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# Current understanding on the neurophysiology of behavioral thermoregulation

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**Keywords:** grooming, thermogradient, thermo-TRP, operant thermoregulatory behavior

**Abbreviations:** AV3V, anteroventral third ventricle; DMH, dorsomedial hypothalamic nucleus; KO, knockout; LPS, lipopolysaccharide; MnPO, median preoptic nucleus; POA, preoptic area of the hypothalamus; $T_c$, core body temperature; TRP, transient receptor potential.

Temperature influence on the physiology and biochemistry of living organisms has long been recognized, which propels research in the field of thermoregulation. With the cloning and characterization of the transient receptor potential (TRP) ion channels as the principal temperature sensors of the mammalian somatosensory neurons, the understanding, at a molecular level, of thermosensory and thermoregulatory mechanisms became promising. Because thermal environment can be extremely hostile (temperature range on earth's surface is from $\sim -69^{\circ}$C to 58°C), living organisms developed an array of thermoregulatory strategies to guarantee survival, which include both autonomic mechanisms, which aim at increasing or decreasing heat exchange between body, and ambient and behavioral strategies. The knowledge regarding neural mechanisms involved in autonomic thermoregulatory strategies has progressed immensely compared to the knowledge on behavioral thermoregulation. This review aims at collecting the up-to-date knowledge on the neural basis for behavioral thermoregulation in mammals in order to point out perspectives and deployment of this research field.

## Introduction

Thermoregulation research has faced a burst in the last 2 decades due to the recognition of the importance of temperature for the complex interactions among physiological, pathological, psychological and environmental factors.[1-5] Homeothermic animals developed the ability of regulating core body temperature ($T_c$) within a narrow range, optimal for cellular and molecular function, even when facing sudden and large variations of environmental temperature. This amazing capacity is due to the orchestration of both behavioral (see below) and autonomic thermoeffectors, e.g. autonomic control of heat loss/conservation through cutaneous vasomotion or water evaporation through skin or respiratory tract, and thermogenesis in special tissues such as brown adipose tissue or through shivering in skeletal muscles.[4] Although the development of autonomic thermoregulatory mechanisms were pivotal for the maneuver of homeothermia, its ability to compensate for thermal loads is limited by body resources such as water (heat loss) and energy (thermogenesis). As a result, behavioral thermoregulation mechanisms are the most efficient available for animals to compensate for greater thermal loads, without compromising valuable body resources. Yet, the nervous system mechanisms and pathways regarding behavioral regulation of body temperature in response to environmental thermal challenges remains a central question in neuroscience.

In 1975, Cabanac[6] pointed to behavior as a "new trend in physiological investigation" with the recognition of behavior as a physiological response. In the face of homeostasis-threatening thermal fluctuations of an environment, behavioral thermoregulation consists of a broadly conserved mechanism which potentially increases the chance of survival of several species.[2,7] In addition, thermoregulatory behavioral adjustments may be interpreted over evolutionary time scales a factor which may have substantial influences on the ecological niche of species. Species can dwell in specific environments basically due to their anatomical and/or biochemical/physiological adaptations to environmental stressors. Such adaptations ensure survival and, consequently, the possibility of procreation of individuals in their natural habitat, even if ambient temperature is considered permanently hostile (e.g., desert regions) or becomes so seasonally or occasionally.[8]

The skin is an important organ that influences in behavioral thermoregulatory mechanisms. Different from autonomic


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thermoregulation that use only feedback signals,[9,10] thermoregulatory behaviors use peripheral thermal feedback signals from the skin and feedforward signals for complex behaviors, like explore the environment using non-hairy skin.[10,11] The spinothalamocortical pathway, in which thermal signals from skin are transmitted by neurons in the lamina I of the spinal cord, and reach the primary somatosensory cortex through a relay in the posterior part of the ventral medial nucleus in the thalamus,[12,13] is mainly responsible for perception and discrimination of cutaneous temperature. Recently, a distinct somatosensory pathway was identified and described as responsible for activation of autonomic defense mechanisms against environmental thermal challenges partially independent from the thalamus. This pathway involves a projection from dorsal horn to the preoptic area of the hypothalamus (POA) with a relay in the external lateral and central parts of lateral parabrachial nucleus (cold signals[14,15]) and in the dorsal part of the lateral parabrachial nucleus, (warmth signals[14,16]) however this pathway uses, even partially, the same signals from the skin pathway, which is thalamus-dependent as describe above.

For autonomic control of body temperature, POA plays an incontestable role orchestrating different neural pathways for each autonomic thermoeffector. From there (POA), neural signals are distributed to distinct neural circuitries which separately control different autonomic thermoeffector, such as brown adipose tissue thermogenesis, peripheral vasomotion and shivering thermogenesis.[17] Although there is evidence suggesting that different thermoregulatory behaviors in the rat (e.g., relaxed postural extension, thermoregulatory grooming, and locomotion) also use distinct neural circuitries,[18,19] the neuroanatomic substrate of no thermoregulatory behavior has been studied extensively. Besides, there is still no evidence of a brain area that plays such a *maestro* role for behavioral thermoregulation as POA plays for autonomic thermoeffectors. Our purpose in this review is to gather what is known regarding the neural mechanisms and brain areas that control some of the most important thermoregulatory behaviors in mammals.

## Behavioral Thermoregulation

### Selection of ambient temperature

Energy expenditure on thermoregulation may somehow impair ontogeny and potentially undermine reproductive success, different from plants in which metabolic expenditure on thermoregulation cannot be naturally reduced by means of relocation.[20] Selection of an ambient temperature (also referred to as warmth- or cold- seeking/avoidance behavior) is a very conservative thermoregulatory strategy; perhaps the most conservative one.

In the laboratory, this behavior can be studied either at a thermogradient apparatus or at a chamber preference setup (**Fig. 1**). The thermogradient consists of a setup with a longitudinal ambient temperature gradient, usually ranging from subneutral cold to supraneutral warmth temperatures, and the animal can choose between a wide range of ambient temperatures.[7] In the chamber preference setup, also known as 2-temperature choice setup, 2 options of thermal environments is offered for the animal (e.g., cold vs neutral, warmth vs neutral, cold vs warmth), and thermoregulation will result from the movement of the animal between the 2 compartments of the chamber.[21] Warmth- and cold-seeking behavior are commonly evoked in a multitude of species belonging to almost all classes of metazoans by significant decreases or increases in ambient temperature — compelling individuals to move to a warmer or colder region/environment to escape serious injury or even death and/or to improve welfare.[2,7] Search for the preferred ambient temperature may also be triggered from stimuli that activate neuro-immune-endocrine responses, such as fever and septic shock (hypothermia followed by fever; for review see Ref.[1]) or reduces $O_2$ availability (hypoxia).[22] The chance of selecting an ambient temperature of preference can be crucial for determining the type of

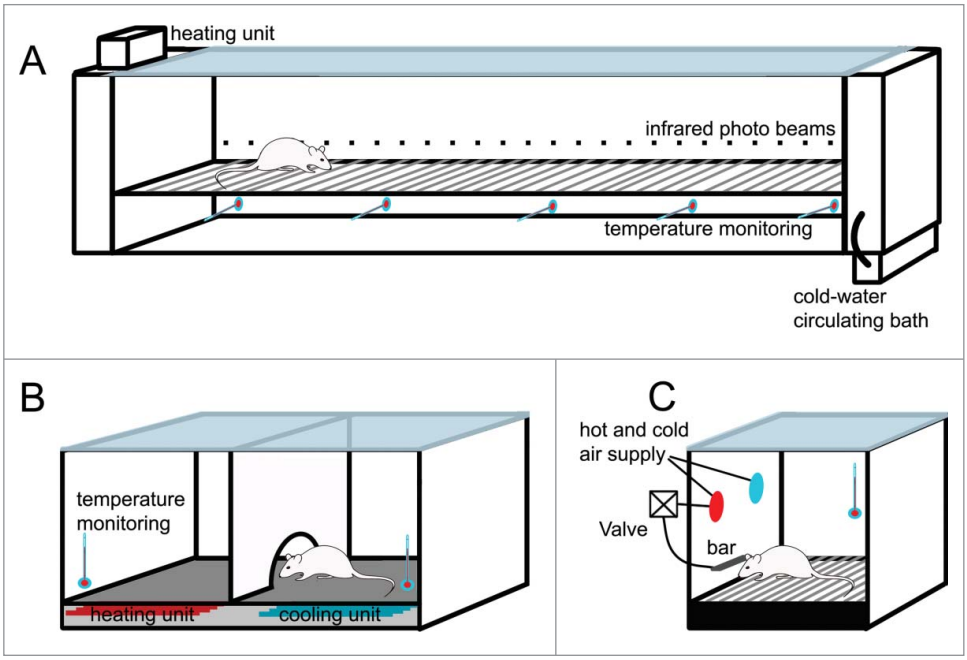


**Figure 1.** Schematic of different apparatus used for the study of behavioral thermoregulatory responses in the laboratory. (**A**) thermogradient apparatus. (**B**) 2-temperature choice setup. (**C**) operant behavior setup. In **A** and **B**, by registering the position of the animal in the apparatus either using infrared photo beams or a camera recorder (not shown in the schematics), the selected ambient temperature can be calculated. In **C**, the parameter analyzed is the ambient temperature. In operant behavior, animals learn to adjust its own environmental temperature by means of pressing a bar or moving to a reward area in order to receive either heat or cold air reinforcement.

thermoregulatory response an animal will display. For instance, Almeida and colleagues[7] have demonstrated that rats treated with a high dose of lipopolisaccharide (LPS) respond with cold-seeking behavior and hypothermia followed by warmth-seeking behavior and fever, whereas the same LPS dose applied to animals kept a subneutral environment develops a greater hypothermic response.[1]

Although the importance of this behavior (selection of ambient temperature) for thermoregulation is unquestionable, the neural pathways controlling this behavior are not completely understood. Studies aimed at understanding the neural pathways controlling behavior started more than 5 decades ago, and used electrodes to stimulate and/or lesion of different areas combined with observation of different types of behavior. By warming regions described as the septal area, ventral midbrain and dorsal medulla, Roberts and Mooney[19] observed an increase in the locomotor activity of rats, which might be related to an attempt to escape an environment and find a more pleasant one. Nevertheless, a later study showed that lesioning these same areas did not affect warmth-induced locomotion,[23] ruling out the participation of these areas in the neural circuitry for the control of behavioral thermoregulatory responses. Interestingly, POA warming or lesioning did not affected warmth-induced locomotion,[19,23] indicating that this area might not be related to behavioral thermoregulation. Later, the hypothesis for POA involvement in the circuitry for the selection of ambient temperature was excluded: by placing a large bilateral electrolytic lesion in the preoptic area, and using different stimuli [physical stimulus– heat and cold; chemical stimulus–agonists of thermo- transient receptor potential (TRP) channels; and inflammatory stimulus – LPS] to induce thermal preference behavior, the authors showed that although all autonomic responses were severely compromised in the POA-lesioned rats, none of the behavioral responses to the different stimuli used were compromised in these animals,[7] excluding the role of POA in thermal preference in mammals.

Involvement of other hypothalamic structures, *viz.*, paraventricular nucleus, or dorsomedial nucleus (DMH), in thermopreferendum was studied in rats stimulated thermally (cold and heat exposure), pharmacologically (by using thermo-TRP channels agonists) or immunologically (LPS challenge). From all behaviors studied, LPS-induced cold-seeking response was the only behavior found to be mediated by neuronal bodies located in the DMH and neural fibers passing through the paraventricular hypothalamic nucleus, which comprise to date the first 2 landmarks of the map of the circuitry of cold-seeking behavior associated with endotoxin shock (**Table 1**).[7]

**Operant thermoregulatory behavior**

Behavioral thermoregulation can also be studied in operant behavior systems (**Fig. 1**). In this paradigm, the animal is placed at a hot or cold environment and will receive a warm or cool reward every time a required action is performed (such as bar pressing or moving to a reward area). This apparatus requires extensive training of the animals, because the action must be learned, yet it has been used in the investigation of the brain areas controlling thermoregulatory behavior.

Because POA has been considered to be the major integrator for the autonomic responses, this was the first site to be investigated when researchers tried to first establish a central neuronal pathway for behavioral thermoregulation. By using central thermodes, it has been shown that local cooling or heating of anterior hypothalamus and POA produces the behavioral motivation for heat and cold,[24,25] indicating that this area may function as a thermosensitive site. In agreement, Fos-like immunoreactivity is observed in the median preoptic nucleus (MnPO) of rats having performed a heat-escape behavior, but also in rats passively heated to the same ambient temperature achieved by animals performing the operant behavior,[26] indicating this area as a thermosensitive, but not necessarily involved in the genesis of the thermoregulatory behavior. Indeed, lesioning of POA has further proved that this area is not involved in the neural circuitry for operant thermoregulatory behavior, as rats trained to press a bar to escape heat showed an increase in this response after bilateral preoptic lesions, being able to maintain stable $T_c$ behaviorally.[27,28] Using a similar approach, Satinoff and Rutstein[29] demonstrated that rats with anterior hypothalamic bilateral lesions could not thermoregulate autonomically, but they kept their ability to thermoregulate behaviorally in a heat-reinforcement operant procedure. Although neurons in POA might not be related to thermal stimulus inducing operant thermoregulatory behavior, there is evidence that this circuitry might be involved in heat escape behavior induced by systemic salt-loading, as ibotenic acid lesions of MnPO inhibits this behavior in rats.[30]

In the early 70′s, the lateral hypothalamus appeared as a promising target for the start of unraveling the neuronal behavioral thermoregulatory pathway, when rats with electrolytic lesion of the lateral hypothalamic area were shown to present severe deficiency in bar pressing for heat-reinforcement.[31] Lateral hypothalamic lesions however, often cause motor deficits of various types as well as a reduction in general motivation, with impairment of behaviors such as feeding, drinking, and shock avoidance, suggesting that impaired thermoregulatory behavior in lateral hypothalamus-lesioned rats is a secondary consequence of motor/motivational impairment. Indeed, further studies that re-evaluated the role of lateral hypothalamus in behavioral thermoregulation, found a similar or even improved performance of lateral hypothalamus-lesioned over sham-operated animals,[32,33] thus discarding the participation of this structure in the neuronal circuitry for behavioral thermoregulation.

Hypothalamic structures are not completely ruled out from the neural circuitry for operant thermoregulatory behavior. Fos-like immunoreactivity is observed in the parastrial nucleus and DMH in rats having performed heat-escaping behavior, but not in rats passively warmed, suggesting these sites as candidates pieces in the circuitry for the genesis of operant behavior.[26] Interestingly, DMH is also a region on the map of the circuitry of cold-seeking behavior associated with endotoxin shock (**Table 1**).[7]

**Posture changes/body extension**

Body extension consists of "relaxed elongation of the body in a prone position with curved spine straightened horizontally,

hind legs flexed toward the sides and rear, and forelegs folded beneath the neck." The head remains in an alert state, but is soon lowered to the floor. This behavior has been observed in different species such as rat and dog, and is important as it alters the ratio surface/volume of the body, facilitating heat transfer to the environment (**Fig. 2**). Different from other thermoregulatory behaviors that are elicited by changes in the skin temperature, prone extension behavior has been shown to be elicited independent from skin temperature, being completely dependent on central sensors.[34] Interestingly, this is also the only thermoregulatory behavior known to include POA in its circuitry control: POA warming elicits relaxed prone extension,[19] whereas POA lesion markedly reduced or eliminated the heat-induced postural extension response.[23]

In addition to the involvement of POA, the periventricular tissue around the anteroventral third ventricle (AV3V) has been reported in relaxed prone extension. In this study, rats with lesion of this region, which included injury in rostral periventricular tissue and medial edge of the medial preoptic nucleus, the organum vasculosum of the lamina terminalis and the ventral part of the MnPO, shown decline in adoption of the the heat-induced body extension (**Table 1**).[35]

### Grooming

Grooming is a common and robust behavior in rodents, and consists of stereotyped forms such as licking, scratching, and face-washing,[36] representing great part of behavior presented in waking time and is triggered by many factors, such as predator stress, injection of drugs, as well as heat,[37] being an important thermoregulatory behavior of heat defense in rats.[38] This behavior allows the spread of saliva onto the body surface and, consequently, promotes the evaporative heat loss (**Fig. 2**).[39] Grooming is one of the main behaviors used by rats for heat loss. For example, at 40°C of ambient temperature, grooming is the mechanism responsible for more than half of the heat lost.[40] It can be studied by observation of the animal, usually recording the behavior for further analyses of the time spent in grooming. As shown by Roberts,[41] rats, guinea pigs and gerbils have an increase of more than 180% in grooming at high ambient temperatures (> 37.7°C). However, other species such as mice and hamster decreased time spent grooming in warmth. Interestingly, the reduced grooming of mouse does not mean that grooming is not a thermoregulatory behavior. The salivary efficiency increases the wetting and compensates the shorter duration, observed by the elevated water loss, around 60%. Hamsters have an efficient evaporative cooling via panting and/or an altered pattern of air movement through the nasal and oral cavities as well.[41]

An efficient thermoregulatory grooming behavior depends on increase in the salivary production, making grooming a mixed autonomic/behavioral thermoeffector. Some brain regions have been reported to be involved in salivation, such as lateral,[42] anterior,[43] and ventromedial hypothalamus,[44] and the periventricular AV3V.[35,45,46] In these studies, it has been shown that lesion in these areas promotes decrease in heat-induced salivation level and, consequently, deficiency in heat loss, promoting increase in $T_c$. Moreover, the organum vasculosum of the lamina terminalis, MnPO and subfornical organ, which together form lamina terminalis, lateral and medial portion of POA

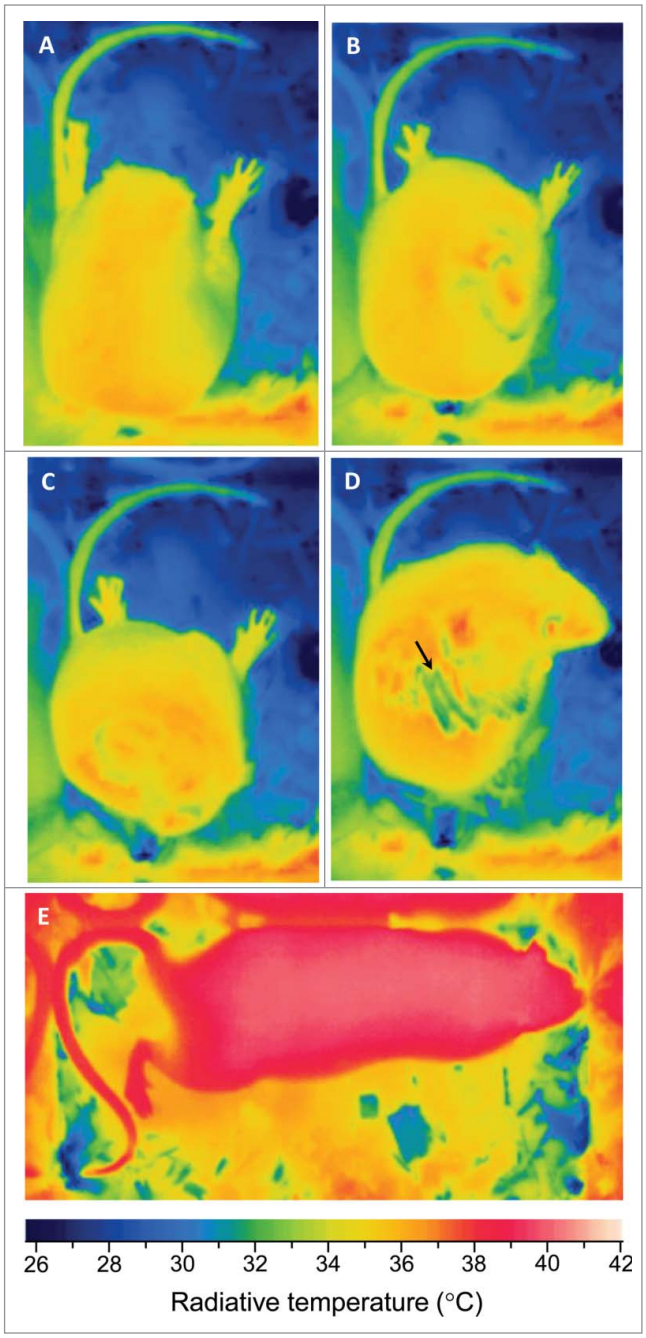


**Figure 2.** Infrared thermograms (unpublished observations) of a rat exposed to supraneutral ambient temperature (~30°C, top view) engaged in a series of grooming behavior, including medial (**A**) and lateral body licking (**B—C**). Note in D the reduction in skin temperature resultant from licking (arrow). In E, a rat exposed to a warmer ambient temperature (~36°C) is expressing prone extension behavior. To obtain this pictures, male Wistar rat (14 weeks old) was exposed individually to ~30 or ~36°C in a temperature-controlled chamber (Eletrolab; Sao Paulo, SP, Brazil) and the behavior of the animal was recorded by an infrared camera (Flir, A300 – Series; Portland, OR, USA) for further analysis of the video. The grooming images were taken in the first 15 min of exposure to ~30°C and prone extension image was taken after 60 min at ~36°C.

**Table 1.** Involvement of hypothalamic structures in thermoregulatory behavior in rats

| Behavior | POA/MnPO | Hypothalamic Area DMH | Posterior Hypothalamus |
|---|---|---|---|
| Selection of Ta | Not involved[5,17,20] | Involved LPS-induced cold-seeking behavior[5] | — |
| Operant Behavior | Involved Salt-loading induced heat scaping behavior[28] | Involved Thermal stimuli induced Fos-like Immunoreactivity[24] | — |
| Body Extension | Involved Warmth-induced prone extension[39] | — | — |
| Grooming | Involved Warmth-induced salivation* | — | Involved Warmth-induced grooming[36,39] |

Ta: ambient temperature.
*other hypothalamic areas were also described for salivation. See text for details and references.

and the insular cortex was shown to have efferent neural pathways to salivary glands.[47,48]

Although salivation and grooming are related and occur in a paired way, they are stimulated independently: salivary secretion can start in a lower temperature threshold and by another kind of stimuli such as taste or bouts of grooming.[49] Furthermore, thermoregulatory grooming behavior and salivation elicited by heat seem to be controlled by different neural pathways,[49] while the anterior hypothalamus has been reported to induce the salivation by warm stimulus,[50] the posterior hypothalamus produce grooming,[19,38] and this behavior is not impaired in rats with lesion in periventricular AV3V,[35] unlike salivation, as described above (**Table 1**).

## Importance of Thermal Sensation for the Initiation of Behavior: Involvement of TRP Channels

As stated above the organisms respond to changes in ambient and body temperature with different behavioral effector. The ability to perceive environmental stimuli is an important adaptive skill, which allows animals to preview thermal ambient conditions minimizing energy expenditure to guarantee internal thermal balance. Although the neural pathways to the behavioral thermoregulatory mechanisms remain largely unknown, the knowledge regarding thermal signal transduction has advanced greatly within the past 2 decades due to the cloning and characterization of thermally sensitive TRP channels, also known as the thermo-TRPs. The understanding of the mechanisms of thermal transduction can be important for guiding the unveiling of the neural pathways that control thermoregulatory behaviors. Thermo-TRPs are activated in a wide range of ambient temperatures, with TRPV1-V4, TRPM2, M4 and M5 activated by warmth, and TRPA1 and TRPM8 activated by cold.[17,51-55] For some thermo-TRP channels physiological functions as thermoreceptors have been proposed, examples include the TRPV3,[56] TRPV4,[21,57] and TRPM8,[52,58,59] and there is some evidence of their involvement in the initiation of thermoregulatory behaviors, as we will discuss below.

### TRPM8

The characterization of TRPM8 channel allowed a breakthrough in understanding of the mechanisms for the detection of cold stimuli.[60-63] This channel is activated *in vitro* by temperatures below ~25°C, and also by chemical compounds such as icilin and menthol.[51] TRPM8 is expressed in small diameter neurons in dorsal root ganglion and trigeminal neurons,[60-62] which is consistent with a role of these channels in thermoreception. Furthermore, there is no co-expression of TRPV1 with TRPM8, indicating that TRPM8 channel is expressed in a population of neurons distinct from those sensitive to warm temperatures.[62]

Primary evidence from TRPM8 role as a thermoreceptor comes from studies that used knockout (KO) animals. In behavioral experiments, TRPM8-KO mice exhibited a strong deficiency in the ability to discriminate between moderately warm and cold temperatures when submitted to 2 temperature preference assay[58,59,64] or to thermal gradient assay.[52] In these studies, null TRPM8 mice spend more time in innocuous and noxious cold temperatures than wild-type mice, while keeping aversion to noxious heat. Furthermore, not only TRPM8-KO animals exhibited this alteration in thermoregulatory behavior, but also adult rats that had ablation of TRPM8-expressing neurons also behave similarly.[65]

In two of these studies, the number of crossings between one ambient and another in animals subjected to 2 temperature preference assay was also studied; this parameter can evaluate cold-avoidance.[64,65] While the control mice reduced the number of crossings when one of the plates was cooled and the other was maintained at 30°C (indicating cold-avoidance), the TRPM8-KO mice and those that had depletion of TRPM8-neurons showed greater thermal mobility not only to innocuous but also for noxious cold temperatures, supporting the hypothesis that these animals display a deficit in the detection of cold stimuli.

Corroborating these findings, mice treated with the TPRM8 antagonist "compound 5" exposed to 2 temperature preference assays (22 vs 30°C), spent more time in the cold (22°C) than in the warmth side (30°C) compared to mice treated with vehicle, showing similar behavior to TRPM8- KO mice, demonstrating that mice sensitivity to cold temperatures was decreased by inhibition of TRM8 channels.[66]

In studies using chemical stimulation, activation of TRPM8 with low intravenous doses of menthol[2,7] or topical application of menthol,[67] warmth-seeking behavior was observed in rats exposed to thermogradient. Similar results were also observed in

mice.[68] Interestingly, M8-B, another antagonist of TRPM8 channels, was shown to attenuate both menthol-induced warmth-seeking behavior and cold-induced increase in c-fos expression in the lateral parabrachial nucleus (LPB),[67] a brain region known to receive cutaneous cold signals from primary sensory neurons,[14,15,69,70] and send these cold signals to the preoptic area of the hypothalamus (POA),[15,71] suggesting that the neural pathway activated to initiate cold-induced warmth seeking behavior may pass through LPB as a result of activation of TRPM8 termorreceptors.

**TRPV1, TRPV3 and TRPV4**

TRPV1 is a non-selective cation channel, highly expressed in small diameter sensory neurons, which can be activated by noxious temperatures > 42°C, protons and chemical agonists as capsaicin.[72,73] Although this channel has been characterized as a thermosensor, evidence shows that the thermoregulatory response evoked by TRPV1 is independent from body or ambient temperature and occurs also at temperatures that are out of activation range of this channel.[74-76]

The TRPV3 and TRPV4 channels are activated by innocuous warmth temperatures (range from ~33 to 39°C and ~25 to 34°C, respectively[77-81]) and they are the 2 main candidates to function as moderated warmth receptors involved in heat defenses in mammals. Other than innocuous warmth, stimuli such as chemical (e.g., camphor, garlic extracts, oregano) can also stimulate TRPV3,[51,82] while TRPV4 are also stimulated by chemical (e.g., 4-αPDD, arachidonic acid metabolites, hyposmolarity, low pH) or mechanical stimuli (shear stress).[83-89]

Consistent with its thermosensitivity *in vitro*, the protein of TRPV3 and V4 channels is located in some elements compatible with thermoregulatory function. TRPV4 is found in endothelial cells, sweat glands, primary sensory neurons and sympathetic afferent neurons in the POA,[77,80,90,91] and both channels are highly expressed in stratified epithelial cells and keratinocytes,[77,78,82,92] suggesting that skin TRPV3 and V4 channels can preliminarily detect warmth stimuli. Indeed, there is evidence that the transduction of thermal stimuli from skin TRPV3 channels to sensory neurons occurs via ATP.[93]

Evidence has supported the participation of TRPV3 channels in behavioral thermoregulation. In studies with ablation of TRPV3 channel, it has been shown that KO mice exposed to a thermogradient behave different from wild wild-type mice. In one study, while WT animals remain most of the time between 30 – 38°C in a thermogradient (temperature range between 15 to 55°C), selecting this temperature range 25 min after starting the test, TRPV3-KO mice showed a preference for this temperature range only after the first hour assessment and, before that time, showed no preference for any temperature.[56] Another study observed that TRPV3 KO mice occupancy in a thermogradient displayed a sharp bias toward cooler temperatures when compared to controls.[94] In another test, using the 2-temperature preference assay, the authors demonstrated that both wild-type and TRPV3-KO have aversion to cold, i.e., spend more time at room temperature when allowed to select between this temperature *vs* cold (15°C). However, when the animals had to choose between room temperature *vs* warmth (35°C), wild-type mice showed a clear preference for the warmth environment (spent more than 90% of the time at 35°C), significantly different from TRPV3-KO mice, which spent only about 65% of the time at 35°C.[56]

It has been proposed that the TRPV4 channel also acts as a thermoreceptor. The involvement of TRPV4 channels in thermoregulatory behavior was preliminarily studied in TRPV4-KO mice that demonstrated preference to warmer temperatures compared to wild type animals when undergoing thermal gradient or 2-temperature selection tasks.[57] Corroborating this result, a recent publication using pharmacological treatment demonstrated that chemical activation of TRPV4 (probably the cutaneous TRPV4 channels) by topical application of TRPV4-agonist leads rats to choose colder temperature in 2-temperature preference assay and in thermogradient assay. In addition, TRPV4-blocking with intravenous administration of a selective antagonist of this channel inhibited warmth-induced cold-seeking behavior in thermogradient.[21]

There is a single study, using the same strain used in the study of Moqrich et al,[56] that has shown that TRPV3-KO mice exhibited no clear alterations in thermoregulatory behavior, although when studying a different strain (TRPV3-KO 129S6), the authors showed a discrete alteration, in which mice preferred a narrower region of cooler temperatures.[95] Furthermore, double TRPV3 and TRPV4-KO C57BL6 mice behavior did not differ from wild-type mice in the thermal gradient, which led the authors to conclude that TRPV3 and TRPV4 ion channels are not major contributors to mouse heat sensation.[95] However, negative results with KO animals are difficult to interpret, because in these animals compensatory mechanisms may occur,[96,97] especially in this case where there is a double TRPV3 and TRPV4- KO. Together, these data and other evidence described above points to a role for TRPV3 and TRPV4 channels in thermoregulation.

Together, these data corroborate the hypothesis that the selection of an appropriate ambient temperature is initiated by signals generated at peripheral shell thermoreceptors, probably involving thermo-TRP channels. Although the afferent and efferent neural circuitries for each behavioral thermoregulatory response is still little established, there is evidence that, at least for cold/TRPM8 dependent signals, the spino-parabrachial pathway might be involved,[67] but whether warmth signals also activate this pathway is still a matter of investigation. The majority of thermoregulatory behaviors studied (except for prone extension behavior) are independent from the integrity of POA, and finding the brain area(s) responsible for integrating these thermal signals and converting into behavioral thermoeffectors is nowadays a challenge in neuroscience.

**Disclosure of Potential Conflicts of Interest**

No potential conflicts of interest were disclosed.

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## References


1. Romanovsky AA, Almeida MC, Aronoff DM, Ivanov AI, Konsman JP, Steiner AA, Turek VF. Fever and hypothermia in systemic inflammation: recent discoveries and revisions. Front Biosci 2005; 10:2193-216; PMID:15970487; http://dx.doi.org/10.2741/1690
2. Almeida MC, Steiner AA, Branco LG, Romanovsky AA. Cold-seeking behavior as a thermoregulatory strategy in systemic inflammation. Eur J Neurosci 2006; 23:3359-67; PMID:16820025; http://dx.doi.org/10.1111/j.1460-9568.2006.04854.x
3. de Paula CA, Santiago FE, de Oliveira AS, Oliveira FA, Almeida MC, Carrettiero DC. The Co-chaperone BAG2 Mediates Cold-Induced Accumulation of Phosphorylated Tau in SH-SY5Y Cells. Cell Mol Neurobiol 2015; PMID:26208804
4. Flouris AD, Piantoni C. Links between thermoregulation and aging in endotherms and ectotherms. Temperature 2015; 2:73-85; http://dx.doi.org/10.4161/23328940.2014.989793
5. Matsuzaki K, Katakura M, Sugimoto N, Hara T, Hashimoto M, Shido O. β-amyloid infusion into lateral ventricle alters behavioral thermoregulation and attenuates acquired heat tolerance in rats. Temperature 2015; 2:4, 418-24; http://dx.doi.org/10.1080/23328940.2015.1044635
6. Cabanac M. Temperature regulation. Annu Rev Physiol 1975; 37:415-39; PMID:123725; http://dx.doi.org/10.1146/annurev.ph.37.030175.002215
7. Almeida MC, Steiner AA, Branco LG, Romanovsky AA. Neural substrate of cold-seeking behavior in endotoxin shock. PLoS One 2006; 1:e1; PMID:17183631
8. Humphries MM, Thomas DW, Speakman JR. Climate-mediated energetic constraints on the distribution of hibernating mammals. Nature 2002; 418:313-6; PMID:12124621; http://dx.doi.org/10.1038/nature00828
9. Werner J. System properties, feedback control and effector coordination of human temperature regulation. Eur J Appl Physiol 2010; 109:13-25; PMID:19787369; http://dx.doi.org/10.1007/s00421-009-1216-1
10. Romanovsky AA. Skin temperature: its role in thermoregulation. Acta Physiol 2014; 210:498-507; http://dx.doi.org/10.1111/apha.12231
11. Cheung S. Responses of the hands and feet to cold exposure. Temperature 2015; 2:105-20; http://dx.doi.org/10.1080/23328940.2015.1008890
12. Craig AD. How do you feel? Interoception: the sense of the physiological condition of the body. Nat Rev Neurosci 2002; 3:655-66; PMID:12154366; http://dx.doi.org/10.1038/nrn894
13. Craig AD, Bushnell MC, Zhang ET, Blomqvist A. A thalamic nucleus specific for pain and temperature sensation. Nature 1994; 372:770-3; PMID:7695716; http://dx.doi.org/10.1038/372770a0
14. Morrison SF, Nakamura K. Central neural pathways for thermoregulation. Front Biosci 2011; 16:74-104; http://dx.doi.org/10.2741/3677
15. Nakamura K, Morrison SF. A thermosensory pathway that controls body temperature. Nat Neurosci 2008; 11:62-71; PMID:18084288; http://dx.doi.org/10.1038/nn2027
16. Nakamura K, Morrison SF. A thermosensory pathway mediating heat-defense responses. Proc Natl Acad Sci U S A 2010; 107:8848-53; PMID:20421477; http://dx.doi.org/10.1073/pnas.0913358107
17. Romanovsky AA. Thermoregulation: some concepts have changed. Functional architecture of the thermoregulatory system. Am J Physiol Regul Integr Comp Physiol 2007; 292:R37-46; PMID:17008453; http://dx.doi.org/10.1152/ajpregu.00668.2006
18. Roberts WW. Differential thermosensor control of thermoregulatory grooming, locomotion, and relaxed postural extension. Ann NY Acad Sci 1988; 525:363-74; PMID:3291670; http://dx.doi.org/10.1111/j.1749-6632.1988.tb38620.x
19. Roberts WW, Mooney RD. Brain areas controlling thermoregulatory grooming, prone extension, locomotion, and tail vasodilation in rats. J Comp Physiol Psychol 1974; 86:470-80; PMID:4592495; http://dx.doi.org/10.1037/h0036147
20. Suguiyama VF, Silva EA, Meirelles ST, Centeno DC, Braga MR. Leaf metabolite profile of the Brazilian resurrection plant Barbacenia purpurea Hook. (Velloziaceae) shows two time-dependent responses during desiccation and recovering. Front Plant Sci 2014; 5:1-13; http://dx.doi.org/10.3389/fpls.2014.00096
21. Vizin RC, Scarpellini CD, Ishikawa DT, Correa GM, de Souza CO, Gargaglioni LH, Carrettiero DC, Bicego KC, Almeida MC. TRPV4 activates autonomic and behavioural warmth-defence responses in Wistar rats. Acta Physiol (Oxf) 2015; 214:275-89; PMID:25739906; http://dx.doi.org/10.1111/apha.12477
22. Malvin GM, Wood SC. Behavioral hypothermia and survival of hypoxic protozoans Paramecium caudatum. Science 1992; 255:1423-5; PMID:1542790; http://dx.doi.org/10.1126/science.1542790
23. Roberts WW, Martin JR. Effects of lesions in central thermosensitive areas on thermoregulatory responses in rat. Physiol Behav 1977; 19:503-11; PMID:613343; http://dx.doi.org/10.1016/0031-9384(77)90226-8
24. Carlisle HJ. Behavioural significance of hypothalamic temperature-sensitive cells. Nature 1966; 209:1324-5; PMID:5956045; http://dx.doi.org/10.1038/2091324a0
25. Satinoff E. Behavioral Thermoregulation in Response to Local Cooling of the Rat Brain. Am J Physiol 1964; 206:1389-94; PMID:14193560
26. Maruyama M, Nishi M, Konishi M, Takashige Y, Nagashima K, Kiyohara T, Kanosue K. Brain regions expressing Fos during thermoregulatory behavior in rats. Am J Physiol Regul Integr Comp Physiol 2003; 285:R1116-23; PMID:12893652; http://dx.doi.org/10.1152/ajpregu.00166.2002
27. Carlisle HJ. Effect of preoptic and anterior hypothalamic lesions on behavioral thermoregulation in the cold. J Comp Physiol Psychol 1969; 69:391-402; PMID:5404472; http://dx.doi.org/10.1037/h0028170
28. Lipton J. Effects of preoptic lesions on heat-escape responding and colonic temperature in the rat. Physiol Behavior 1968; 3:165-9; http://dx.doi.org/10.1016/0031-9384(68)90049-8
29. Satinoff E, Rutstein J. Behavioral thermoregulation in rats with anterior hypothalamic lesions. J Comp Physiol Psychol 1970; 71:77-82; PMID:5452104; http://dx.doi.org/10.1037/h0028959
30. Konishi M, Kanosue K, Kano M, Kobayashi A, Nagashima K. The median preoptic nucleus is involved in the facilitation of heat-escape/cold-seeking behavior during systemic salt loading in rats. Am J Physiol Regul Integr Comp Physiol 2007; 292:R150-9; PMID:17197642; http://dx.doi.org/10.1152/ajpregu.00769.2005
31. Satinoff E, Shan SY. Loss of behavioral thermoregulation after lateral hypothalamic lesions in rats. J Comp Physiol Psychol 1971; 77:302-12; PMID:5117208; http://dx.doi.org/10.1037/h0031652
32. Refinetti R, Carlisle HJ. A reevaluation of the role of the lateral hypothalamus in behavioral temperature regulation. Physiol Behav 1987; 40:189-92; PMID:3628527; http://dx.doi.org/10.1016/0031-9384(87)90206-X
33. Refinetti R, Carlisle HJ. Effects of lateral hypothalamic lesions on thermoregulation in the rat. Physiol Behav 1986; 38:219-28; PMID:3797489; http://dx.doi.org/10.1016/0031-9384(86)90157-5
34. Roberts WW, Martin JR. Peripheral thermoreceptor control of thermoregulatory responses of the rat. J Comp Physiol Psychol 1974; 87:1109-18; PMID:4443452; http://dx.doi.org/10.1037/h0037584
35. Whyte DG, Brennan TJ, Johnson AK. Thermoregulatory behavior is disrupted in rats with lesions of the anteroventral third ventricular area (AV3V). Physiol Behavior 2006; 87:493-9; PMID:16487986; http://dx.doi.org/10.1016/j.physbeh.2005.11.019
36. Bolles RC. Grooming behavior in the rat. J Comparative Physiol Psychol 1960; 53:306-10; PMID:13802322; http://dx.doi.org/10.1037/h0045421
37. Kalueff AV, Tuohimaa P. Grooming analysis algorithm for neurobehavioural stress research. Brain Res Brain Res Protoc 2004; 13:151-8; PMID:15296852; http://dx.doi.org/10.1016/j.brainresprot.2004.04.002
38. Tanaka H, Kanosue K, Nakayama T, Shen Z. Grooming, body extension, and vasomotor responses induced by hypothalamic warming at different ambient temperatures in rats. Physiol Behavior 1986; 38:145-51; PMID:3786495; http://dx.doi.org/10.1016/0031-9384(86)90145-9
39. Hainsworth FR, Stricker EM. Evaporative cooling in the rat: effects of partial desalivation. Am J Physiol 1969; 217:494-7; PMID:5799377
40. Hainsworth FR. Evaporative water loss from rats in the heat. Am J Physiol 1968; 214:979-82; PMID:5647201
41. Roberts WW, Mooney RD, Martin JR. Thermoregulatory behaviors of laboratory rodents. J Comp Physiol Psychol 1974; 86:693-9; PMID:4823242; http://dx.doi.org/10.1037/h0036138
42. Hainsworth FR, Epstein AN. Severe Impairment of Heat-Induced Saliva-Spreading in Rats Recovered from Lateral Hypothalamic Lesions. Science 1966; 153:1255-7; PMID:5920755; http://dx.doi.org/10.1126/science.153.3741.1255
43. Stricker EM, Hainsworth FR. Evaporative cooling in the rat: effects of hypothalamic lesions and chorda tympani damage. Can J Physiol Pharmacol 1970; 48:11-7; PMID:5414311; http://dx.doi.org/10.1139/y70-002
44. Flynn FW, Evey LA, Mitchell JC. Heat-induced saliva secretion and thermoregulation in female rats with ventromedial hypothalamic lesions. Physiol Behav 1981; 26:779-82; PMID:7267771; http://dx.doi.org/10.1016/0031-9384(81)90098-6
45. Whyte DG, Johnson AK. Lesions of periventricular tissue surrounding the anteroventral third ventricle (AV3V) attenuate salivation and thermal tolerance in response to a heat stress. Brain Res 2002; 951:146-9; PMID:12231469; http://dx.doi.org/10.1016/S0006-8993(02)03238-9
46. Whyte DG, Johnson AK. Lesions of the anteroventral third ventricle region (AV3V) disrupt cardiovascular responses to an elevation in core temperature. Am J Physiol Regul Integr Comp Physiol 2005; 288:R1783-90; PMID:15650127; http://dx.doi.org/10.1152/ajpregu.00524.2004
47. Hubschle T, McKinley MJ, Oldfield BJ. Efferent connections of the lamina terminalis, the preoptic area and the insular cortex to submandibular and sublingual gland of the rat traced with pseudorabies virus. Brain Res 1998; 806:219-31; PMID:9739143; http://dx.doi.org/10.1016/S0006-8993(98)00765-3
48. Hubschle T, Mathai ML, McKinley MJ, Oldfield BJ. Multisynaptic neuronal pathways from the submandibular and sublingual glands to the lamina terminalis in the rat: a model for the role of the lamina terminalis in the control of osmo- and thermoregulatory behavior. Clin Exp Pharmacol Physiol 2001; 28:558-69; PMID:11458883; http://dx.doi.org/10.1046/j.1440-1681.2001.03487.x
49. Yanase M, Kanosue K, Yasuda H, Tanaka H. Salivary secretion and grooming behaviour during heat exposure in freely moving rats. J Physiol 1991; 432:585-92; PMID:1886070; http://dx.doi.org/10.1113/jphysiol.1991.sp018402
50. Kanosue K, Nakayama T, Tanaka H, Yanase V, Yasuda H. Modes of action of local hypothalamic and skin thermal stimulation on salivary secretion in rats. J Physiol 1990; 424:459-71; PMID:2391658; http://dx.doi.org/10.1113/jphysiol.1990.sp018077
51. Caterina MJ. Transient receptor potential ion channels as participants in thermosensation and thermoregulation. Am J Physiol Regul Integr Comp Physiol 2007; 292:R64-76; PMID:16973931; http://dx.doi.org/10.1152/ajpregu.00446.2006
52. Dhaka A, Murray AN, Mathur J, Earley TJ, Petrus MJ, Patapoutian A. TRPM8 Is Required for Cold Sensation in Mice. Neuron 2007; 54:371-8; PMID:17481391; http://dx.doi.org/10.1016/j.neuron.2007.02.024
53. Patapoutian A, Peier AM, Story GM, Viswanath V. ThermoTRP channels and beyond: mechanisms of temperature sensation. Nat Rev Neurosci 2003; 4:529-39; PMID:12838328; http://dx.doi.org/10.1038/nrn1141

54. Vay L, Gu C, McNaughton PA. The thermo-TRP ion channel family: properties and therapeutic implications. Br J Pharmacol 2012; 165:787-801; PMID:21797839; http://dx.doi.org/10.1111/j.1476-5381.2011.01601.x
55. Laursen W, Anderson E, Hoffstaetter L, Bagriantsev S, Gracheva E. Species-specific temperature sensitivity of TRPA1. Temperature 2015; 2:214-26; http://dx.doi.org/10.1080/23328940.2014.1000702
56. Moqrich A, Hwang SW, Earley TJ, Petrus MJ, Murray AN, Spencer KS, Andahazy M, Story GM, Patapoutian A. Impaired thermosensation in mice lacking TRPV3, a heat and camphor sensor in the skin. Science 2005; 307:1468-72; PMID:15746429; http://dx.doi.org/10.1126/science.1108609
57. Lee H, Iida T, Mizuno A, Suzuki M, Caterina MJ. Altered thermal selection behavior in mice lacking transient receptor potential vanilloid 4. J Neurosci 2005; 25:1304-10; PMID:15689568; http://dx.doi.org/10.1523/JNEUROSCI.4745.04.2005
58. Bautista DM, Siemens J, Glazer JM, Tsuruda PR, Basbaum AI, Stucky CL, Jordt SE, Julius D. The menthol receptor TRPM8 is the principal detector of environmental cold. Nature 2007; 448:204-8; PMID:17538622; http://dx.doi.org/10.1038/nature05910
59. Colburn RW, Lubin ML, Stone DJ, Jr., Wang Y, Lawrence D, D'Andrea MR, Brandt MR, Liu Y, Flores CM, Qin N. Attenuated cold sensitivity in TRPM8 null mice. Neuron 2007; 54:379-86; PMID:17481392; http://dx.doi.org/10.1016/j.neuron.2007.04.017
60. McKemy DD, Neuhausser WM, Julius D. Identification of a cold receptor reveals a general role for TRP channels in thermosensation. Nature 2002; 416:52-8; PMID:11882888; http://dx.doi.org/10.1038/nature719
61. Nealen ML, Gold MS, Thut PD, Caterina MJ. TRPM8 mRNA is expressed in a subset of cold-responsive trigeminal neurons from rat. J Neurophysiol 2003; 90:515-20; PMID:12634279; http://dx.doi.org/10.1152/jn.00843.2002
62. Peier AM, Moqrich A, Hergarden AC, Reeve AJ, Andersson DA, Story GM, Earley TJ, Dragoni I, McIntyre P, Bevan S, et al. A TRP channel that senses cold stimuli and menthol. Cell 2002; 108:705-15; PMID:11893340; http://dx.doi.org/10.1016/S0092-8674(02)00652-9
63. Thut PD, Wrigley D, Gold MS. Cold transduction in rat trigeminal ganglia neurons in vitro. Neuroscience 2003; 119:1071-83; PMID:12831865; http://dx.doi.org/10.1016/S0306-4522(03)00225-2
64. Knowlton WM, Bifolck-Fisher A, Bautista DM, McKemy DD. TRPM8, but not TRPA1, is required for neural and behavioral responses to acute noxious cold temperatures and cold-mimetics in vivo. Pain 2010; 150:340-50; PMID:20542379; http://dx.doi.org/10.1016/j.pain.2010.05.021
65. Knowlton WM, Palkar R, Lippoldt EK, McCoy DD, Baluch F, Chen J, McKemy DD. A sensory-labeled line for cold: TRPM8-expressing sensory neurons define the cellular basis for cold, cold pain, and cooling-mediated analgesia. J Neurosci 2013; 33:2837-48; PMID:23407943; http://dx.doi.org/10.1523/JNEUROSCI.1943-12.2013
66. Feketa VV, Balasubramanian A, Flores CM, Player MR, Marrelli SP. Shivering and tachycardic responses to external cooling in mice are substantially suppressed by TRPV1 activation but not by TRPM8 inhibition. Am J Physiol Regul Integr Comp Physiol; 305:R1040-50; PMID:24005250; http://dx.doi.org/10.1152/ajpregu.00296.2013
67. Almeida MC, Hew-Butler T, Soriano RN, Rao S, Wang W, Wang J, Tamayo N, Oliveira DL, Nucci TB, Aryal P, et al. Pharmacological blockade of the cold receptor TRPM8 attenuates autonomic and behavioral cold defenses and decreases deep body temperature. J Neurosci 2012; 32:2086-99; PMID:22323721; http://dx.doi.org/10.1523/JNEUROSCI.5606-11.2012
68. Tajino K, Matsumura K, Kosada K, Shibakusa T, Inoue K, Fushiki T, Hosokawa H, Kobayashi S. Application of menthol to the skin of whole trunk in mice induces autonomic and behavioral heat-gain responses. Am J Physiol Regul Integr Comp Physiol 2007; 293(5):R2128-35:00377.2007
69. Cechetto DF, Standaert DG, Saper CB. Spinal and trigeminal dorsal horn projections to the parabrachial nucleus in the rat. J Comp Neurol 1985; 240:153-60; PMID:3840498; http://dx.doi.org/10.1002/cne.902400205
70. Feil K, Herbert H. Topographic organization of spinal and trigeminal somatosensory pathways to the rat parabrachial and Kolliker-Fuse nuclei. J Comp Neurol 1995; 353:506-28; PMID:7759613; http://dx.doi.org/10.1002/cne.903530404
71. Bratincsak A, Palkovits M. Activation of brain areas in rat following warm and cold ambient exposure. Neuroscience 2004; 127:385-97; PMID:15262329; http://dx.doi.org/10.1016/j.neuroscience.2004.05.016
72. Caterina MJ, Schumacher MA, Tominaga M, Rosen TA, Levine JD, Julius D. The capsaicin receptor: a heat-activated ion channel in the pain pathway. Nature 1997; 389:816-24; PMID:9349813; http://dx.doi.org/10.1038/39807
73. Tominaga M, Caterina MJ, Malmberg AB, Rosen TA, Gilbert H, Skinner K, Raumann BE, Basbaum AI, Julius D. The cloned capsaicin receptor integrates multiple pain-producing stimuli. Neuron 1998; 21:531-43; PMID:9768840; http://dx.doi.org/10.1016/S0896-6273(00)80564-4
74. Garami A, Shimansky YP, Pakai E, Oliveira DL, Gavva NR, Romanovsky AA. Contributions of different modes of TRPV1 activation to TRPV1 antagonist-induced hyperthermia. J Neurosci 2010; 30:1435-40; PMID:20107070; http://dx.doi.org/10.1523/JNEUROSCI.5150-09.2010
75. Romanovsky AA, Almeida MC, Garami A, Steiner AA, Norman MH, Morrison SF, Nakamura K, Burmeister JJ, Nucci TB. The transient receptor potential vanilloid-1 channel in thermoregulation: a thermosensor it is not. Pharmacol Rev 2009; 61:228-61; PMID:19749171; http://dx.doi.org/10.1124/pr.109.001263
76. Steiner AA, Turek VF, Almeida MC, Burmeister JJ, Oliveira DL, Roberts JL, Bannon AW, Norman MH, Louis JC, Treanor JJ, et al. Nonthermal activation of transient receptor potential vanilloid-1 channels in abdominal viscera tonically inhibits autonomic cold-defense effectors. J Neurosci 2007; 27:7459-68; PMID:17626206; http://dx.doi.org/10.1523/JNEUROSCI.1483-07.2007
77. Guler AD, Lee H, Iida T, Shimizu I, Tominaga M, Caterina M. Heat-evoked activation of the ion channel, TRPV4. J Neurosci 2002; 22:6408-14; PMID:12151520
78. Peier AM, Reeve AJ, Andersson DA, Moqrich A, Earley TJ, Hergarden AC, Story GM, Colley S, Hogenesch JB, McIntyre P, et al. A heat-sensitive TRP channel expressed in keratinocytes. Science 2002; 296:2046-9; PMID:12016205; http://dx.doi.org/10.1126/science.1073140
79. Smith GD, Gunthorpe MJ, Kelsell RE, Hayes PD, Reilly P, Facer P, Wright JE, Jerman JC, Walhin JP, Ooi L, et al. TRPV3 is a temperature-sensitive vanilloid receptor-like protein. Nature 2002; 418:186-90; PMID:12077606; http://dx.doi.org/10.1038/nature00894
80. Watanabe H, Vriens J, Suh SH, Benham CD, Droogmans G, Nilius B. Heat-evoked activation of TRPV4 channels in a HEK293 cell expression system and in native mouse aorta endothelial cells. J Biol Chem 2002; 277:47044-51; PMID:12354759; http://dx.doi.org/10.1074/jbc.M208277200
81. Xu H, Ramsey IS, Kotecha SA, Moran MM, Chong JA, Lawson D, Ge P, Lilly J, Silos-Santiago I, Xie Y, et al. TRPV3 is a calcium-permeable temperature-sensitive cation channel. Nature 2002; 418:181-6; PMID:12077604; http://dx.doi.org/10.1038/nature00882
82. Xu H, Delling M, Jun JC, Clapham DE. Oregano, thyme and clove-derived flavors and skin sensitizers activate specific TRP channels. Nat Neurosci 2006; 9:628-35; PMID:16617338; http://dx.doi.org/10.1038/nn1692
83. Gao X, Wu L, O'Neil RG. Temperature-modulated diversity of TRPV4 channel gating: activation by physical stresses and phorbol ester derivatives through protein kinase C-dependent and -independent pathways. J Biol Chem 2003; 278:27129-37; PMID:12738791; http://dx.doi.org/10.1074/jbc.M302517200
84. Kohler R, Heyken WT, Heinau P, Schubert R, Si H, Kacik M, Busch C, Grgic I, Maier T, Hoyer J. Evidence for a functional role of endothelial transient receptor potential V4 in shear stress-induced vasodilatation. Arterioscler Thromb Vasc Biol 2006; 26:1495-502; PMID:16675722; http://dx.doi.org/10.1161/01.ATV.0000225698.36212.6a
85. Liedtke W. TRPV4 plays an evolutionary conserved role in the transduction of osmotic and mechanical stimuli in live animals. J Physiol 2005; 567:53-8; PMID:15961428; http://dx.doi.org/10.1113/jphysiol.2005.088963
86. O'Neil RG, Heller S. The mechanosensitive nature of TRPV channels. Pflugers Arch 2005; 451:193-203; PMID:15909178; http://dx.doi.org/10.1007/s00424-005-1424-4
87. Strotmann R, Harteneck C, Nunnenmacher K, Schultz G, Plant TD. OTRPC4, a nonselective cation channel that confers sensitivity to extracellular osmolarity. Nat Cell Biol 2000; 2:695-702; PMID:11025659; http://dx.doi.org/10.1038/35036318
88. Suzuki M, Mizuno A, Kodaira K, Imai M. Impaired pressure sensation in mice lacking TRPV4. J Biol Chem 2003; 278:22664-8; PMID:12692122; http://dx.doi.org/10.1074/jbc.M302561200
89. Voets T, Talavera K, Owsianik G, Nilius B. Sensing with TRP channels. Nat Chem Biol 2005; 1:85-92; PMID:16408004; http://dx.doi.org/10.1038/nchembio0705-85
90. Alessandri-Haber N, Yeh JJ, Boyd AE, Parada CA, Chen X, Reichling DB, Levine JD. Hypotonicity induces TRPV4-mediated nociception in rat. Neuron 2003; 39:497-511; PMID:12895423; http://dx.doi.org/10.1016/S0896-6273(03)00462-8
91. Liedtke W, Choe Y, Marti-Renom MA, Bell AM, Denis CS, Sali A, Hudspeth AJ, Friedman JM, Heller S. Vanilloid receptor-related osmotically activated channel (VR-OAC), a candidate vertebrate osmoreceptor. Cell 2000; 103:525-35; PMID:11081638; http://dx.doi.org/10.1016/S0092-8674(00)00143-4
92. Chung MK, Lee H, Mizuno A, Suzuki M, Caterina MJ. TRPV3 and TRPV4 mediate warmth-evoked currents in primary mouse keratinocytes. J Biol Chem 2004; 279:21569-75; PMID:15004014; http://dx.doi.org/10.1074/jbc.M401872200
93. Mandadi S, Sokabe T, Shibasaki K, Katanosaka K, Mizuno A, Moqrich A, Patapoutian A, Fukumi-Tominaga T, Mizumura K, Tominaga M. TRPV3 in keratinocytes transmits temperature information to sensory neurons via ATP. PfÃ¼gers Archiv - European J Physiol 2009; 458:1093-102; PMID:19669158; http://dx.doi.org/10.1007/s00424-009-0703-x
94. Marics I, Malapert P, Reynders A, Gaillard S, Moqrich A. Acute heat-evoked temperature sensation is impaired but not abolished in mice lacking TRPV1 and TRPV3 channels. PLoS One 2014; 9:e99828; PMID:24925072; http://dx.doi.org/10.1371/journal.pone.0099828
95. Huang SM, Li X, Yu Y, Wang J, Caterina MJ. TRPV3 and TRPV4 ion channels are not major contributors to mouse heat sensation. Mol Pain 2011; 7:37; PMID:21586160; http://dx.doi.org/10.1186/1744-8069-7-37
96. Lariviere WR, Chesler EJ, Mogil JS. Transgenic studies of pain and analgesia: mutation or background genotype? J Pharmacol Exp Ther 2001; 297:467-73; PMID:11303031
97. Mogil JS, Yu L, Basbaum AI. Pain genes?: natural variation and transgenic mutants. Annu Rev Neurosci 2000; 23:777-811; PMID:10845081; http://dx.doi.org/10.1146/annurev.neuro.23.1.777