in the current study was applied using 5 s trials separated by at least 45 s. This stimulation changed carotid transmural pressure for five consecutive seconds, and therefore increased mean carotid sinus pressure by a factor approximately equal to the chamber pressure achieved for each trial (i.e. mean estimated carotid sinus pressure is increased or decreased by the pressure attained in the collar) (Querry *et al.* 2001). However, 500 ms pulses, such as those used by Crandall *et al.* (1996), only briefly change carotid sinus

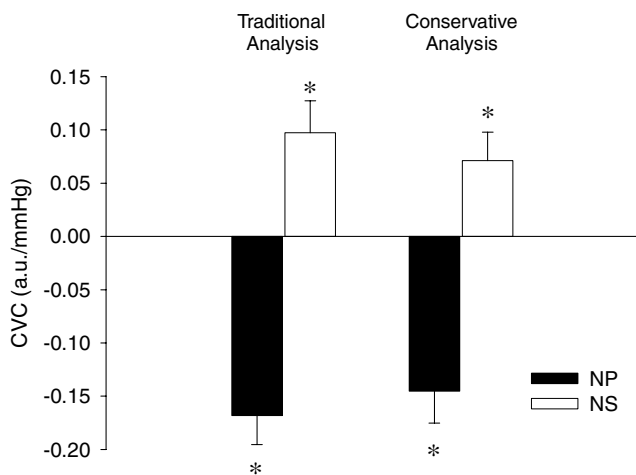


Figure 4. Carotid baroreflex-mediated changes in cutaneous vascular conductance (CVC) in response to neck pressure (NP, filled bars) and neck suction (NS, open bars) during whole-body heating

Data are presented as a change from baseline ($0.81 \text{ a.u. mmHg}^{-1}$). Traditional analysis and conservative analysis (see Methods) of the same data are presented as means \pm S.E.M. *Significantly different from prestimulus ($P < 0.05$). $n = 7$

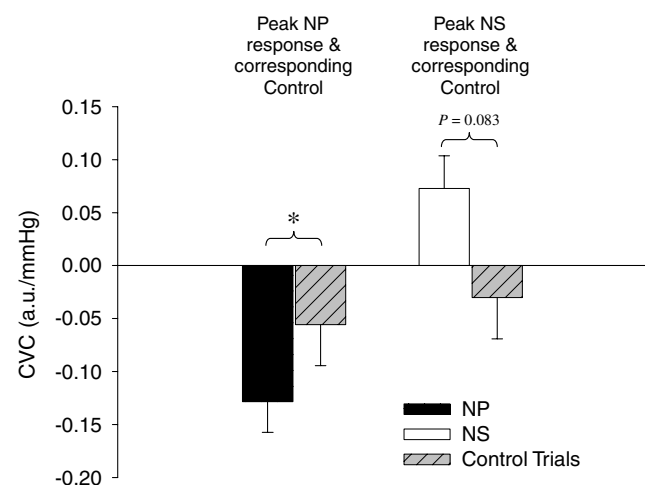


Figure 5. Comparison of neck pressure (NP) and neck suction (NS) trial to breath-hold/airflow control trials (Control Trials) during whole-body heating

Peak changes in cutaneous vascular conductance (CVC) in response to NP and NS were compared to the respective control trials from data obtained at the same time relative to the carotid stimulus. Data are presented as a change from baseline ($0.81 \text{ a.u. mmHg}^{-1}$).

*Significantly different ($P < 0.05$). $n = 7$

pressure, primarily during the systolic phase of the cardiac cycle, and therefore did not alter mean carotid arterial pressure to the same extent as the present protocol. Another possible explanation for the discrepancy between these studies is that the role of the aortic baroreflex between the two studies was probably different. Potts *et al.* (1993) described the analysis of the 5 s stimulation of the carotid baroreceptors as being primarily an open-loop system (i.e. aortic baroreflex independent) due to the brevity of the resulting changes in arterial pressure after carotid stimulation. However, the resulting change in mean arterial pressure reported by Crandall *et al.* (1996) over the 3 min of NP application (i.e. a reported blood pressure increase of ~ 10 mmHg) probably engaged the aortic baroreceptor population, which may have inhibited the effectiveness of carotid baroreceptor hypotension in altering CVC. However, this is purely speculative, as the relative role, or directional control of the cutaneous vasculature by the carotid *versus* aortic baroreflexes has not been examined.

Possibly more relevant to the difference between the findings of the current study and that of Crandall *et al.* (1996) is the nature of the analysis of the responses. In the current study, the dynamic response (i.e. within seconds) to individual trials of carotid baroreceptor stimulation was assessed, while Crandall *et al.* (1996) compared 1 min averages of CVC during NP to prestimulus baseline CVC. The latter technique (Crandall *et al.* 1996) clearly minimizes the ability to detect dynamic baroreflex-mediated fluctuations in CVC that may have occurred during NP. That is, the lack of change in mean CVC, averaged over 1 min, does not negate the notion that carotid baroreceptor stimulation is capable of causing dynamic oscillations in CVC.

Beiser *et al.* (1970) demonstrated a decrease in cutaneous vascular resistance of forearm skin in response to loading the carotid baroreceptors during normothermic conditions. Unlike the findings of Beiser *et al.* (1970), the application of NS did not increase CVC (i.e. decrease resistance) during normothermia in the current investigation. However, Beiser *et al.* (1970) used iontophoresis of adrenaline (epinephrine) to constrict the cutaneous vasculature of one arm prior to carotid baroreceptor stimulation, and compared changes in the 'preconstricted arm' to the other, non-treated arm. Also, Beiser *et al.* (1970) used 2 min trials of NS and strain gauge plethysmography of the forearm, compared to the 5 s NS stimuli and laser Doppler flowmetry indices of skin blood flow used in the current study. These methodological differences may explain differences in CVC responses to NS between these studies. Furthermore, it is possible that, in the current study, the normothermic condition (i.e. the water temperature used) resulted in relatively higher skin temperatures (albeit, only slightly) compared to those found by Beiser *et al.* (1970), and that this subtle

elevation in skin temperature resulted in a withdrawal of sympathetic vasoconstrictor activity directed to the skin. Such an occurrence may have minimized the effectiveness of the hypertensive carotid baroreceptor challenge with NS in response to further withdrawal skin sympathetic activity. However, this hypothesis is purely speculative.

Studies investigating the arterial baroreflex control of sSNA are limited in number. In four subjects, electrical stimulation of the carotid sinus nerve performed by Wallin *et al.* (1975) did not reproducibly change sSNA. However, changes in skin blood flow were not measured by Wallin *et al.* (1975), and reliance on changes in integrated sSNA may limit the interpretation with respect to CVC responses. Furthermore, Wilson *et al.* (2001), observed no change in sSNA in response to pharmacologically induced baroreceptor stimulation. Similar to the work of Wallin

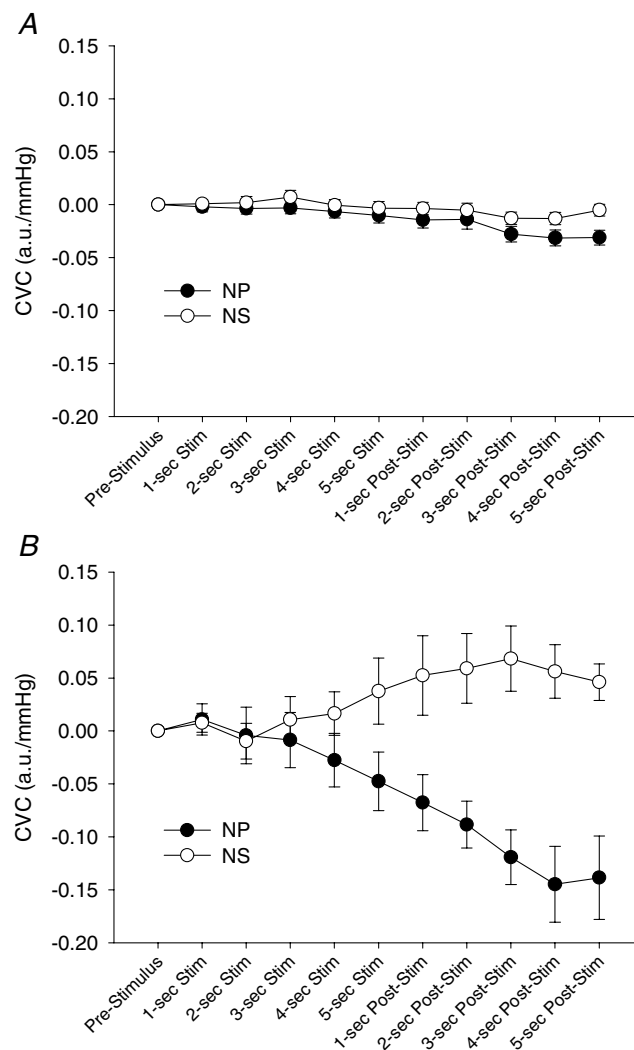


Figure 6. Second-by-second group average responses for cutaneous vascular conductance (CVC) to neck pressure (NP) and neck suction (NS) during normothermia (A) and whole-body heating (B)

et al. (1975), Wilson *et al.* (2001) also did not examine skin blood flow during baroreceptor stimulation due to the direct effect of the infused drugs on the cutaneous vasculature. The cutaneous vasculature is under direct neural control of both an active vasoconstrictor system and an active vasodilator system. It is therefore difficult to distinguish between concomitant changes in these two systems in the sSNA recording, as this may result in no net change in the integrated neural signal at a time when CVC is changing. Perhaps related to this, it has been demonstrated that changes in CVC are not always accompanied by changes in sSNA (Cui *et al.* 2004). This mismatch between sSNA and CVC may further complicate the interpretation of findings regarding arterial baroreflex control of CVC. While analysis of sSNA was not examined in the current study, the end-organ response (i.e. changes in skin blood flow and calculated CVC) was assessed during acute carotid baroreceptor stimulation.

The differential responses to NS between normothermia and whole-body heating in the current study (i.e. the absence during normothermia *versus* the presence during whole-body heating) may be related to steady-state CVC for the respective conditions. That is, when steady-state CVC is high, as during whole-body heating, changes in cutaneous vascular tone are more easily expressed, or identified. Another possible explanation for these differences is that the heat stress itself (i.e. elevated internal temperature, elevated skin temperature, etc) may directly influence the carotid baroreflex, or its associated neural pathways, 'unveiling' reflex control of the skin during a time when neural control of the skin becomes more important for arterial blood pressure regulation. Finally, given that during heat stress CVC is primarily controlled by an active vasodilator system, it may be that carotid baroreceptor control of CVC primarily occurs through modulation of the active vasodilator system whose activity is absent or greatly reduced during normothermia.

The influence of carotid baroreflex-mediated changes in CVC on mean arterial blood pressure is unclear and would depend on the thermal status of the individual. Due to the small fraction of cardiac output directed to the skin during normothermic conditions, carotid baroreflex-mediated changes in CVC will minimally contribute to total vascular conductance and, therefore, arterial blood pressure, in normothermic conditions. However, during heat stress conditions, cardiac output has been reported to more than double, with 100% of that increase going to the skin (Rowell *et al.* 1969). For example, if one assumes that cardiac output increases from $\sim 5 \text{ l min}^{-1}$ to 11 l min^{-1} during a given heat stress at a blood pressure (gradient) of 85 mmHg, then total vascular conductance would be $\sim 129 \text{ ml min}^{-1} \text{ mmHg}^{-1}$. Assuming that $\sim 6.25 \text{ l min}^{-1}$ is directed to the skin in this example, equivalent carotid baroreflex-mediated changes in CVC to those observed

in the current investigation during whole-body heating (i.e. -15% to $+10\%$, or $\sim 25\%$ range), would correspond to $\sim 18 \text{ ml min}^{-1} \text{ mmHg}^{-1}$ ($\sim 14\%$ of total vascular conductance). This would represent an $\sim 11 \text{ mmHg}$ range in which arterial blood pressure may be modulated by carotid baroreflex control of the cutaneous vasculature during whole-body heating.

Both NP and NS reduced T_{neck} during normothermia and whole-body heating (see Methods). These control trials may help in discriminating between the role of true baroreflex-mediated changes in CVC and the role of potential T_{neck} changes, as well as the effect of the breath-hold itself. Although slight reductions in CVC were observed during these control trials in both thermal conditions, when the changes in CVC observed during the control trials were subtracted from the corresponding those of NP and NS trials, appropriate changes in CVC to these perturbations remained during whole-body heating. However, simple subtraction relies on the responses being additive (i.e. no interaction between baroreflex and potential breath-hold or T_{sk} responses), and therefore may not be the most appropriate analysis. Regardless, potential effects of the breath-hold and changes in T_{neck} are inadequate to describe different directional responses in CVC between NP and NS stimuli during whole-body heating (Fig. 6B).

In summary, carotid baroreceptor unloading and loading, using the neck chamber technique, caused cutaneous vasoconstriction and vasodilation, respectively, during whole-body heating. These data strongly suggest that the carotid baroreflex exhibits an efferent limb governing the cutaneous vasculature, and therefore the cutaneous vasculature appears to be involved in moment-to-moment regulation of arterial blood pressure through the carotid baroreflex.

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