Age-specific modification of local cutaneous vasodilation by capsaicin-sensitive primary afferents

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In response to increases in core body temperature, older individuals have a reduced ability to increase skin blood flow (SkBF) (15, 16). To investigate these age-related decrements in SkBF, several researchers have used local-heating methods to examine cutaneous vascular function in aged skin. Attenuated local SkBFs have consistently been observed at rest and during local heating (8, 22, 24, 28, 30, 37, 38). Structural changes associated with aging in the skin and cutaneous microvasculature have provided much of the explanation for the decrements observed in local SkBF. Furthermore, there is evidence that the reduced cutaneous vasodilation during local heating of aged skin is due to an impaired axon reflex-mediated vasodilation, as well as diminished nitric oxide (NO) production and/or responsiveness in the skin of older individuals (24).

In his classic observations, Lewis (18) described a pronounced reddening of the skin in response to localized heat that was presumably caused by vasodilation of cutaneous blood vessels. It was postulated that this vasodilation was caused initially by a relaxation of vascular tone in the heated region and indirectly by a release of vasodilator substances in the skin that occurs at higher temperatures (18). Furthermore, it was noted that this reddening occurs independent of central nervous system activation.

It is likely that the initial vasodilator effect of local heating on the skin results from a reduction of noradrenergic vasoconstriction due to a decrease in the affinity of α2-adrenoreceptors for norepinephrine (7). However, the vasodilation caused by local heating exceeds that caused by noradrenergic blockade alone, suggesting that there are other mechanisms involved in addition to a release of vasoconstrictor tone (39). Recent evidence indicates that the vasodilation that occurs during local heating of the skin is primarily mediated by NO (14). Although SkBF responses to local heating of the skin vary depending on the temperature and the length and rate at which it is applied (1, 14, 21, 27), a typical response can be described in which there is an initial rapid rise in blood flow mediated by axon reflexes at the onset of heating, a brief nadir, and then a second slower rise and sustained plateau that is primarily dependent on local NO production (23).

The initial axon reflex vasodilation that occurs during rapid local heating of the skin may be partially mediated by heat-sensitive nociceptors that are capable of releasing vasoactive neurotransmitters in response to temperatures below those that produce a conscious perception of pain (21). Although it was found that blockade of the axon reflex did not affect cutaneous vasodilation during sustained local heating at 40°C (23), other investigators suggest that heat-sensitive nociceptors are important for this response (33). There is a certain population of heat-sensitive sensory nerves in the skin that are also sensitive to...
capsaicin, the pungent ingredient in hot peppers. These capsaicin-sensitive primary afferent (CSPA) sensory nerves include many, but not all, C-fiber nociceptors, some C-fiber warmth receptors, and some Aδ-fiber polymodal nociceptors (10, 20, 29). Although acute capsaicin stimulates these nerves, chronic capsaicin administration causes a reversible desensitization that specifically blocks their function in the skin (2, 4, 6, 19, 26, 31). As such, axon reflexes normally elicited by CSPAs can be blunted or abolished with repeated applications of capsaicin (2, 4, 19, 31).

It is not known whether CSPAs are the sensory nerves that mediate the initial axon reflex vasodilation during rapid heating of the skin or exactly how these nerves contribute to the sustained increase in SkBF during prolonged heating of the skin. Furthermore, impaired sensory nerve function in aged skin may cause an attenuated neurogenic vasodilation in response to local heating. The purpose of this study was to determine the age-specific modification of cutaneous vasodilation by CSPAs during local heating of the skin. It was hypothesized that CSPA activity would contribute modestly to the increase in SkBF evoked by local heating and that this contribution would be diminished in aged skin.

METHODS

Subjects. Nine young (18–30 yr old), eight middle-aged (40–55 yr old), and eight older (65–80 yr old) healthy men participated in the experiments. Health status was determined by a physician-supervised screening at The Pennsylvania State University General Clinical Research Center. Subjects were noninstitutionalized and normally active. Exclusionary criteria included obesity (body mass index ≥ 30 kg/m²), hypertension (systolic > 140 mm Hg and/or diastolic > 90 mm Hg), underweight (body mass index < 20 kg/m²), smoking, any medication with the potential to alter cardiovascular or thermoregulatory control or response, allergies to hot peppers, and various dermatological conditions or diseases. Table 1 describes the physical characteristics of the subjects. All experimental procedures were explained to the subjects, and they were given an opportunity to ask questions. Before participation, all subjects signed an informed consent approved by the Institutional Research Board of The Pennsylvania State University.

Table 1. Subject characteristics

<table>
<thead>
<tr>
<th></th>
<th>Young* (n = 9)</th>
<th>Middle-Aged (n = 8)</th>
<th>Older (n = 8)</th>
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<tr>
<td>Age yr</td>
<td>24.3 ± 2.1</td>
<td>44.8 ± 6.0†</td>
<td>69.7 ± 4.0‡</td>
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<tr>
<td>Height, cm</td>
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<td>176.4 ± 2.9</td>
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<tr>
<td>V0₂ max, ml·kg⁻¹·min⁻¹</td>
<td>52.7 ± 11.7</td>
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<td>26.9 ± 5.6‡</td>
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<tr>
<td>Body fat, %</td>
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<td>17.7 ± 4.6</td>
<td>25.4 ± 4.2‡</td>
</tr>
<tr>
<td>Body mass index</td>
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<td>23.7 ± 2.3</td>
<td>26.3 ± 3.1‡</td>
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<td>Systolic BP, mm Hg</td>
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<td>132.0 ± 13.1</td>
<td>139.3 ± 6.1</td>
</tr>
<tr>
<td>Diastolic BP, mm Hg</td>
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<td>81.0 ± 8.9</td>
<td>84.2 ± 2.8</td>
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<td>MAP, mm Hg</td>
<td>92.8 ± 6.7</td>
<td>98.0 ± 9.4</td>
<td>102.6 ± 1.8†</td>
</tr>
</tbody>
</table>

Values are means ± SD; n, no. of subjects. V0₂ max, maximal O₂ consumption; BP, blood pressure; MAP, mean arterial pressure.

*One young subject participated only in the rapid-heating protocol.
†Significantly different (P = 0.05) from young.
‡Significantly different (P = 0.05) from middle-aged.

Screening. For each subject, an initial visit to the laboratory included a physical examination by the staff doctor and a medical history assessment. A graded exercise test was also performed to determine maximal aerobic capacity. A 0.075% capsaicin cream (Clay-Park Labs, Bronx, NY) was applied to the back of one hand for 30–40 min to identify any adverse reactions to capsaicin. In the absence of symptoms indicating hypersensitivity, including an unusually painful or hyperemic response to capsaicin, the subjects were cleared to participate in the study.

Experimental design. The study consisted of two experimental days typically separated by 1 wk of topical capsaicin pretreatment (CP). On the 1st day of experiments, rapid and slow local-heating protocols were performed at control sites on one arm. After the opposite arm was subjected to CP for 7 days (see below), the efficacy of CP was verified at the beginning of the 2nd experimental day. If the criteria for CSPA desensitization were met, the two local-heating protocols were repeated at CP sites.

Protocol. Temperature and humidity were maintained at thermoneural conditions (~23°C and ~30% relative humidity) in subject’s environment. The subject reclined comfortably on an adjustable hospital bed. During an initial rest period of 5–10 min, each arm was supinated and set on separate, movable armrests. The inflatable cuff of a Finapres blood pressure monitor (Ohmeda, Madison, WI) was placed on the middle finger of the nonexperimental arm to estimate blood pressure and heart rate. Finapres output was monitored for stability before commencement of the experiments.

Rapid heating. After an experimental arm was randomly selected, a standard type-T thermocouple was affixed to a flat area on the distal part of the ventral forearm. The thermocouple was covered at the end by a cork disk to minimize the influence of surrounding air temperature on the measurement of local skin temperature (Tloc). The tip of the thermocouple was located within a ~1-cm-diameter circle that was drawn onto the skin and served as the target for the laser-Doppler imaging (LDI) beam. The LDI was configured to take single-point measurements, an option that is fundamentally the same as that used with standard laser-Doppler flowmetry (i.e., the laser beam remains in a fixed position over a single point on the skin). Local heating was accomplished convec-
tively by directed warm air. A modified blow dryer with a double-vented nozzle (~4-cm² opening) was positioned in an adjustable stand to heat the measurement site without obstructing the LDI beam. A Powerstat voltage controller regulated the output of the blow dryer. Using the visual display of Tloc measured by the thermocouple, the investigator could maintain precise manual control over the amount of heating provided by the blower. A fenestrated drape was placed over the forearm, exposing only a small area (4 cm²) to the directed warm air.

The heated site (4 cm²/0.62 in.²) was kept at 33°C for 10 min during an initial baseline period. At the 10th min, the blower was turned up to a setting on the Powerstat that caused Tloc to increase from 33 to 40°C within ~2 min. The same Powerstat setting was used for every trial. Tloc was maintained at 40°C for 50 min, during which LDI was measured continuously. After 50 min of heating to a Tloc of 40°C, the blower was turned off and 5 min of LDI measurements were obtained during a recovery phase.

Slow heating. The slow-heating protocol was performed at the proximal end of the ventral forearm. A fenestrated drape with a larger opening (16 cm²) was used to cover the forearm at this site. The insulated thermocouple was relocated and used to measure Tloc. A spot ~2 cm away from the thermocouple placement was marked with a soft pen to identify the
center of the measurement site. For the slow-heating period, LDI scans were employed. The LDI system configurations were as follows: scan size = 1.6 × 1.6 cm, scan time = 11 s, scan speed = 4 ms/pixel.

First, an initial LDI scan of the site was taken to serve as a baseline. The blower was then positioned directly above the measurement site so that the stream of air was perpendicular to the surface of the forearm. Tloc was raised in a stepwise manner at a rate of 1°C/min from 35 to 42°C and then maintained at 42°C for an additional 30 min. Every 5 min (at the end of each increment), an LDI scan was taken. During each scan, the blower was removed from its cradle and held by a researcher from the side to allow an unobstructed measurement. After completion of the final scan (15th in the series), all instrumentation and heating equipment were removed, ending the slow-heating period.

CP. After the initial experimental session, instructions for CP were given. Three application sites (25 cm²) were marked with a soft pen on the ventral aspect of the nonexperimental forearm. Subjects were provided with a supply of 0.075% capsaicin cream (Clay-Park Labs), applicators, and bandages. They were given a demonstration of how to apply the cream that included precautions associated with its pungency. The application sites were thinly coated with the capsaicin cream and covered with a bandage twice daily for 7 days. The cream was left on the sites continuously, except when the subjects were bathing/showing, swimming, or exercising.

Verification of CP efficacy. The efficacy of CP was verified after 7 days of application. A baseline scan of a CP site was taken, and then patches containing 300 μl of a vehicle solution (95% ethanol) and 300 μl of a 1.0% capsaicin (Sigma Chemical, St. Louis, MO) solution were applied to separate CP sites. After 32 min, the patches were removed and LDI scans were immediately taken. Mean LDI flux values for all three scans (baseline, vehicle, and capsaicin) were analyzed and compared. The criterion for desensitization was that the mean LDI flux value at the capsaicin site had to be <150% of that at the vehicle site. If this standard was met, the two local-heating protocols were performed at CP sites. For further verification, responses to 1.0% capsaicin at CP sites were also compared with those at non-CP sites (control). The CP site where 1% capsaicin was applied was used exclusively to verify CP efficacy. The CP site where vehicle was applied was later used for the slow-heating protocol. At least 75 min elapsed from the end of the vehicle application to the commencement of slow heating at that site. Baseline values before heating were not different from those at control sites, indicating that the local erythema had subsided.

LDI data analysis. The LDI system (Moor Instruments) used a visible red helium-neon laser operating at a wavelength of 632.8 nm. This low-power laser beam scanned the surface of the skin in a raster pattern and was processed to create a color-coded image of skin perfusion. Each pixel in the 1,600-pixel (40 × 40 pixel) scan had a unique flux value, proportional to SkBF. The principal measurement of interest was the mean flux value, found by averaging the mean flux values of five 900-pixel (30 × 30 pixel) regions of interest (ROIs) uniformly placed at each corner and in the center of each scan. The ROIs overlapped such that the center of each scan was oversampled. This method allowed all 1,600 pixels to be used without giving a disproportionately high weighting to pixels on the periphery that sometimes had a greater variability. A mean flux value for each ROI was calculated by LDI software.

All LDI measurements were indexed to percentage of maximal cutaneous vascular conductance (%CVCmax) by dividing mean cutaneous vascular conductance (CVC) by CVCmax. Mean CVC was determined as the ratio of LDI flux to mean arterial pressure (MAP) at the time of the scan (CVC = LDI flux/MAP). Local heating of the skin to 42°C for 30–60 min has been shown to evoke maximal cutaneous vasodilation (13, 34). Accordingly, CVCmax for each subject was found by averaging the mean CVC values of five 25-pixel ROIs within the LDI scan taken during the slow-heating protocol that had the highest mean CVC while Tloc was at 42°C. In addition to being used for the slow-heating protocol, this maximal CVC value was also applied to the normalization of data obtained during the rapid-heating protocol and the CP efficacy protocol.

Because pixels could not have been matched exactly from scan to scan (so that each pixel could be indexed to its own unique value for CVCmax), it was necessary to use a mean CVCmax determination. In pilot work, LDI scans were analyzed such that various pixel samples (1, 4, 9, 25, and 1,600 pixels) that had the highest CVC values in a particular scan were identified. After the CVC values of these different pixel samples were compared, it was discovered that they were not significant from each other in such that meaningful CVC determinations at the highest pixel value was proportional to the CVC of the entire 1,600-pixel area. The CVCmax determination we used (averaging five 25-pixel ROIs) produced a value that allowed the rapid-heating (single-site CVC) and slow-heating (40 × 40-pixel-area CVC) data to be indexed in a reasonable way that was consistent with previous values in the literature. This novel method of normalization assumes that the calculated CVCmax from an LDI scan of a relatively large area provides an index of CVCmax that is not site specific but, rather, characteristic of the functional capacity of an individual’s skin.

The percentage of the area that was vasodilated (%AVD) was also determined for the slow-heating scans taken when Tloc = 36, 38, and 40°C and for three of the scans taken when Tloc = 42°C. A threshold value that represented vasodilation (defined in this study as CVC > 25% CVCmax) was subtracted from each scan, and the LDI software calculated the percentage of valid pixels (those still remaining) to yield %AVD. Then the %CVCmax of only those areas that were identified as being vasodilated was determined. Finally, dose-response curves were fitted to the overall %CVCmax data from the slow-heating protocol using a four-parameter logistic function curve model of the following form: \( y = y_0 + \frac{(y_m - y_0)}{[1 + \left(\frac{x}{x_0}\right)^{b}]}} \) (SigmaPlot).

One-minute flux averages were calculated for the single-site tracings of the rapid-heating protocol. All flux measurements were converted to CVC and expressed as %CVCmax as described previously. Baseline was the average CVC during the first 10 min of the period. The initial peak value was calculated as the 1-min mean CVC around the highest flux value immediately following rapid heating. The nadir value was considered to be the 1-min mean CVC around the single lowest flux value during the reduction in SkBF after the initial peak. The value for the plateau phase was the highest 10-min average following the nadir. Finally, the recovery value was the calculated 3-min mean CVC following termination of the heating.

Statistical analysis. Physical characteristics among the three age groups were compared using a one-way ANOVA (Excel). When significant main effects of age were found, t-tests were performed using Bonferroni’s corrections to determine between which groups the differences existed. A three-way ANOVA with repeated measures was initially conducted for the slow- and rapid-heating protocols. A priori simple interactions using a two-way ANOVA with repeated
measures were also performed to determine treatment effects at each level of age. When significant differences were found, appropriate follow-up post hoc analyses were completed. All ANOVAs were computed with SAS software (SAS Institute, Cary, NC) and utilized the Proc Mixed procedure for mixed models. The significance level for all statistical tests was set at $\alpha = 0.05$. Values are means $\pm$ SE, unless otherwise noted.

RESULTS

Verification of CP efficacy. For each age group, there was no difference ($P > 0.05$) in %VCVmax at CP sites where 1.0% capsaicin and vehicle solutions were applied (Fig. 1). This was interpreted as an indication of capsaicin-specific desensitization. Furthermore, in the young and middle-aged groups, the SkBF response to 1.0% capsaicin was significantly reduced at CP sites compared with non-CP sites (24.6 vs. 61.0% CVCmax and 14.2 vs. 51.0% CVCmax, respectively, $P < 0.05$). In the older group, a less apparent decrement was observed, but the relatively small initial response to 1.0% capsaicin at control sites likely prevented detection of a significant difference ($P = 0.051$).

Rapid heating. Rapid heating resulted in the characteristic biphasic SkBF response previously described (23), i.e., an initial increase and peak at the onset of heating, a brief nadir, and then a slower rise and plateau (Fig. 2). This pattern was seen for all three age groups before and after CP. The initial peak response was significantly decreased (53.9 vs. 74.4% CVCmax, $P < 0.05$) at CP sites in young individuals (Fig. 3A). However, in no instance was the sensory nerve-mediated initial peak response completely eliminated after CP. Smaller reductions occurred in the nadir and plateau phases of the young group after CP, but they did not reach the level of statistical significance. The effect of CP on the rapid-heating SkBF responses in the middle-aged and older groups did not reach statistical significance (Fig. 3). When the phase responses of the different age groups were compared, there were no significant differences before or after CP.

Slow heating. CP significantly attenuated the SkBF responses to slow heating in the middle-aged and older groups ($P < 0.05$; Fig. 4). Reductions also occurred in the young group, but they did not reach statistical significance ($P = 0.056$). For the older group, diminished areas of vasodilation and decreased magnitudes of vasodilation within those areas accounted for the smaller %VCVmax at CP sites (Figs. 5 and 6). In the middle-aged group, only %AVD was significantly altered ($P < 0.05$) by CP and was entirely responsible for the reduced %VCVmax.

There were no age differences in the response to slow local heating before or after CP as indicated by ANOVA and dose-response curves. Nevertheless, the significant reductions with CP in the middle-aged and older groups suggest age-specific effects of CP on SkBF responses to slow local heating. It is worth noting that there was a tendency for %VCVmax to rise at control sites after $T_{loc}$ reached 42°C, a temperature near the in vivo thermal activation threshold for the capsaicin [vanilloid receptor subtype 1 (VR1)] receptor (5). This persistent increase after $T_{loc}$ reached 42°C was eliminated at the CP sites.

DISCUSSION

This study provides evidence that CSPAs make only a minor contribution to the cutaneous vasodilation elicited by local heating of the skin below temperatures that initiate pain and are not primarily responsible for the initial SkBF caused by rapid local heating of the skin to 40°C. Furthermore, this study demonstrated...
that chronic CP 1) attenuated cutaneous vasodilation during progressive, local heating of the skin in middle-aged and older individuals and 2) reduced the initial SkBF peak during rapid local heating of the skin in young individuals. These results reflect age-specific alterations in CSPA-mediated local vasodilation that may be associated with attenuated SkBF during whole body heating.

The rapid local-heating experiment was performed primarily to test whether CSPAs are involved in the initial axon reflex-mediated peak in SkBF that occurs immediately at the onset of heating (23). CSPAs were hypothesized to be involved in the response on the basis of 1) their expression of the heat-gated VR1

Fig. 3. SkBF response to rapid local heating by age. Effect of 7 days of CP on %CVC\textsubscript{max} during rapid local heating of young (A), middle-aged (B), and older (C) skin is illustrated. Overall %CVC\textsubscript{max} was significantly reduced in the young group after CP, as was the sensory nerve-mediated initial peak in SkBF after the start of heating (A). *Significantly different from control ($P < 0.05$).

Fig. 4. SkBF response to slow local heating by age. %CVC\textsubscript{max} to slow local heating before and after CP in young (A), middle-aged (B), and older (C) individuals is shown. After a baseline measurement, local skin temperature was increased to 35°C and subsequently raised in a stepwise manner at a rate of 1°C/5 min. An LDI scan was taken at the end of every 5-min heating increment. After measurement at minute 35, local skin temperature was elevated to 42°C and clamped there for the remainder of the protocol. CP resulted in significantly different responses for middle-aged and older groups. *Significantly different from CP site ($P < 0.05$).
receptor, 2) their ability to cause neurogenic vasodilation, and 3) previous indications that heat-sensitive nociceptors were important for local heating-induced vasodilation (5, 10, 11, 33). Although the activation threshold for VR1 is thought to be higher than the 40°C \( T_{\text{loc}} \) used previously by Minson et al. (23), activation characteristics of sensory neurons are dependent on the rate at which temperature changes as well as the magnitude of the temperature itself (3, 5, 21, 32, 35).

Fig. 5. Vasodilated area during slow local heating. Areas of vasodilation in response to slow local heating in young (A), middle-aged (B), and older (C) skin before and after CP are illustrated. CP resulted in significantly reduced areas of vasodilation during slow local heating in middle-aged and older groups. *Significantly different from CP site \((P < 0.05)\). (Note fewer x-axis values than in Fig. 4, inasmuch as this analysis was not performed at every temperature of the protocol.)

Fig. 6. Magnitude of SkBF in vasodilated area during slow local heating. Magnitude of SkBF in vasodilated areas (Fig. 5) is shown for young (A), middle-aged (B), and older (C) individuals. There was a significant reduction in \( %\text{CVC}_{\text{max}} \) after CP in the older group (C). *Significantly different from CP site \((P < 0.05)\). (Note fewer x-axis values than in Fig. 4, inasmuch as this analysis was not performed at every temperature of the protocol.)
With faster rates of change, activation thresholds for thermal-sensitive neurons are reduced (21). Therefore, it is possible that heat-sensitive CSPAs can respond to rapid increases in $T_{loc}$ by mediating a protective neurogenic vasodilation.

The initial peak response to rapid local heating was significantly reduced after CP only in the young group. Because the initial peak response is known to be sensory nerve mediated (23), this result is consistent with findings from our laboratory and elsewhere of impaired neurogenic vasodilation in aged skin (9, 17). Another notable finding was that although the peak response was reduced in younger skin after CP, the response was only modestly reduced and never completely abolished. This observation suggests that cutaneous sensory nerves that are not capsaicin sensitive participate in this response to a greater extent. Recently, a newly identified vanilloid receptor-like subtype 3 (VRL-3) was discovered that is heat sensitive but capsaicin insensitive (32, 40). In vitro, it has a lower thermal activation threshold than the VR1 receptor found on CSPAs (~39°C vs. ~43°C) (5, 32, 40), putting it directly in range of the 40°C $T_{loc}$ used in the present study. Because CP would have only desensitized CSPAs, sensory nerves that were not capsaicin sensitive would have remained active. Thus it is likely that neurons expressing the VRL-3 receptor are at least partially responsible for mediating the initial peak response.

During prolonged local heating of the skin at a $T_{loc}$ that does not elicit pain (37–42.5°C), there is a marked increase in blood flow that occurs through the action of local mechanisms, primarily mediated by NO, and this increase in blood flow is confined to the skin (1, 12, 14). The relatively modest effect of CP on the overall cutaneous vasodilation during prolonged heating of the skin reflects the predominant role of NO-mediated mechanisms in this response. Consequently, the neurogenic vasodilation resulting from CSPA activation is additive to the overall SkBF response, such that a widespread vasodilation and vascular recruitment exists independent of CSPA activity. Therefore, increases in %AVD via CSPA activity were probably not achieved by further vascular recruitment but, rather, were due to additional vasodilation of vessels stimulated by CSPA neuropeptides that elevated their magnitude of flow above the threshold for vasodilation used in this analysis.

In a recent study investigating age-related cutaneous vasodilation during local heating, it was concluded that older individuals have a diminished capacity to produce and/or respond to NO (24). Impaired NO production or responsiveness in aged skin could mean that CSPA activation contributes more toward the maximal SkBF response to local heating in these individuals. The fact that SkBF was significantly reduced by CP in the middle-aged and older groups but not in the young group during slow local heating supports this assertion. An alternative explanation may be that after CSPA function was blocked by CP, a more robust NO production and/or responsiveness in younger skin was engaged and restored SkBF to a greater extent than in the other two groups. In other words, a greater NO reserve activity may have masked the deleterious effects of CP to a larger extent, even if CSPA-mediated vasodilation was initially more extensive in younger skin. Nevertheless, cutaneous vasodilation in the skin during prolonged local heating is only modestly dependent on CSPA activity in all age groups, as NO-mediated mechanisms prevail.

There were significant reductions in SkBF at CP sites during slow heating, but not during the later phases of rapid heating, which was a similar period of prolonged heating. A potential explanation for this discrepancy may be that, during the slow-heating protocol, the final 30 min were clamped at a $T_{loc}$ of 42°C, whereas in the rapid-heating protocol, the prolonged heating was clamped at a $T_{loc}$ of 40°C. This relatively small difference is important if the proximity to the VR1 thermal threshold (~43°C) is considered (5). At 42°C, subthreshold firings of CSPAs are likely to occur with more rapidity than at 40°C. Thus, at temperatures a few degrees below the activation threshold of these neurons, the difference in CSPA activity is presumably great. Alternatively, because there were significant differences in %CVC$_{max}$ for the older subjects at 40°C in the slow- but not the rapid-heating protocol, variations in the temporal pattern of heating may have contributed to this finding.

Previous studies have consistently shown attenuations in absolute SkBF associated with aging during local heating of the skin, including reduced maximal responses (22, 30). Therefore, although subjects from all age groups reached a similar %CVC$_{max}$ during both protocols of this study, presumably lower CVC$_{max}$ values in aged skin would translate to a lower absolute SkBF for a given %CVC$_{max}$. This finding of older and younger individuals reaching a similar %CVC$_{max}$ during local heating is consistent with previous observations during whole body heating (16). SkBF was indexed to %CVC$_{max}$ in an attempt to minimize the influence of structural changes in the skin, because it has been suggested that maximal SkBF “reflects a fundamental property of the vasculature of an individual’s skin” and that this property is “the appropriate basis for scaling the range of responses in an individual and for comparison with other individuals” (30). However, in addition to structural properties, functional components may limit CVC$_{max}$ elicited by local heating in aged skin (24). As a result, indexing SkBF to %CVC$_{max}$ in this study may not have completely normalized the influence of structural changes in the skin. Consequently, structural limitations, including decreased functional capillary plexus units, damaged or obliterated vessels, and/or attenuated precapillary recruitment, may also have contributed to the age-specific SkBF responses that were observed (8, 22, 25, 28, 38).

Interpreting the results of these experiments is dependent on accepting the assumptions of the CVC$_{max}$ normalization that was used and the effectiveness of CP in blocking CSPA activity. Although all subjects
met the criteria for desensitization, it is possible that desensitization was incomplete or different among individuals. The nearly complete absence of any vasodilatory response to acute capsaicin in middle-aged and older skin after CP suggests an effective desensitization in these groups. In fact, the significantly reduced SkBF response to acute capsaicin at control sites in aged skin compared with younger skin indicates that desensitization existed before CP in the older group. Because the goal of this study was to determine the age-specific contribution of CSPAs to SkBF responses during local heating of the skin, this finding was not limiting. Intuitively, an age-related desensitization would lead one to conclude that the contribution of CSPAs is reduced or absent in aged skin; however, because the SkBF response to slow heating was attenuated by CP in the older group, CSPAs apparently retain their ability to respond to cutaneous vasodilatory stimuli of sufficient intensity.

Although a slight vasodilation in response to 1% capsaicin persisted in younger skin after CP, the absolute reduction in SkBF at CP sites compared with control was substantial and at least as much as that achieved in the other two groups. Additionally, vasodilation resulting from nonspecific effects of ethanol in younger skin may have been responsible for the persistent SkBF responses seen at vehicle and 1% capsaicin sites (36). Furthermore, previous studies employing similar CP methods to younger skin have demonstrated desensitization, defunctionalization, and reversible neuronal damage within the length of pretreatment used in the present study (6, 26, 31).

In conclusion, CSPAs contribute only modestly to the cutaneous vasodilation that occurs in response to local heating of the skin at temperatures below those that elicit pain. SkBF was preferentially attenuated by CP I) in middle-aged and older skin during progressive local heating and 2) in younger skin during rapid local heating. These results imply that, with advanced age, CSPA activity is more important to the maximal SkBF response during prolonged local heating, whereas it has a reduced role in the initial SkBF peak elicited by rapid local heating. The interaction of reflex mechanisms with these modifications of local SkBF may contribute to the age-related attenuation in cutaneous vasodilation during whole body heat stress.

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DISCLOSURES

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REFERENCES


