Local response to cold in rat tail after spinal cord transection

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Kalincik T, Jozefcikova K, Waite PM, Carrive P. Local response to cold in rat tail after spinal cord transection. J Appl Physiol 106: 1976–1985, 2009. First published April 2, 2009; doi:10.1152/japplphysiol.00095.2009.—Subjects with severe chronic spinal cord injury (SCI) are prone to hypothermia when they are exposed to relatively low environmental temperatures that are normally well tolerated by healthy individuals. This impaired thermoregulation is presumably due to disconnection of territories below the SCI from supraspinal thermoregulatory centers. However, it is not known how these territories respond to low temperatures. Using a complete transection at T11 in rats, we examined the responses of the tail to cold (6–9°C) by measuring changes in tail blood flow and skin temperature weekly for 8 wk after SCI. Despite no significant change in baseline mean flow or temperature in the tail, the transection effectively removed the sympathetically mediated supraspinal control of the tail vasculature, since the amplitude of the pulse flow was markedly increased and the natural variations of the mean flow were almost abolished. As expected, the cold challenge before SCI caused a marked drop in mean flow, pulse amplitude, and temperature of the tail. Surprisingly, the drops in mean blood flow and temperature were observed after SCI, although the decrease in flow was slower and the pulse amplitude was not reduced. The results suggest that the cutaneous vasculature of the tail is sensitive to cold and will constrict, despite disconnection from supraspinal centers. This local effect is slow but may be sufficient to maintain some level of thermoregulation to cold. Without this vascular reaction, the effects of SCI on temperature regulation to cold would probably be much worse.

sympathetic nervous system; thermoregulation; skin blood flow; infrared thermography; Doppler ultrasonography; Raynaud’s phenomenon

IN HOMEOTHERMIC ORGANISMS, core body temperature is closely controlled and maintained within a relatively narrow range. Spinal cord injury (SCI) may lead to dysfunction of temperature homeostasis, which, in extreme cases, can render the organism poikilothermic (34, 35, 40). Consequently, tetraplegic individuals are prone to hypothermia or hyperthermia during exposure to temperature changes that are well tolerated in healthy individuals (14, 24, 27).

Despite the scientific attention that thermoregulation has received, whether the dermatomes innervated from the isolated spinal cord are capable of local thermoregulatory responses to environmental temperature changes has not been conclusively determined. Some studies in patients with a clinically complete SCI reported an absence of thermoregulatory responses (i.e., sweating, shivering, cutaneous vasodilatation, or vasoconstriction) below the neurological level (13, 14, 30, 36). On the other hand, a number of studies reported signs of thermoregulatory responses below the SCI when patients were exposed to high or low environmental temperatures (10, 37, 39, 43). This obvious lack of consensus could stem from the fact that, in the aforementioned studies, the completeness of the SCI was not structurally confirmed and specific autonomic functions were not assessed. Instead, the completeness of SCI was inferred from clinical examinations of sensory and motor systems [with the exception of the study by Seckendorf and Randall (40), where the injury was anatomically verified]. The limited sparing of local thermoregulatory functions below the SCI in these studies could therefore be attributed to the preservation of descending fibers originating from supraspinal centers (14).

To clarify this point, we sought to evaluate the local response to cold in the tail of spinalized rats up to 8 wk after a complete T11 spinal cord transection. The tail is an important thermoregulatory organ in the rat, inasmuch as it plays a major role in heat loss or conservation. This role is mediated through its vasculature: the vasoconstrictor tone of the tail vasculature is controlled by sympathetic vasomotor fibers (31, 33). By performing a T11 transection, we severed descending fibers projecting to 98% of sympathetic preganglionic neurons innervating the tail arteries (45), thus depriving the tail vasculature of its central sympathetic regulation and, in effect, disconnecting it from thermoregulatory centers. We hypothesized that this injury, the level and completeness of which were fully controlled, should lead to severe disturbances of the normal vascular and thermal responses of the tail to cold. Tail arterial blood flow was recorded by Doppler ultrasonography, and surface temperature was recorded by infrared thermography.

MATERIALS AND METHODS

All experimental protocols and procedures were approved by the Animal Care and Ethics Committee of the University of New South Wales and conformed to the rules and guidelines on animal experimentation in Australia.

Seven naive male Australian Albino Wistar rats (Biological Resources Centre, University of New South Wales, Sydney, Australia; 350–400 g body wt) were housed separately in plastic home boxes (65 × 40 × 22 cm). They had ad libitum access to food and water and were maintained on a 12:12-h light-dark cycle.

Implantation of Doppler probes. The Doppler ultrasonic transducer was implanted to the ventral tail artery according to a standard protocol (12). Briefly, rats were anesthetized with a mixture of ketamine hydrochloride (100 mg/kg ip; Parnell Laboratories, NSW, Australia) and xylazine (7 mg/kg ip; Ilum Xylazil-20, Troy Laboratories, NSW, Australia). A supplementary dose of ketamine (30 mg/kg ip) was administered every 60 min following the initial dose until all surgical procedures were completed. A 15-mm longitudinal midline skin incision centered over the midportion of the coccygeal vertebrae (4th-5th coccygeal level) was made on the ventral surface of the tail, ~1 cm caudal to the anal opening. The skin was dissected from the underlying tendons and fascial sheaths. The fascia overlying the ventral tail artery was cut longitudinally, and a drop of 0.5% bupivacaine hydrochloride (0.1 ml; Pharmacia, WA, Australia) was applied to the exposed artery. After the insulated wires of the Doppler transducer (0.8-mm model E cuff probe, Iowa Doppler Products) were fed...
through an opening created laterally between the tendons and sutured to the fascia, the artery was placed within the probe cuff. The wires were tunneled subcutaneously to the dorsal thoracic region, where they were externalized.

Spinal cord transection. At 1 wk after probe implantation, rats were anesthetized with ketamine-xylazine, as described above. Hair in the dorsal thoracic region was shaved, and the area was scrubbed with an antiseptic solution. The animal was kept on a heating pad throughout the surgery to maintain core temperature at 37 ± 1°C. The operation field was infiltrated with 0.5% bupivacaine, and a 2- to 5-cm dorsal midline incision of the skin was made over the lower thoracic vertebrae. The T10 vertebra was exposed, and its laminae and spinous process were removed. The dura mater was opened longitudinally, and a drop of bupivacaine was applied to the exposed spinal cord. The spinal cord was then completely transected with microscissors, and the transection was confirmed by passage of a sharp scalpel through the lesion site several times. A block of sterile porcine gelatin sponge (2 × 1 × 1 mm; Gelfoam, Pharmacia & Upjohn, Kalamazoo, MI) was inserted in the gap between the cord stumps. Another block of Gelfoam was placed on the dorsal surface of the spinal cord to separate the nervous tissue from surrounding connective tissue. Muscles and skin were sutured in layers.

Postoperative care. The SCI animals were kept at 25–26°C. Nor-motonic sodium chloride (0.9%, 5–10 ml) was administered twice daily until the rats were able to drink normally. Animals received prophylactic cephalin sodium (60 mg/kg sc; VIC, Australia), as a preventive measure against urinary tract infections, twice daily for 5 days after SCI and the analgesic carprofen (5 mg/kg sc; Rimadyl, Australia, Australia) for 3 days after SCI. The urinary bladder was manually emptied twice daily for up to 2 wk, when autonomous bladder voiding reflexes gradually reestablished. Animals were inspected daily for skin irritation, decubitus, and hematuria, soiled animals were bathed, and minor wounds were treated.

Cold challenge. The animals were exposed to the cold environment on a weekly basis in the week preceding SCI and up to 8 wk after SCI. After 5 min in a pretest box (25–26°C), the animal was placed in a cold chamber (modified refrigerator, 6–9°C) for 20 min (Fig. 1). Immediately after the cold challenge, the rat was returned to the pretest box for 5 min. Control experiments were carried out according to the same protocol, except the chamber was set at room temperature (i.e., 25–26°C). The rats were habituated to the experimental conditions by introduction to the pretest box and the cold chamber on 2 separate days within the week preceding the first cold challenge. No data were acquired during the habituation sessions.

Blood flow in the ventral tail artery was recorded during the cold challenge and the control experiments. Data from chronically implanted Doppler transducers were acquired using a directional pulsed Doppler flowmeter (model 545C-4, University of Iowa) and MacLab Chart 5.0 software (ADInstruments, Bella Vista, Australia). The system was calibrated artificially (0 mV = 0 kHz, 0.5 mV = 1 kHz), inasmuch as the experimental settings did not allow for arterial occlusion, which is essential to obtain the real zero value of the blood flow. The data, recorded as a Doppler shift of the transmitted beam (kHz), were then converted to flow velocity (cm/s) using a conversion formula

\[ v = \frac{cf_D}{2f_c \cos \alpha} \]

where \( v \) is flow velocity, \( c \) is sound velocity in the tissue (1,540 m/s), \( f_0 \) is frequency of the Doppler shift, \( f_i \) is frequency of the transmitted beam (20 MHz), and \( \alpha \) is angle of incidence between the ultrasound beam and the direction of blood flow (45°). The measured variables (mean and pulsatile flow) were sampled at 100 Hz. Two parameters, mean arterial flow and pulse amplitude, were analyzed (Fig. 2A). Mean flow was calculated as follows: 1/3 systolic flow + 2/3 diastolic flow. Pulse amplitude was calculated as the difference between maximum and minimum flow recorded over each second (i.e., 5–9 heart cycles). The calculated values were averaged every 20 s.

Surface temperature of the rat and its immediate surroundings was obtained with an infrared thermographic camera (TheraCam P45, FLIR Systems, Boston, MA), as previously described (21, 49). Emissivity was set at 0.98 (the emissivity of skin and fur). The sensitivity and accuracy of the system were 0.1°C and 0.6°C, respectively. Thermographic images of the animals were taken from a distance of 110 cm at 1-min intervals in parallel with the blood flow recording. Single-pixel temperature values at four locations (interscapular region, sacral region, middle of the tail, and tail tip), in areas that were previously shaved to allow for direct assessment of skin temperature (Fig. 2B), were extracted from the images with ThermaCAM QuickView software. In addition, at 5 wk after SCI, the rectal temperature of the animals during the cold challenge was recorded using a digital thermometer (model BAT-10, Physitemp Instruments, Clifton, NJ). For calculation of baseline mean blood flow and temperature values, data recorded in the pretest box during the 5-min period preceding the cold challenge or the control experiment were averaged. Coefficients of variation (CoV) of mean flow and pulse amplitude

Fig. 1. Ventral tail artery blood flow during cold challenge before injury (A) and 5 wk after complete T11 spinal cord transection (B). Note negative (reverse) flow during cold challenge after transection.
(calculated as standard deviation/mean of pretest values) were used as measures of flow variation in the pretest period.

To further characterize the effect of cold exposure on changes in the recorded parameters, we compared average tail blood flow and surface temperature recorded during minutes 7 and 23 of the experiment (i.e., after 2 and 18 min of cold exposure) over the weeks before and after SCI.

Statistical analysis. Values are means ± SE. The software package SPSS 16.0 for Windows was used for all statistical analyses. Effects were considered significant if \( P \leq 0.05 \).

One-way repeated-measures ANOVA was used to compare blood flow and temperature responses across weeks (from the week before SCI through 8 wk after SCI). The repeated measure was time in minutes, and the main factor was weeks. Post hoc analyses were carried out with Dunnett’s and Tukey’s honestly significant difference tests. In addition, pulse amplitude and interscapular temperature, which showed significant or nearly significant changes in their baselines, were analyzed with a one-way repeated-measures analysis of covariance (ANCOVA), where the repeated measure was time in minutes, the main factor was weeks, and the covariate was the average of values acquired before the test (i.e., in minutes 0–5). Using this procedure, we were able to evaluate the effect of the cold exposure/control experiment while controlling for changes in baseline.

Blood flow and temperature values before the tests and at minutes 7 and 23 of the experiment (after 2 and 18 min of cold exposure) were compared with a repeated-measures ANOVA, where the repeated measure was time in weeks starting from the week before SCI to 8 wk after SCI. Within-subject effects were assessed. To adjust for the violation of sphericity assumption, we corrected between-group degrees of freedom with Huynh-Feldt epsilon. A similar analysis was used to evaluate changes in rectal temperature over 20 min of cold exposure recorded at 5 wk after SCI.

RESULTS

Baseline tail blood flow and temperature. Examples of baseline tail blood flow before and 5 wk after SCI can be seen in the 5-min (pretest) period shown in Fig. 1. In this pretest period, the most obvious difference between the two recordings was the increase in pulse amplitude, which appeared to be greater after SCI. Sudden changes in pulse amplitude could still be observed; however, they seemed to occur randomly and much less often than before SCI, where they were usually associated with alerting stimuli. These sudden drops in pulse amplitude after SCI were due to reductions in systolic blood flow, with little or no change in diastolic blood flow. In fact, diastolic blood flow remained very low in these animals, oscillating at around 0. Occasionally, small negative incursions were observed, indicating minor backflow in the artery.

Group data for tail blood flow during the pretest period (mean arterial flow, pulse amplitude, and their CoV) were compared before and 1–8 wk after SCI (Fig. 3). Although no
significant difference in mean blood flow was observed between the time points \((P = 0.3,\) by repeated-measures ANOVA), an increase in pulse amplitude after SCI was apparent and was confirmed by repeated-measures ANOVA \((P = 0.01).\) Post hoc analysis revealed a significant difference between the week before SCI and 1, 7, and 8 wk after SCI \((P \leq 0.04,\) by \(t\)-tests with Dunnett’s correction). CoV of mean flow and pulse amplitude were also significantly decreased, indicating a reduction of the natural variations in blood flow after SCI. In both instances, the CoV decreased 1 wk after SCI and persisted until the end of the study \((P < 0.001,\) by repeated-measures ANOVA; \(P \leq 0.02,\) by \(t\)-tests with Dunnett’s correction).

Surface temperature during the pretest period was measured weekly at four locations: one rostral (interscapular region) and three caudal (sacral region, middle of the tail, and tail tip) to the SCI. No significant changes in temperature of these regions were observed after SCI \((P \geq 0.07,\) by repeated-measures ANOVA). However, the sacral and interscapular skin temperatures showed opposing trends after SCI: sacral skin temperature tended to decrease, and interscapular temperature tended to increase.

Tail blood flow and temperature changes during cold exposure. The animals were exposed weekly to a cold environment \((6–9°C)\) for 20 min, while their tail blood flow and surface temperature were monitored (Figs. 1 and 4). As expected, cold exposure before SCI caused a marked drop in tail blood flow with clear reductions of its pulse amplitude and mean. This effect was maintained throughout the cold challenge. In contrast, no reduction in pulse amplitude was observed after SCI, but, surprisingly, there was still a marked reduction in mean blood flow (Fig. 4). A close look at Fig. 1 shows that pulse amplitude was maintained, because systolic and diastolic flows dropped, with clearly negative (reverse) diastolic flow. In other words, blood flow was reversed in the artery during diastole. As a result, mean flow was actually reduced to levels close to those observed before SCI (compare pulsatile traces in insets of Fig. 1; also see Fig. 2 for definition of mean blood flow).

However, Fig. 4 also shows a steep drop of the mean flow before SCI that became apparent immediately after initiation of the cold challenge \((20 \text{ s});\) after SCI, the decrease was more gradual \((5 \text{ min}).\) When the mean flow response was tested with a one-way repeated-measures ANOVA, a significant group effect \((P = 0.01).\) Post hoc Dunnett’s test showed that the difference occurred between the week before SCI and each week after SCI \((P \leq 0.03).\) The difference in pulse amplitude was confirmed by a repeated-measures ANOVA \((P < 0.001,\) group effect for weeks) and was further verified by a repeated-measures ANCOVA \((P < 0.001,\) with average pretest value of pulse amplitude as covariate) and Dunnett’s post hoc analysis (before SCI vs. each week after SCI, \(P < 0.001).\)

At 5 wk after SCI, rectal temperature of the injured animals during the cold challenge was assessed. The temperature gradually decreased by \(1°C\) over 20 min \((P = 0.004,\) by repeated-measures ANOVA; Fig. 4). Before and 1–8 wk after SCI, the cold air caused a quick and marked drop in skin surface temperature as measured by infrared thermography, an effect that is caused by a direct cooling of the skin, rather than a regulated response (23). The response was reversed when the animals were returned to ambient air at room temperature. As shown in these recordings, small differences in skin temperature, which are worth reporting, were observed between pre- and post-SCI. 1) Tail temperature appeared to drop less rapidly after SCI, consistent with the blood flow data. However, the effect was not confirmed by the one-way repeated-measures ANOVA \((P > 0.2).\) 2) Sacral temperature appeared to drop faster after SCI, whereas interscapular temperature was consistently higher throughout the cold exposure. Although the difference in sacral temperature was not statistically significant \((P = 0.4,\) by repeated-measures ANOVA), there was a difference in interscapular temperature \((P = 0.001,\) by repeated-measures ANOVA; \(P = 0.005,\) by repeated-measures ANCOVA). Post hoc Dunnett’s test revealed a difference between the week before SCI and 2, 3, 4, 5, 6, and 8 wk after SCI \((P \leq 0.01).\)

To examine in more detail early and late effects of cold exposure on blood flow and surface temperature, we assessed the values recorded at minutes 7 and 23 as for baseline (pretest) values (Fig. 5). Less pronounced decreases in mean blood flow and pulse were observed after compared to before SCI at minutes 7 and 23. At minute 7, this difference was statistically significant for both parameters \((P < 0.03,\) by repeated-measures ANOVA). The changes reached significance 2, 3, 4, 6, and 8 wk after SCI for mean flow \((P \leq 0.04,\) by \(t\)-tests with Dunnett’s correction) and 1–8 wk after SCI for pulse amplitude \((P \leq 0.005,\) by \(t\)-tests with Dunnett’s correction). At minute 23, the difference in mean flow was only a trend that did not reach the level of significance \((P = 0.07,\) by repeated-measures ANOVA), whereas the difference in pulse amplitude was significant 1–8 wk after SCI \((P < 0.01,\) by repeated-measures ANOVA; post hoc \(P \leq 0.006,\) by \(t\)-tests with Dunnett’s correction). A less pronounced reduction in interscapular temperature after SCI at minute 23 was observed and confirmed by repeated-measures ANOVA \((P = 0.01).\) Post hoc analysis found a difference between the week before SCI and 3, 4, 6, and 8 wk after SCI \((P \leq 0.04,\) by \(t\)-tests with Dunnett’s correction). Otherwise, no effect of SCI on surface temperature was seen at any time.

Tail blood flow and temperature changes during exposure to room temperature. Tail blood flow and surface temperature in control experiments were recorded with the animal in the chamber used for cold exposure, but at room temperature \((25–26°C).\) As shown in Fig. 6, exposure to the chamber did not evoke any major change in any of the recorded parameters, which remained similar to the values acquired in the pretest box.

Blood flow during the control experiment did not change significantly after SCI. No difference in mean arterial flow \((P = 0.6,\) by repeated-measures ANOVA) or pulse amplitude \((P = 0.1,\) by repeated-measures ANOVA; \(P = 0.5,\) by repeated-measures ANCOVA) was observed between pre- and post-SCI. Similarly, sacral, tail middle, and tail tip surface temperatures did not show any significant changes after SCI \((P > 0.7,\) by repeated-measures ANOVA). Interscapular temperature was higher after SCI, but there was no increase during the control experiment compared with baseline. This finding was confirmed with a repeated-measures ANCOVA \((P = 0.063).\)
Fig. 4. Mean arterial flow, pulse amplitude, and surface temperatures before and after SCI during cold challenge. Rectal temperature was recorded 5 wk after SCI. A significant effect of injury (difference between weeks) on mean arterial flow ($P = 0.01$, by repeated-measures ANOVA), pulse amplitude ($P < 0.001$, by repeated-measures ANOVA and repeated-measures analysis of covariance (ANCOVA)), and interscapular temperature ($P = 0.001$, by repeated-measures ANOVA; $P = 0.005$, by repeated-measures ANCOVA) was found. Values recorded at minutes 7 and 23 (arrows) were further analyzed (see Fig. 5).
DISCUSSION

We have shown that local responses of the rat tail to cold are not abolished by complete T11 spinal cord transection. The SCI prevents the reduction in pulse amplitude and slows the initial decrease in mean arterial flow in response to cold. However, it does not change the magnitude of the decrease in mean arterial blood flow, nor does it change the drop in tail surface temperature. In the present study, we evaluated changes in thermoregulatory responses to cold in the dermatomes caudal to the SCI in an animal model of spinal cord transection. T11 spinal cord transection allows for surgical verification and uniformity of the SCI. This level of transection leads to a disconnection of 98% of sympathetic preganglionic neurons innervating the vascular system of the tail from the supraspinal thermoregulatory centers (45). Thus the model we have used is suitable for assessment of local thermoregulatory responses free from supraspinal neural control.

Thermoregulation at room temperature (baseline). We have seen that T11 transection does not produce any significant changes in baseline mean arterial flow in the ventral tail artery. This observation is in agreement with a study by Karlsson and colleagues (19), who found no alterations in baseline arterial flow, vascular resistance, or norepinephrine spillover in lower extremities of spinal patients. However, the decrease in CoV of mean flow and pulse amplitude in the present study indicates that fluctuations in tail blood flow are markedly reduced after SCI. This reduction in blood flow variability is, in large part, due to a lack of reactivity to alerting stimuli, a well-known...

Fig. 5. Ventral tail artery blood flow and skin temperature at minutes 7 and 23 of the cold challenge (i.e., after 2 and 18 min of cold exposure) before SCI (intact) and 1–8 wk after SCI. *P < 0.05; **P < 0.01 (post hoc Dunnett’s test).
feature of tail blood flow in the intact and conscious rat (32, 54). It can therefore be directly attributed to the loss of supraspinal input (tonic, as well as phasic) to the sympathetic preganglionic neurons controlling the vasculature of the tail, a conclusion that confirms the effectiveness of our T11 transection (8, 19, 54). Interestingly, reductions in pulse and mean flow, either spontaneous or evoked by handling, could be observed after SCI. Because they were associated with a reduction in pulse amplitude (as in intact animals), it is likely that they were sympathetically mediated but originated from the isolated cord below, rather than above, the SCI. They may be part of abnormal and exaggerated reflexes similar to those observed during autonomic dysreflexia in response to stimuli caudal to the transection (22, 24).

Fig. 6. Mean arterial flow, pulse amplitude, and surface temperature before and after SCI during the control experiment at room temperature. No significant effect of injury (difference between weeks) was found.
Consistent with the baseline blood flow, SCI did not cause any significant changes in skin temperature at the four sites on the body of the animal. This indicates indirectly that T11-transected animals are able to regulate core temperature and maintain it close to homeostatic levels. Interestingly, opposite changes in baseline interscapular and sacral temperatures were noted after SCI: although the temperature of the sacral skin tended to decrease, the temperature of the interscapular skin tended to increase. The decrease in sacral temperature is consistent with a previous report from our laboratory that described a permanent drop in core temperature after T4 spinal cord transection (21). Under these circumstances, it is likely that the increment in interscapular temperature reflects an increase in the temperature of the interscapular brown adipose tissue, located directly under the skin in this region (41). Such increased brown fat metabolism could be a compensatory mechanism for the increased heat loss in spinalized animals or the result of its repeated activation by the weekly exposures to cold (5, 38, 41, 47).

Thermoregulatory responses during cold challenge. The cold challenge involves handling and transferring the rat to a different environment. To control for these components of the procedure, which could have stressed the animals, the same procedure was used, but in an environment at room temperature. Since none of the parameters show any significant changes during the control experiment, we can confidently attribute all the significant changes observed during cold exposure to the cold stimulus.

The exposure to 6–9°C was an effective cold stimulus, since it caused a moderate decrease in the rectal temperature of the SCI animals (1°C over 20 min). We have observed that the blood flow response of the ventral tail artery to cold exposure is altered in SCI animals, since the initial drop is markedly slower than in the intact animals (5 min vs. 20 s). This can be attributed directly to the loss of supraspinal input from thermoregulatory centers, the role of which, in this case, is to mount a quick, neurally mediated vasoconstrictor response to reduce heat loss. However, the minimum values of the mean arterial flow, reached before the end of the cold challenge, do not differ between pre- and post-SCI. This indicates that full tail vasoconstriction to a cold environment, which is the normal response of intact rats, still occurs in SCI rats, even though its initiation is somewhat delayed. In contrast, the cold-evoked decrease in pulse amplitude, which occurs at the same time as the reduction in mean blood flow in intact rats, does not occur in the spinalized animals. This can be attributed to a parallel decrease in systolic and diastolic blood flow, which resulted in a reduction in mean flow without an alteration of the pulse amplitude. The reduction in systolic and mean flow indicates an increase in resistance in the vascular bed supplied by the ventral tail artery. However, it is unlikely that this increased resistance was sympathetically mediated, since pulse amplitude was not reduced (in contrast to the spontaneous vasoconstrictions described above). The most likely explanation is that it was a vascular response distal to the ventral tail artery. This would also explain the negative (reverse) flow observed during diastole, because the flaccidity, or increased compliance of the ventral tail artery due to the chronic loss of basal vascular tone, would have offered less resistance than the constricted distal vasculature during the low-pressure diastolic phase.

In any case, it appears that this reduction in mean blood flow without reduction in pulse amplitude effectively reduces heat loss from the tail, since its surface temperature during the cold challenge does not differ significantly between SCI and intact animals. In fact, the whole body response to cold may have been as effective in the SCI rats as in the intact rats, since no significant differences in sacral temperature were observed. We also observed that the interscapular temperature was higher in the SCI rats during cold exposure. This difference may reflect the increased thermogenic capacity of the interscapular brown adipose tissue in the spinalized rats, as suggested above, and most probably results from an increased drive from thermoregulatory centers in response to cold (28). It would have contributed significantly to the whole body thermoregulatory response of the SCI animals to cold.

Alterations of blood flow and skin temperature occurred by 1 or 2 wk after SCI and, thereafter, remained relatively stable, until the end of the study at 8 wk after SCI. This observation suggests that ventral tail artery blood flow and skin temperature neither recover nor further deteriorate with the advance of SCI into a more chronic phase.

Mechanisms for vascular responses to cold below SCI. The data obtained in the present study suggest that spinal cord transection does not sever all mechanisms responsible for effective thermoregulatory responses in the rat tail. We can consider several mechanisms that could contribute to the preserved thermoregulatory reactions to cold after the central sympathetic control of the vascular tone, provided by descending pathways, is lost. The first possibility, which, as discussed above is unlikely, is that the mechanism might be sympathetically mediated and originate from the isolated portion of the spinal cord. It is well known that a spinal cord disconnected from supraspinal regulation is capable of producing reflex changes in vascular tone in response to afferent input (7, 52, 53), as occurs in the clinical condition of autonomic dysreflexia (8, 11, 46). Indeed, in a study on an acute preparation of isolated neonatal rat spinal cord by McKenna and Schramm (26), sympathetic preganglionic neurons were sympathetically activated by stimulation of dorsal root neurons. However, there is no evidence that similar output can be driven by peripheral thermal stimuli. There is evidence of thermal sensors within the spinal cord itself (29, 44, 51), but because of the time course of the responses seen here, it is unlikely that the decrease in blood flow was driven by a drop in spinal cord temperature. The second, and more likely, possibility is that the vasoconstriction in the vessels caudal to the spinal transection is a direct response of the peripheral vasculature to cold. An in vivo study in tetrodotoxin-treated animals showed that the cutaneous circulation is regulated locally by direct action of cooling on the skin (17). In addition, it was previously reported that local cooling leads to a sensitization of the vessels to norepinephrine (9, 15, 48), which is mediated through an increase in expression of α2C-adrenoceptors and ATP secretion (ATP secretion stimulates secretion of norepinephrine through preganglionic P2 purinoceptors) (2, 18, 20). Cooling also increases reactivity of α2C-adrenoceptors to circulating catecholamines (17). It is known that cold exposure results in an increase in levels of plasma catecholamines and that the vasoconstriction in the tail artery is mainly triggered via activation of α1- and α2C-adrenoceptors (4, 6, 25, 53). Thus, in T11-transected rats, where the substantial part of the sympathetic
output to cutaneous vasculature and adrenal glands is rostral to the SCI, the elevated levels of circulating catecholamines of adrenal or sympathetic origin (a consequence of the reaction to the cold challenge rostral to the lesion) could lead to stimulation of the cold- and SCI-sensitized tail vasculature and, subsequently, to vasoconstriction (1, 42, 50, 53). This hypothesis is supported by the fact that the cold-evoked drop in mean blood flow in the ventral tail artery, reported in the present study, occurred with some delay in the spinal animals, as opposed to intact rats, which also suggests hormonal regulation.

Interestingly, a similar mechanism of abnormal vascular reactivity involving α2-adrenoceptors has been proposed for Raynaud’s phenomenon, a disease characterized by unusually strong vasoconstriction in fingers and toes in response to cold (for reviews see Refs. 3 and 16). It may well be that the effects we have described here in SCI rats have the same pathophysiological origin as the symptoms of Raynaud’s phenomenon.

Clinical implications. The present data strongly suggest that complete SCI does not abolish local thermoregulatory responses to cold at locations below the lesion. However, these responses are slower than normal, which may partly explain the increased susceptibility of SCI patients to changes in environmental temperature. Clearly, species differences may also be important. Improved knowledge of the mechanisms controlling local vascular responses to cold would allow for development of therapeutic strategies targeting improved temperature homeostasis in SCI patients. It may also be used in the clinical management of some disorders of vascular reactivity, such as Raynaud’s phenomenon.

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