Peripheral Mechanism Involved in Response of Dogs' Cutaneous Veins to Local Temperature Change

By Michael M. Webb-Peploe, M.B., and John T. Shepherd, M.D., D.Sc.

ABSTRACT

The cutaneous veins of the dog constrict and dilate with local cooling and warming, respectively. This response, which is dependent on the integrity of the adrenergic nerves, might be explained by an altered response of the smooth muscle to a given frequency of sympathetic nerve impulses or by local temperature receptors that initiate alterations in sympathetic nerve activity.

In the lateral saphenous vein perfused at 42°, 27°, and 17°C, the constriction caused by stimulation of lumbar sympathetic nerves was 40%, 160%, and 200%, respectively, of that at 37°C; the constriction caused by norepinephrine at the same temperatures was 70%, 187%, and 318% of that at 37°C. Division of the dorsal spinal roots from L-1 to S-1 made no difference to the veno-constrictor response to local cooling. It is concluded that an alteration in the sensitivity of the smooth muscle to arriving sympathetic nerve impulses accounts for the venous responses to local temperature changes seen in the intact dog.

ADDITIONAL KEY WORDS
norepinephrine and temperature
veins and temperature
local temperature and veins

temperature and sympathetic impulses
veins and norepinephrine
posterior nerve roots and veins
venomotor reflexes

Previous studies (1, 2) have shown that the superficial limb veins of the dog constrict in response to either a decrease in central temperature or to a cooling of the blood perfusing the vein. Whereas central cooling causes constriction of the cutaneous veins in all the limbs, the constriction with local cooling is confined to the cooled limb. The magnitude of the response to a fixed local cold stimulus is governed by the central temperature; with a decrease in central temperature of a few degrees the response is much greater, whereas after central warming it is much less. The response to changes in both local and central temperature depends on the integrity of the sympathetic nervous system.

These studies did not indicate the nature of the local thermoregulatory mechanism, nor did they shed any light on the site at which integration of local and central thermal drives takes place. Two hypotheses appear to fit the facts (1). Local cooling enhances the response of the smooth muscle of the vein to a given frequency of sympathetic impulses, but does not change the frequency. Central cooling increases the frequency. Integration of local and central thermal drives occurs in the vein wall (2). Local cooling stimulates temperature receptors in the limb, causing an increase in frequency of the sympathetic impulses to the veins of that limb. Integration of local and central thermal drives occurs in the spinal cord or higher centers.

The experiments in series 1 were designed to test the first hypothesis by studying the effect of changes in local temperature on the venous response to electric stimulation of the
sympathetic nerves and to norepinephrine infusion.

In series 2 experiments, we tested the second hypothesis by attempting to find an afferent limb of the reflex arc.

Methods

SERIES 1

In dogs anesthetized with thiopental, 15 mg/kg, and chloralose, 80 mg/kg iv, and artificially ventilated with oxygen, the lateral saphenous vein was cannulated at the ankle and perfused at constant flow by a roller pump with blood taken from the median sacral artery. A heat exchanger was placed in the circuit, and perfusion and femoral vein pressures were measured. During experiments, the common iliac artery was occluded. With this method (3) any alteration in driving pressure, that is, the difference between perfusion (inflow) pressure and femoral vein (outflow) pressure, is due to a change in venomotor activity, because blood flow through the vein is constant.

Altering the temperature of the water flowing through the heat exchanger made it possible to obtain rapid and reproducible changes in temperature in the vein (1). Blood temperature was measured just upstream to the cannula in the lateral saphenous vein.

Sympathetic Stimulation.—The lumbar sympathetic chain was divided at the level of the second or third vertebral body and dissected free down to the level of the sixth vertebral body. The chain was stimulated with an electric stimulator (Grass Instruments Co., model S4) via a platinum bipolar electrode, using a stimulus of 8 v applied for 1 msec with no delay. Previous experiments (3) had shown that this voltage provides a stimulus of supramaximal intensity. Stimulation frequencies of 2, 6, and 10 cps were used.

Norepinephrine Infusion.—Norepinephrine (Levophed, Winthrop Laboratories, New York) was infused upstream from the roller pump at rates of 2.5, 5, 10, and 20 μg/min, using a Harvard infusion pump.

SERIES 2

Three sets of experiments were conducted. In three dogs, the lateral saphenous vein was perfused at constant flow and the effect of local changes in temperature was assessed before and after the vein was dissected free of skin and surrounding connective tissue.

In another three dogs, the lateral saphenous vein was severed, and the two segments were perfused separately at constant flow; two roller pumps were used so that temperature conditions could be varied independently in the adjacent segments.
parts of the vein. One inflow cannula was placed at the ankle, and the vein was double cannulated about 4 to 6 cm proximally. The more distal of these two cannulas carried blood out of the distal segment of vein and via a short length of Silastic tubing into the femoral vein of the opposite hindleg. The more proximal of the two cannulas served as the inflow cannula for the proximal segment of vein.

Finally, in six dogs the effect of section of the lumbar dorsal spinal roots on the venous temperature responses was assessed. The dogs were placed prone in a cast, and both lateral saphenous veins were perfused at constant flow by two roller pumps. Perfusion and femoral vein pressures were measured in each leg. Bilateral lumbar laminectomy exposed the spinal cord. In three dogs, the posterior roots L-4 through L-7 and S-1 on one side, together with the cauda equina, were cut; in the other three dogs, only the posterior roots L-1 through L-4 were cut on one side. The responses to local changes in temperature were compared in the two legs before and after unilateral division of the posterior roots.

Results

**SERIES 1**

After acute lumbar sympathectomy, changes in temperature of the perfusate had little effect on saphenous venomotor tone. In the 10 dogs studied (6 dogs, Table 1; 4 dogs, Figs. 3 and 5), the driving pressure increased by an average of only 6 mm Hg with a decrease in perfusate temperature from 42° to 27°C.

**Sympathetic Stimulation**—The maximal increment in driving pressure during stimulation of the lumbar sympathetic chain at 2 and 6 cps was measured at perfusion temperatures of 37°, 42°, and 27°C in five dogs and 37°, 47°, and 57°C in one dog. The responses to stimulation of 10 cps also were obtained at these temperatures in four of the six dogs (Table 1). Between each series of stimulations at a temperature other than 37°C, the response to stimulation of 10 cps and 37°C was used as a control. Thus it was possible to be certain that any change in response at a temperature other than 37°C was due to the temperature change and was not the result of changes in electrode position or of fatigue. Changes in temperature had a similar effect on the responses to all three stimulation frequencies, although the results at 2 cps were more variable than at the higher frequencies. A rise in temperature from 37° to 42°C reduced the responses by an average of 59%, and a decrease in temperature from 37° to 27°C increased the responses by an average of 45%. In the one dog in which a temperature of 47°C was used, there was no response to stimulation.

Changes in temperature not only affected the magnitude of the response to stimulation but also influenced the latent period between

![Graph](image-url)
Venomotor responses to electric stimulation of lumbar sympathetic chain at 10 cps during perfusion of lateral saphenous vein with blood at temperatures of 17°, 27°, 37°, and 42°C.

The two signals on base line of each record indicate onset and termination of stimulation (dog 2 in Table 1). Ordinate scale (cm Hg) refers to saphenous vein perfusion and femoral vein pressure.

Figure 2

The onset of stimulation and the start of vasoconstriction. This lag could be measured accurately at all four temperatures (17°, 27°, 37°, and 42°C) only when stimulation of 10 cps was used, and averaged 29 seconds at 17°C, 10.5 seconds at 27°C, 6 seconds at 37°C, and 2.5 seconds at 42°C.

Temperature also had an effect on the rate at which the vasoconstriction induced by sympathetic stimulation passed off once the stimulus was stopped (Fig. 1, left). The higher the temperature, the faster the relaxation. Similar results were obtained in all animals.

The responses to stimulation of 10 cps at 17°, 27°, 37°, and 42°C in one animal are shown in Figure 2, and the records illustrate the effect of temperature changes on the magnitude, latent period, and rate of relaxation of the response.

At low temperatures, the long latent period and slow rate of tension development necessitated prolonged periods of stimulation before the response reached its peak, and it was easy for the magnitude of the response to be underestimated. To avoid this difficulty, the effect of changes in temperature on the response to continuous sympathetic stimulation was assessed in four more dogs (Fig. 3). With this method, a rise in temperature from 37° to 42°C reduced the responses by an average of 57%; at 37°C, the responses were increased by an average of 64%; and at 17°C, they were still further increased by an average of 97%, as compared to the responses obtained at 37°C.

Figure 4 shows the records from one of these experiments in which we assessed the effect of temperature variations on the venous responses to continuous sympathetic stimulation at frequencies of 0, 0.5, 1, 2, and 4 cps. The results indicate that minor changes in the frequency of sympathetic nerve stimulation caused significant alterations in the response of the veins to changes in local temperature—the higher the frequency, the greater the response to changes in temperature.

Norepinephrine Infusion.—The effect of temperature variations on the venous responses to continuous infusion of norepinephrine was assessed in four dogs (Fig. 5). As with sympathetic nerve stimulation, an increase in temperature from 37° to 42°C reduced the responses by an average of 29%, whereas reducing the temperature from 37°

Circulation Research, Volume XXIII, December 1968
to 27°C increased the response by an overall 187%, and at 17°C the response was increased by 318% compared to that at 37°C. Increasing the rate of infusion in any animal enhanced the effect of local changes in temperature. Variations in local temperature not only modified the magnitude of the response to norepinephrine but also affected the rate of relaxation after cessation of the infusion. Figure 1 (right) shows the effect of a difference of 10°C in temperature on the rate of relaxation. At 27°C, it was much slower than at 37°C.

SERIES 2

Removal of the skin from over the vein, and even dissection of the vein free of surrounding connective tissue, made no difference to the venous responses to changes in the temperature of the blood flowing through it. This indicated that if temperature receptors were involved in this response, they had to be situated in the vein wall.

In the experiments in which adjacent segments of the same vein were separately perfused and one segment was cooled and the other was maintained at 37°C, venoconstriction was confined to the cooled segment and never involved the one maintained at 37°C.

Finally, division of the dorsal spinal roots
Records from experiment investigating effect of temperature variations in the order 37°, 42°, 27°, 17°, and 37°C on responses to sympathetic stimulation at frequencies of 0.5, 1, 2, and 4 cps. (Onset and termination of stimulation are indicated by the short black bars at bottom of each tracing.) Also shown is effect of variations in temperature in absence of sympathetic stimulation. Upper tracing records temperature of blood perfusing lateral saphenous vein (scale marked °C). Scale marked mm Hg refers to saphenous perfusion and femoral-vein pressures. Changes in perfusion pressure reflect changes in vasoconstrictor activity since outflow (femoral-vein) pressure and flow through vein are constant. Note the increasing effect of changes in temperature with increasing frequency of stimulation.

Discussion

After sympathectomy, changes in temperature of the blood perfusing the lateral saphenous vein had little effect on its smooth muscle. Hence the active changes in venous wall tension with local temperature changes were dependent on the sympathetic nerves, and were not caused by a direct temperature effect on the venous smooth muscle, nor were they the result of alterations in the properties of the perfusate. Attempts to demonstrate the afferent limb of a possible reflex are from temperature receptors in the limb to the spinal cord and back to the vein via the sympathetic nerves led to the following conclusions (1). If temperature receptors in the limbs were involved, they had to be situated in the vein wall and not in the skin (2). The afferent fibers did not run with other sensory fibers, because cutting the appropriate dorsal spinal roots did not abolish the temperature response. Cooper and Kerslake (4, 5) have suggested that afferent fibers from temperature receptors run in the lumbar sympathetic nerve chains in man. However, in the present experiments in the dog, section of the upper lumbar-dorsal roots in the region of the sympathetic outflow to the hindlegs failed to abolish the response (3). If a reflex existed, then a complex system composed
Effect of temperature on saphenous venous motor response to continuous infusion of norepinephrine. The four graphs represent data from four animals. Each line on any graph represents the rate of infusion of norepinephrine given on right of each line. Note the increasing sensitivity to changes in temperature with increasing rate of infusion.

Further, the alteration in sensitivity of venous smooth muscle to both nerve stimulation and norepinephrine infusion (Table 1, Figs. 3 and 5) caused by changes in temperature was sufficient to account for the responses to local cooling and warming seen in the intact animal without the need to postulate a local temperature reflex. Of particular interest was the finding that minor changes in frequency of sympathetic nerve stimulation profoundly altered the venous response to changes in local temperature. In one animal, for example, the increase in driving pressure resulting from a decrease of 20°C (from 37° to 17°C) in perfusate temperature was 5 mm Hg in the absence of sympathetic stimulation, 15 mm Hg at a stimulation frequency of 0.5 cps, 35 mm Hg at 1 cps, 55 mm Hg at 2 cps, and 86 mm Hg at 4 cps. In the present experiments, an increase in rate of sympathetic stimulation from 0.5 to 4 cps produced an increase of response to a reduction of 20°C in local temperature which was similar to that induced by a reduction of central temperature of 4°C.

The increase in the sensitivity of venous smooth muscle to sympathetic nerve impulses...
WEBB-PEPLOE, SHEPHERD

and infused norepinephrine produced by cooling has not been described before, to our knowledge. There is valid evidence that uptake and binding of "free" norepinephrine within the sympathetic nerve ending constitutes a major means by which it is removed and inactivated (for review see Wurtman [6]). With heart slices (7), and isolated perfused rat hearts (8), this uptake occurs against a concentration gradient and involves both diffusion and an active transport mechanism. Norepinephrine is also eliminated from the neighborhood of the receptor sites by O-methylation to normetanephrine, a reaction catalyzed by the enzyme catechol-O-methyl transferase (9), and by simple diffusion, finding its way unchanged into the circulation. These processes are all likely to be temperature sensitive, and similar reactions in synapse and striated neuromuscular junctions have indeed been shown to possess a high temperature coefficient (10). A possible explanation for the increased responses to both sympathetic stimulation and norepinephrine infusion with cooling is that the rate of removal of transmitter substance from the receptor site is slowed more than its rate of arrival. The slow rate of relaxation at low temperatures after nerve stimulation or norepinephrine infusion also might be due to a decreased rate of removal of neurotransmitter from the receptor site. The observation that cooling enhances the norepinephrine response more than it does the response to nerve stimulation indicates that cooling may affect the processes involved in the liberation of neurotransmitter by an arriving impulse in addition to slowing its rate of removal from the receptor site.

In summary, the data suggest that changes in central temperature result in appropriate alterations in sympathetic nerve traffic to the cutaneous veins and that changes in local vein temperature profoundly affect the responsiveness of the venous smooth muscle to arriving nerve impulses (an increase of 5°C in perifusate temperature reducing the response by about 60%, and a decrease of 10°C increasing the response by about the same amount). The effect of changes in environmental temperature, and thus of cutaneous thermoreceptor activity, on the venomotor reactions was not examined.

Acknowledgments

Robert R. Lorenz and Roger L. Ready rendered valuable technical assistance.

References

Peripheral Mechanism Involved in Response of Dogs' Cutaneous Veins to Local Temperature Change
MICHAEL M. WEBB-PEPLOE and JOHN T. SHEPHERD

Circ Res. 1968;23:701-708
doi: 10.1161/01.RES.23.6.701
Circulation Research is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 1968 American Heart Association, Inc. All rights reserved.
Print ISSN: 0009-7330. Online ISSN: 1524-4571

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