Central Pathways Controlling Brown Adipose Tissue Thermogenesis

Shaun F. Morrison

Neurological Sciences Institute, Oregon Health and Science University, Beaverton, Oregon 97006

Heat production in brown adipose tissue contributes to cold defense, to stress-induced increases in body temperature, and to energy balance. Elucidating the functional organization of the central network controlling the sympathetic outflow to brown adipose tissue could provide a framework for understanding how dysregulation of thermogenesis contributes to hyperthermia and to obesity.

In small mammals, the defense of body temperature in a cold environment presents a challenge to homeostasis that is met through sympathetically regulated, nonshivering (or adaptive) thermogenesis, coupled with a reduction in cutaneous blood flow. Brown adipose tissue (BAT), the principal effector of thermogenesis, is composed of adipocytes replete with mitochondria that contain uncoupling protein-1, which shunts the energy obtained from the oxidation of free fatty acids into heat, which is then convected to BAT vasculature and distributed throughout the body. The consumption of fatty acids during diet-induced thermogenesis in BAT has also received recent attention as a component in the energy balance equation that influences the level of stored calories (adiposity). The release of norepinephrine from sympathetic ganglion cell terminals innervating β3-adrenergic receptors on brown adipocytes regulates the level of heat production in BAT by 1) augmenting the activity of the lipase that determines the amount of cytoplasmic free fatty acids available for mitochondrial oxidation and 2) increasing the content of uncoupling protein-1 in the mitochondria by increasing transcription (14).

This review summarizes our current understanding of the neural circuits within the central nervous system that control BAT heat production through regulation of the level of BAT sympathetic nerve activity. The goal of developing an informative schematic model of the functional organization of the neuronal populations regulating BAT thermogenesis is approached retrogradely, beginning with the peripheral components and moving centrally through the brain stem to cell groups in the hypothalamus. The focus is on cell groups and pathways mediating the BAT thermogenic component of the thermoregulatory response to cold. Additional information is introduced on the neural circuits controlling nonshivering thermogenesis as it relates to energy balance where central regulation of heat production in BAT serves as a model for the control of calorie consumption that is directed toward reducing the adipose energy reserve. The marked differences in the signals from afferents, reflexes, and hormones that are expected to influence the sympathetic outflow to BAT, the effector in each of these two distinct homeostatic processes, make the elucidation of the central circuits controlling BAT sympathetic nerve activity both challenging and fascinating.

Innervation of BAT

Although deposits of BAT are distributed throughout the body, the largest depot in the rat and mouse are the lobules of the interscapular BAT. Application of retrograde tracer (11) or pseudorabies virus (15) into the rat interscapular BAT indicates that the majority of sympathetic ganglion cells innervating the interscapular BAT are located in the middle cervical (stellate) ganglion, and these receive excitatory, cholinergic inputs from sympathetic preganglionic neurons contained primarily within the third and fourth thoracic segments (Fig. 1). The most potent determinant of the discharge of BAT sympathetic preganglionic neurons is expected to be the activity of their direct inputs from supraspinal, BAT sympathetic premotor neurons.

The most powerful technique permitting localization of the sympathetic premotor neurons controlling the sympathetic outflow to BAT is transsynaptic retrograde tracing using pseudorabies virus injected into the interscapular BAT. In this application of this technique, the supraspinal cell groups with the earliest appearance of an infection after viral injection into the interscapular BAT pads are in the anatomic position of sympathetic premotor neurons for BAT because they would have become infected through their synaptic contact with BAT sympathetic preganglionic neurons in the upper thoracic spinal cord. Following injections of the pseudorabies virus into interscapular BAT fat pads, the earliest retrogradely infected neurons were found in the raphe pallidus/magnus area, in the parapyramidal region ventral to the inferior olivary complex, in the region of the rostral ventrolateral medulla, in the A5 noradrenergic cell group, and in the paraventricular hypothalamus (1, 5). Although these anatomic results strongly imply roles for neurons in each of these regions in the premotor regulation of BAT sympathetic nerve activity, only the raphe pallidus/magnus area has been examined physiologically.

Role of raphe pallidus neurons in BAT thermogenesis

A representative population of BAT sympathetic premotor neurons in the raphe pallidus is shown in Fig. 1. One of the criteria for BAT sympathetic premotor neurons is that their activation should lead to increased BAT sympathetic nerve activity both challenging and fascinating.
bicuculline leads to a prompt and marked increase in BAT activity and BAT thermogenesis. The potential mechanisms through which rostral raphe pallidus neurons are excited following local GABA\(_A\) receptor blockade may be similar to those described in the next section on the effect of brain stem transections. Their elucidation remains critical to our understanding of the fundamental brain stem network regulating BAT thermogenesis and the accompanying stimulation of heart rate. Thus, although the existence of BAT sympathetic premotor neurons in the raphe pallidus/magnus region is supported by the findings that raphe pallidus neurons project to BAT sympathetic preganglionic neurons in the thoracic spinal cord and that activation of neurons in this area increases BAT thermogenesis, additional data are needed to demonstrate, for instance, that BAT thermogenic responses are eliminated by inhibition of raphe pallidus neurons and that at least some of the raphe pallidus neurons that project to the thoracic spinal cord exhibit the appropriate characteristics, conduction velocity and discharge patterns to be classified as sympathetic premotor neurons for BAT. Of related interest are the recent findings supporting the existence in the same medullary raphe region of sympathetic premotor neurons regulating the sympathetic outflow to skin blood vessels in the tail (16) and the rabbit ear (3). Cutaneous blood flow in these vessels determines the amount of heat lost to the environment and their sympathetically regulated vasoconstriction parallels the sympathetic drive to BAT in thermoregulatory responses. The similar localization of neurons regulating cutaneous blood flow and BAT thermogenesis suggests an organization of medullary sympathetic premotor neurons in which those of the raphe pallidus control thermoregulatory and metabolic effectors, whereas those in the rostral ventrolateral medulla regulate cardiovascular targets to maintain tissue perfusion and arterial pressure (9).

Brain stem involvement in BAT thermogenesis

In light of this new information on the location of BAT sympathetic premotor neurons in the medulla oblongata, the results of transection experiments have important implications for understanding the functional organization of the brain stem components of the central sympathetic network regulating BAT thermogenesis. We first consider the effects on BAT temperature in anesthetized rats that follow transections through the neuraxis at two separate locations in the brain stem. As shown in Fig. 3A (cut A), a transverse transection of the pons and medulla (schematized as Fig. 1, bar A (17) or infusion of procaine into the pons (18) produces a large increase in BAT thermogenesis and in core temperature. A similar transection in the area between the rostral pons and the mamillary bodies (Fig. 3A, inset, cut B; schematized as Fig. 1, bar B) has no effect on BAT temperature (17). These data imply the existence of a tonically active, descending pathway that traverses the region of the caudal transection (Fig. 3A, cut A; Fig. 1, bar A) and produces an inhibition of the activity of BAT sympathetic premotor neurons, possibly those in raphe...
pallidus, located caudal to this transection. This inhibitory pathway is schematized in Fig. 1. In this regard, the large BAT thermogenic response to administration of bicuculline into raphe pallidus (Fig. 2) (12) indicates the existence of tonically active, GABA_A receptor-mediated, inhibitory inputs to the neurons in this region controlling BAT thermogenesis. Coupled with the fact that the level of BAT sympathetic nerve activity is quite low during normothermia in the anesthetized rat, these findings suggest that disinhibition of BAT sympathetic premotor neurons in the rostral raphe pallidus is a key regulator of the level of BAT sympathetic nerve activity and BAT thermogenesis. Three additional implications may be derived from the thermogenic effects of these transections. First, because eliminating the descending inhibitory input with the transection (Fig. 1, bar A) results in stimulation of BAT thermogenesis, the neurons comprising this inhibitory input should be tonically active in the anesthetized rat (note the hypothetical excitatory input to the pontine inhibitory neurons and the medial preoptic area in Fig. 1). Second, at least some of these inhibitory neurons are located within the pons, since the transection rostral to the pons (Fig. 1, bar B) did not produce a similar increase in BAT thermogenesis. Third, within the medulla (i.e., below the level of transection A in Fig. 1) one would hypothesize the existence of a tonically active source of excitation (Fig. 1, pathway 1) to the BAT sympathetic premotor neurons, or they may possess inherent membrane properties for sustained activation following removal of the GABAergic inhibition. This mechanism would be necessary to provide the increase in their discharge that underlies the stimulation of BAT thermogenesis seen following transection caudal to the pons.

Neurons in the inferior olive (19) have been suggested to mediate the thermogenic response to elimination of the pontine inhibitory input. However, this conclusion remains in doubt for several reasons. First, no retrogradely infected cells have ever been reported in the inferior olive after pseudorabies virus injections into BAT (1, 5), even after very long survival times. These data indicate that inferior olivary cells are not within central pathways that influence the sympathetic nerve outflow to BAT. Indeed, inferior olivary cells are considered to be a homogeneous cell population providing the climbing fiber excitation exclusively to cerebellar Purkinje cells. Second, the electrical stimulation and the procaine and electrolytic lesion techniques used in these studies do not discriminate between effects on cell bodies and on axons of passage. Third, the parameters used in these experiments, such as the large concentration and volume of the intraparenchymal glutamate infusion, were beyond those known to have regionally restricted effects, introducing the possibility that the results could be attributed to neurons outside the inferior olive.

Pontine neurons that may contribute to the descending inhibitory pathway are schematized in Fig. 1. (a) In this regard, the large BAT thermogenic response to administration of bicuculline into raphe pallidus (Fig. 2) (12) indicates the existence of tonically active, GABA_A receptor-mediated, inhibitory inputs to the neurons in this region controlling BAT thermogenesis. These are accompanied by a small rise in arterial pressure (AP) and a marked increase in heart rate (HR). The elevation in expired CO_2 (Exp CO_2) is likely produced by the increased metabolism in BAT and in heart, the latter due to the evoked tachycardia. Horizontal calibration bar represents 2 min, vertical bar is 100 μV for IBAT sympathetic nerve activity. Inset: histological section containing a dye spot (arrow) marking the bicuculline microinjection site (left), and a modified atlas drawing through the rostral raphe pallidus indicating the position of the microinjection site (circle) (right) are shown. PrH, prepositus hypoglossus; Sol, nucleus solitarius; RMg, raphe magnus; LPGi, lateral paragigantocellularis; 7, facial motor nucleus; 4V, fourth ventricle. Data represent similar responses to those in Fig. 1 of Ref. 12.
bition of BAT thermogenesis have been localized to a large region in the vicinity of the retrorubral field (19), although these experiments involved the infusion of large volumes of procaine, which is not selective for inhibition of cell bodies. The only other midbrain region that has been examined for effects on BAT thermogenesis is the caudal periaqueductal gray, in which activation of neurons elicits a rise in BAT temperature. The specific role of these cells in the circuits controlling BAT thermogenic responses remains to be determined.

Control of BAT thermogenesis in thermoregulation and fever

A reduction in skin or core temperature, usually resulting from a fall in the temperature of the animal’s environment, is the primary thermoregulatory stimulus for the activation of BAT thermogenesis. Although rudimentary thermal responses to cold can be evoked from most levels of the neuraxis, the most complete responses involve the medial preoptic area of the anterior hypothalamus, which contains neurons with characteristic thermal sensitivities. Populations of warm-sensitive neurons that are activated by an increase in preoptic area temperature and cold-sensitive neurons, stimulated by local cooling, integrate the effects of local temperature on their membrane excitability with inputs from 1) thermally insensitive, tone-setting neurons in the medial preoptic area and 2) information from spinal afferent pathways conveying thermal sensation from cutaneous and other tissue receptors. Medial preoptic area neurons with different thermal sensitivities are differentially involved in mediating thermogenic vs. heat loss responses (6). Through descending pathways eventually influencing the discharge of sympathetic premotor neurons, medial preoptic area efferent neurons can produce a continuum of responses. In a hot environment, BAT sympathetic nerve activity is inhibited, BAT lipid metabolism and BAT thermogenesis are reduced, and cutaneous vasoconstriction is relaxed. In contrast, cold stimuli evoke simultaneous increases in sympathetic outflow to BAT to stimulate nonshivering thermogenesis and to the cutaneous vasculature to reduce heat loss.

Clues to the understanding of the hypothalamic networks responsible for the activation of BAT sympathetic nerve activity and thermogenesis in the normal thermoregulatory response to cold may be derived from studies on the neural pathways mediating the metabolic heat production during the febrile component of the acute phase reaction to infection or immune challenge. Peripheral pyrogenic signals initiate changes in central neuronal activity that result in stimulation of BAT thermogenesis and an increase in body temperature. Although the underlying mechanisms continue to be debated, considerable evidence indicates a role for the interaction of locally released prostaglandin E₂ (PGE₂) with prostaglandin receptors on neurons in the region of the medial preoptic area, the median preoptic nucleus, or the organum vasculosum of the laminae terminalis. Additional pathways may include blood-borne immune signaling molecules entering the brain via a circumventricular organ such as the organum vasculosum of the laminae terminalis or immune signals initiating fever as a reflex response through their effects on vagal sen-
sory terminals. Since intracerebroventricular injection of PGE$_2$ or direct microinjection of PGE$_2$ into the medial preoptic area elicits large increases in BAT sympathetic outflow and BAT thermogenesis, they provide a useful stimulus to identify a thermogenic pathway from the medial preoptic area that may also function in normal thermoregulation.

As with thermogenic responses to a cold environment, the activation of BAT sympathetic outflow and BAT thermogenesis evoked by central application of PGE$_2$ is prevented or reversed by inactivation of neurons in the raphe pallidus (10, 13). Thus, although other populations of BAT sympathetic premotor neurons may also be activated during the febrile response, those in the rostral raphe pallidus are an essential component of the thermogenic efferent pathway from the medial preoptic area. The large increases in BAT and core temperatures (Fig. 3B) produced by transections restricted to the fiber tracts immediately caudal to the medial preoptic area (Fig. 1, bar C), or by inhibition of medial preoptic area neurons with tetrodotoxin, indicate that the efferent pathway from the medial preoptic area (Fig. 1, pathway 4) mediates a tonic inhibition of BAT thermogenesis in normothermic, anesthetized rats (6). Because warm-sensitive neurons would be expected to inhibit thermogenesis, this result suggests that the influence of the medial preoptic area on the level of BAT thermogenesis is determined predominantly by the level of inhibition that warm-sensitive neurons exert on the activity of the sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat production stimulated by exposure to cold or to agents producing sympathetic nerves to BAT. In particular, the increased heat produc...
other. The former would be the case if the descending excitatory pathway (hypothesized in the dorsomedial hypothalamus) was the target of the tonic inhibitory output from the medial preoptic area (Fig. 1, pathway 4).

Beyond the preoptic area, regions in the hypothalamus that contain retrogradely labeled neurons following pseudorabies virus injections into BAT and thus are within pathways connected to BAT sympathetic preganglionic neurons include the dorsomedial hypothalamic nucleus, the paraventricular nucleus, the arcuate nucleus, and the lateral hypothalamus (1, 5, 15). Anatomically, the small size, close proximity, and interconnections of these areas, coupled with the fact that efferent axons from one region traverse neighboring cell groups in their descending path, have complicated attempts to dissect the functional roles and interrelationships of hypothalamic neurons controlling BAT thermogenesis. Physiologically, because the hypothalamus receives many afferent signals, including neuronal, hormonal, and thermal, and because it functions to modulate and coordinate activity in many tissues, it is difficult to "assign" a selective function to neurons in a particular region. In many cases, the experimental techniques employed, in particular electrical stimulation, electrolytic lesion, and microinjection of local anesthetics, also confound the interpretation of the results because they fail to differentiate between effects on local neurons and axons of passage. For instance, nearly all of the studies indicating a role for the ventromedial hypothalamus in mediating BAT thermogenic responses involve such techniques, and thus the results could be attributable solely to axons passing through this area. This concern is supported by the absence of retrogradely infected cells in the ventromedial hypothalamus following application of pseudorabies virus into BAT.

Regarding the paraventricular, lateral, and arcuate nuclei, our understanding of their role in the control of BAT thermogenesis is limited (1) to the finding that cells in these regions are infected after injection of pseudorabies virus into BAT and (2) to determinations that microinjections of excitatory amino acids or peptides can elicit changes in BAT uncoupling protein or temperature. Although information on the functional relationship of each of these regions to the control of BAT thermogenesis is sparse, the dorsomedial hypothalamic nucleus is a potential synaptic integration site in the indirect pathway through which medial preoptic area neurons could influence BAT thermogenesis.

**Role of dorsomedial hypothalamic neurons in thermogenic responses**

"Stressful" stimuli such as restraint, noxious pinching, anxiety paradigms, and placement in a novel cage evoke a...
stellation of autonomic responses in preparation for potential danger, escape, conflict, or injury. Similar to the febrile response, these include an increase in BAT thermogenesis supporting a sustained elevation in body temperature (“stress hyperthermia”). Neurons in the dorsomedial hypothalamus participate in the pathways mediating the autonomic responses to certain stressful stimuli. As illustrated in Fig. 4, disinhibition of dorsomedial hypothalamic neurons with the GABAA receptor antagonist bicuculline increases BAT thermogenesis, body temperature, and other autonomic variables in a similar way to that seen during stress (20). The strong activation of BAT thermogenesis following blockade of GABAA receptors in the dorsomedial hypothalamus (Fig. 4) indicates the presence of both a potent, tonically active GABAergic inhibition of the dorsomedial hypothalamic neurons that influence BAT thermogenesis as well as a source of activation of these neurons upon removal of this inhibition. This tonic inhibition could arise from medial preoptic area neurons, and this hypothesis is illustrated as pathway 4 in Fig. 1. As described above, a tonic inhibition from neurons in the medial preoptic area could silence dorsomedial hypothalamic neurons and account for the absence of an effect on BAT thermogenesis following transections of their axons in the rostral pons (Fig. 1, bar B). In addition, removal of this inhibition from the medial preoptic area could account for the activation of BAT thermogenesis observed 1 with cuts behind the medial preoptic area (Fig. 1, bar C; Fig. 3B), 2 with activation of negatively coupled prostaglandin receptors following application of PGE2 into the medial preoptic area (13), 3 with a reduction in medial preoptic area warm-sensitive neuronal discharge by cold stimuli or pyrogens (4), or 4 with microinjection of bicuculline into dorsomedial hypothalamus (20). Although retrograde tracing experiments indicate a strong projection from the dorsomedial hypothalamus to the rostral raphe pallidus, no functional data are available to indicate whether dorsomedial hypothalamic efferent neurons (dotted pathway 3 in Fig. 1) directly excite BAT sympathetic premotor neurons or whether they increase BAT thermogenesis by inhibiting the discharge of descending pontine thermolytic neurons.

Ultimately, a model depending on the regulation of neuronal activity through disinhibition must include a mechanism responsible for exciting the neuronal population being studied. Neither the required medullary source of the excitatory drive to sympathetic premotor neurons for BAT in raphe pallidus (Fig. 1, pathway 1) nor an endogenous cellular mechanism supporting their spontaneous discharge has been identified. This is also true for the other neuronal populations in the proposed model, including those in the medial hypothalamic area, the dorsomedial hypothalamus, and the pons. Interestingly, a similar quandary regarding the activation of cardiovascular sympathetic premotor neurons in the rostral ventrolateral medulla after removal of their principal inhibitory input from the caudal ventrolateral medulla has also proven elusive. In this regard, both tonically active excitatory inputs and endogenous pacemaker capability have been proposed to account for the tonic discharge of these cardiovascular sympathtic premotor neurons.

Energy balance as a stimulus for BAT thermogenesis

Sympathetically regulated heat production in BAT occurs through the consumption of free fatty acids by the brown adipocyte mitochondria. Thus stimulation of BAT thermogen-esis provides a mechanism through which the central nervous system can increase the animal's overall energy expenditure and, in turn, influence the animal's supply of stored energy. This “secondary” role of BAT as a consumer of stored energy has received increased attention recently as the growing epidemic of human obesity has motivated research to improve our understanding of the neural mechanisms and networks that regulate adiposity and body weight. It is not surprising that studies have consistently shown a reciprocal relationship between an agent's effect on appetite and feeding and that on energy expenditure, suggesting the engagement of adipolytic or adipogenic “programs” that produce complementary responses in a range of effector mechanisms. Although several hormonal (leptin, insulin), substrate (glucose), and neural (vagal afferents from the liver) signals have been identified as potential regulators of the activity in neuronal pathways influencing energy balance and many of these have been postulated to alter the level of BAT sympathetic neuronal activity and BAT metabolism, direct examination of these hypotheses has been undertaken in only a few instances and determination of the neural pathways mediating these effects has not been accomplished.

Leptin is a hormone secreted by adipose tissue that provides those neurons containing leptin receptors with a signal related to body adiposity. In a simple negative feedback model, increased leptin signaling not only reduces appetite but also activates sympathetic outflows to white adipose tissue to increase lipolysis and the availability of circulating fatty acids and to BAT to stimulate mitochondrial oxidation of fatty acids, together reducing adipose tissue mass and returning the leptin signal and body adiposity to a “set point” value. Either intravenous or intracerebroventricular leptin administration increases not only BAT sympathetic nerve activity and BAT heat production but also renal sympathetic nerve activity but has only a small effect on arterial pressure.

Leptin is thought to initiate its multiple actions through binding to receptors on hypothalamic neurons, particularly those in the arcuate nucleus (Fig. 1) (7), to reduce the secretion of the orexigenic peptide, neuropeptide Y, and to stimulate release of α-melanocyte-stimulating hormone, which binds to melanocortin-4 receptors to increase energy expenditure. The paraventricular and lateral nuclei of the hypothalamus, both of which have descending projections to the

“This “secondary” role of BAT as a consumer of stored energy has received increased attention recently as the growing epidemic of human obesity has motivated research...”
medulla and spinal cord, have been identified as important projection targets of arcuate neurons (Fig. 1). Application of neuropeptide Y to the paraventricular hypothalamus reduces uncoupling protein-1 in BAT (2). Intracerebroventricular administration of the melanocortin-4 receptor agonist MTII potently activates BAT sympathetic nerve activity, BAT thermogenesis, and energy expenditure, as well as renal sympathetic nerve activity, in a manner similar to leptin administration (8). However, the location and transmitter phenotype of the melanocortin-4 receptor-containing neurons responsible for BAT activation remain unknown, as do the pathways and neurotransmitters through which they produce activation of BAT sympathetic premotor neurons and, ultimately, energy expenditure.

In conclusion, although many of the details of the central pathways determining the sympathetic outflow driving BAT metabolism and thermogenesis remain to be discovered, considerable recent progress in this area has been stimulated by the potential that increased understanding of these central mechanisms may provide for novel interventions to combat hyperthermia and obesity. In addition, the finding that alterations in the early development environment can influence sympathetic function in the adult (11) suggests that a variety of maternal and environmental factors can play important roles in determining the optimal function of the thermoregulatory and energy balance mechanisms in the adult. A location of putative sympathetic premotor neurons controlling BAT, the medullary cornerstone of the BAT thermogenic control network, has been identified in the rostral raphe pallidus, but determining the phenotype of these cells and the mechanisms governing their activity and responsiveness remain challenging experimental goals. The identification of hypothalamic regions sensitive to thermoregulatory and to metabolic signaling coupled with the localization of the sympathetic premotor neurons conveying an integrated excitatory drive to the spinal sympathetic neurons regulating BAT sympathetic nerve activity holds the promise to make significant progress in understanding the functional organization of the hierarchical central circuits governing BAT thermogenesis. The possibility that disinhibition of the sympathetic premotor neurons for BAT may be the principal mode for regulating BAT sympathetic nerve activity represents a novel sympathetic control strategy whose explication for BAT thermogenesis could provide clues not only to the sympathetic regulation of other metabolic targets but also to the cellular interactions responsible for tonic excitation within sympathetic generating networks.

I thank Dr. Christopher Madden for his comments on a draft version of this review.

I am grateful for the support of the Department of Health and Human Services, National Institutes of Health, through National Institute of Diabetes and Digestive and Kidney Diseases Grants DK-57838 and DK-20378 and National Institute of Neurological Diseases and Stroke Grant NS-40987.

References


