

Central Neural and Endocrine Mechanisms of Non-Exercise Activity Thermogenesis and Their Potential Impact on Obesity

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The rise in obesity is associated with a decline in the amount of physical activity in which people engage. The energy expended through everyday non-exercise activity, called non-exercise activity thermogenesis (NEAT), has a considerable potential impact on energy balance and weight gain. Comparatively little attention has been paid to the central mechanisms of energy expenditure and how decreases in NEAT might contribute to obesity. In this review, we first examine the sensory and endocrine mechanisms through which energy availability and energy balance are detected that may influence NEAT. Second, we describe the neural pathways that integrate these signals. Lastly, we consider the effector mechanisms that modulate NEAT through the alteration of activity levels as well as through changes in the energy efficiency of movement. Systems that regulate NEAT according to energy balance may be linked to neural circuits that modulate sleep, addiction and the stress response. The neural and endocrine systems that control NEAT are potential targets for the treatment of obesity.

Key words: physical activity, energy balance, hypothalamus.

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Obesity in the context of decreasing physical activity

Obesity is an ever-increasing problem now that more than one billion people worldwide are overweight or obese (1). More than one-half of Americans are overweight, and the proportion of the population that is overweight, obese, or extremely obese continues to grow (2, 3). Developing nations are quickly catching up with more developed countries with respect to rates of obesity and related health problems (4). This trend cuts across ethnicity, age, gender, and education level (5, 6). Childhood obesity, which portends adult obesity and predicts health problems in adulthood (7), continues to increase (8). Obesity is associated with a slew of deleterious health effects and health-related risk factors, including diabetes, cardiovascular disease, high cholesterol, asthma, arthritis, sleep apnoea, and cancer (6, 9–13), and is also linked with decreased life span (14), disability (15), and a lower in quality of life (16).

Weight gain results when energy expenditure is inappropriately low for the caloric intake of an individual. The role of dietary intake and its underlying mechanisms have been the principal focus when examining the rise in obesity (17, 18). Obesity incidence has risen, however, even as dietary quality has improved or energy intake has been reduced (17, 18). This circumstance necessitates a critical examination of the other side of the energy balance equation,

namely energy expenditure. As illustrated in Fig. 1, total daily energy expenditure (TDEE) consists of basal metabolic rate (BMR), the thermic effect of food, and the energy expenditure of activity. Some studies find a decreased BMR in those predisposed to become obese (19–21). Even in these obesity-prone individuals, however, the decrease in BMR could not fully explain the positive energy balance incurred (19), and the energy expenditure of physical activity was directly implicated in the development of obesity (20). Smaller, tightly controlled studies lend little support to the belief that differences in resting energy expenditure contribute to the tendency of an individual to gain weight (22, 23). Next, the thermic effect of food contributes little to TDEE in terms of both absolute kcal/day and variability. This suggests that the remaining component of TDEE is likely to change with obesity and the propensity to gain weight, specifically the energy expenditure of activity. At the population level, sedentary behaviour has increased steadily as physical activity has decreased, and this corresponds to increased obesity and decreased health in both children and adults (22, 24–28). The current trend in the general population is opposed to communities with heavy physical activity demands, which show very low rates of obesity (29). Indeed, increasing physical activity levels may be the missing link needed to reverse the current obesity trends (18, 22, 24, 25, 27, 30).

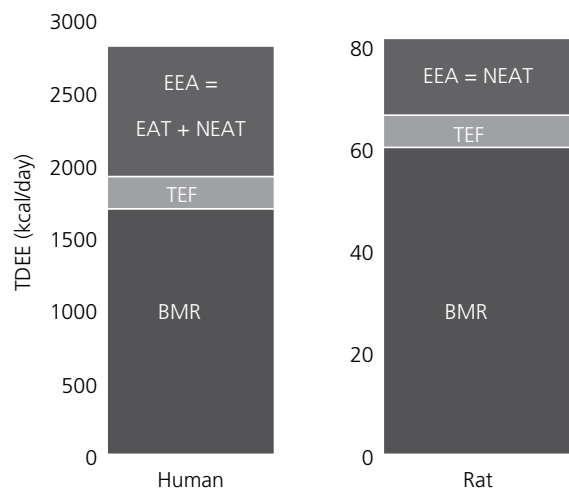


Fig. 1. Total daily energy expenditure (TDEE) in humans and rats. In all mammals, TDEE is comprised of basal metabolic rate (BMR), the thermic effect of food (TEF), and the energy expenditure of activity (EEA). In humans, the energy expenditure of activity is further divided up into exercise activity thermogenesis (EAT), which is the energy expended through volitional exercise, and non-exercise activity thermogenesis (NEAT), which is comprised of the energy expended through all other activity. In rodents, which do not have volitional exercise *per se*, all of the EEA can be considered NEAT.

The energy expended in activity can be separated into exercise and non-exercise activity thermogenesis (EAT and NEAT; Fig. 1). This final component, NEAT, is the most highly variable portion of TDEE among individuals, accounting for between 100 and 800 kcal/day (31, 32). The activities that comprise NEAT (i.e. everything from shopping to yard work to fidgeting) have an important influence on energy balance. Even small changes in the energy expenditure of physical activity can, over time, have a substantial impact on body weight (33, 34). Overfeeding results in compensatory increases in energy expenditure (35, 36). Moreover, individuals who show high NEAT in the face of increased caloric intake are more able to fend off weight gain than those who do not increase their NEAT (37). Lastly, there may be inherent, biologically based, familial traits that shape NEAT, which are reflected in differences in energy expenditure and body weight (38, 39). Both overall NEAT and the response of NEAT to dietary manipulation are highly variable between individuals, and individuals' physical activity levels have several biological and nonbiological influences that are not directly affected by energy balance (40). Social, cultural, environmental, and cognitive inputs may override biological cues for physical activity levels, allowing caloric intake to unduly influence energy balance. Evidence points to a prominent role of decreased physical activity and NEAT in the increasing rates of obesity in our increasingly deskbound society, and enhancing the contribution of NEAT to energy expenditure can help equalise the energy balance equation and prevent weight gain. Because NEAT is both highly variable and highly labile, it has a large potential to impact energy balance and body weight. Dysregulation of NEAT may have a considerable influence on body weight regulation and obesity. Here, we describe the potential phys-

iological mechanisms driving physical activity that may vary across the population and interact with changes in caloric intake. As human NEAT has been previously reviewed at length (41), we concentrate on animal studies that reveal promising potential biological regulators of NEAT.

The definition of NEAT in humans is clear: all energy expended due to activity excluding volitional exercise is considered NEAT. But how do we define NEAT in animals? Because animals do not have volitional exercise *per se*, we define animal NEAT as encompassing the energy expenditure of all activity, including spontaneous, locomotor, and more stationary or repetitive activities such as grooming. All animal physical activities expend energy and are subject to biological regulation and thus are a part of NEAT and equivalent to human NEAT. Confusion may also occur when examining activity in a running wheel. Although stimuli that heighten NEAT in animals may similarly increase wheel running, it is clear that wheel running and spontaneous physical activity are not equivalent (42). Running wheels often induce anomalous effects on the quantity and patterns of activity as well as on food intake and other behaviours (43–46), including rewarding properties of stimuli (47). Sometimes, the running wheel is even used to model 'exercise' in rodents (48). Therefore, a running wheel introduces confounding effects on the amount of activity and energy expenditure that are independent of energy balance regulation. Although studies employing the activity wheel can give us additional insight into the activity-promoting properties of hormones and neuropeptides, we chose to focus preferentially on brain systems that regulate general activity and energy expenditure.

Although NEAT is a critical variable when considering energy balance, little is known about the neural, endocrine, and molecular mechanisms affecting physical activity and NEAT. Changes in food availability can change NEAT in humans (37). Conversely, food restriction and starvation also predictably alter locomotor activity and energy expenditure in several species, including humans (49–54). The effects of caloric restriction depend on several variables (length and amount of restriction, ambient temperature, size of animal) and should be considered in the context of the species, environment, and energy balance: in general, food restriction results in an acute increase in activity and foraging, whereas long-term, severe restriction or starvation decrease physical activity, which then conserves energy. However, little is known about the mechanisms of these effects (54). There is an abundance of data describing mechanisms controlling the other side of the energy balance equation, namely food intake, and the biological mechanisms driving it. Physical activity has equal potential to affect weight change, however, and relatively little attention is being paid to the biological mechanisms regulating it. It is interesting that many of the same molecular, neural, and endocrine factors that affect feeding also modulate physical activity. Consequently, this rich, abundant source of knowledge can be used to gather information regarding how NEAT is regulated in animals and how these changes in physical activity affect body weight. In this review, we examine the endocrine and neural bases of physical activity and how they may potentially affect body weight through NEAT.

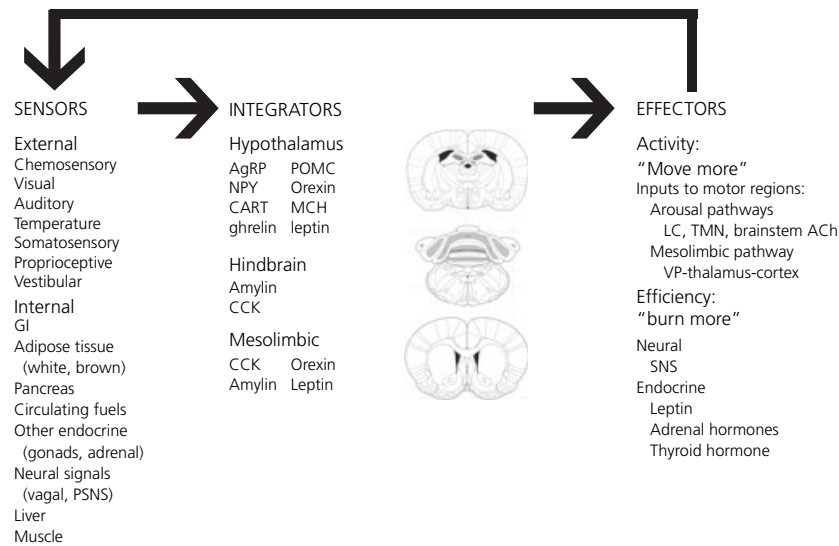


Fig. 2. The proposed model for the neuroendocrine regulation of non-exercise activity thermogenesis (NEAT) in the service of energy balance. To regulate NEAT according to changes in energy balance, the brain interprets external sensory cues of energy availability as well as internal metabolic cues. These signals are interpreted and integrated by several brain regions, including hypothalamic energy balance sites such as the arcuate nucleus, hindbrain regions including the area postrema and nucleus of the solitary tract, and the mesolimbic dopamine pathway. Lastly, these brain systems have ascending and descending projections that affect the amount of physical activity through arousal and limbic pathways, as well as descending neural projections and endocrine signals that alter the energy efficiency of physical activity. In this way, the brain can use NEAT to adjust energy balance in cases of energy surplus or deficiency. Brain atlas figures taken from Paxinos and Watson (263). ACh, acetylcholine; AgRP, agouti-related peptide; CART, cocaine- and amphetamine-regulated transcript; CCK, cholecystokinin; LC, locus coeruleus; MCH, melanin-concentrating hormone; NPY, neuropeptide Y; POMC, pro-opiomelanocortin; PSNS, parasympathetic nervous system; SNS, sympathetic nervous system; TMN, tuberomammillary nucleus; VP, ventral pallidum.

Biological regulatory systems can be understood based on their inputs and outputs: how stimuli are sensed, how the information they convey is integrated, and how the effector systems then alter physical activity and the associated energy expenditure through central and peripheral mechanisms (Fig. 2, Table 1). Through a systemic approach to investigating changes and individual differences in NEAT, we may discover potential targets for the manipulation of NEAT in the service of altering energy balance and treating obesity. We hypothesise that NEAT functions to promote negative energy balance in the face of increased caloric intake. To alter NEAT according to energy balance, sensory mechanisms need to detect changes in energy balance or energy availability. First, we need to identify the sensor mechanisms that inform the brain of excess caloric intake. Second, we need to determine how these signals are integrated. Lastly, effector mechanisms that increase NEAT need to be activated.

Sensors

To adjust energy expenditure according to positive or negative energy balance, the current metabolic status must be sensed. These sensor mechanisms might utilise visual, auditory, chemosensory, somatosensory, proprioceptive, vestibular, or temperature cues. Sensor mechanisms can detect external or environmental cues that inform the animal of the ready availability of food as well as pregastric sensory contact with food that mediates cephalic-phase

responses (55). Olfactory and gustatory (i.e. chemoreceptive) processing of food also influences satiety (56). Chemosensory tissues contain receptors for leptin (57, 58) and orexin (59, 60), which may alter gustatory and olfactory perception. Indeed, fasting increases olfactory acuity (61). Conversely, olfactory bulbectomy differentially alters appetite in obesity-prone and obesity-resistant rats, and increases novel open field locomotor activity in both types of rats (62). The current obesity trends underscore the impact of environmental factors in the tendency to gain weight. Obesity has risen sharply in the past few decades, a time scale that does not allow for change in the genetic or physiological systems underlying energy balance. Thus far, we are failing to effectively deal with emerging environmental and social factors that promote positive energy balance and obesity. A poor diet is not the only factor contributing to the current obesity trends, however, as demonstrated by increasing obesity even in situations where dietary fat or calorie consumption does not increase (17, 18). The modern environment also permits sedentary behaviour, which contributes to positive energy balance by decreasing energy expenditure (63), just as other aspects of Western living also affect energy balance through homeostatic and cognitive, nonhomeostatic cues (64, 65). Neither of these sets of environmental cues (i.e. those of excess food availability or decreased need for movement) are sufficient in and of themselves to prompt effective compensatory mechanisms that either decrease energy intake or increase energy expenditure, at least in some individuals. It is possible that sensory cues such as

Table 1. Central and Peripheral Signals of Energy Balance That Modulate Appetite and Have the Potential to Affect the Energy Expenditure of Activity.

Hormone/neuropeptide	Appetite	Site of synthesis	Activity and/or NEAT	Reference
Ghrelin	↑	GI	↓	(69, 71)
Obestatin	↓?	GI	No change	(76, 77)
Cholecystokinin	↓	GI	↓	(79, 81, 83, 84)
Glucagon-like peptide-1	↓	GI	No change	(85, 262)
Peptide YY	↓	GI	No change	(85)
Leptin	↓	Adipose, GI	↑	(90–94)
Insulin	↓	Pancreas	No change	(99)
Amylin	↓	Pancreas	↓	(101, 103, 104)
Glucagon	↓	Pancreas	No change	(106)
Free fatty acids	↓	Diet (circulation), adipose	↓	(112)
Glucose	↓	Diet (circulation)	No change	(109)
Thyroid hormone	↑	Thyroid	↑	(113)
Glucocorticoids	↑	Adrenal cortex	–	(116)
Testosterone	↑	Testis	↑	(118, 119)
Oestradiol	↓	Ovary	↑	(118, 120, 124–128)
Progesterone	↑	Ovary	↓	(122)
Neuropeptide Y	↑	Brain (hypothalamus)	↑↓	(69, 147–151)
Agouti-related peptide	↑	Brain (hypothalamus)	↓	(69, 145)
Cocaine- and amphetamine-regulated transcript	↓	Brain (hypothalamus)	↑	(136)
Pro-opiomelanocortin	↓	Brain (hypothalamus)	↑	(140–142)
Orexin	↑	Brain (hypothalamus)	↑	(72, 73)
Melanin-concentrating hormone	↑	Brain (hypothalamus)	↓	(165–168)
Thyrotrophin-releasing hormone	↓	Brain (hypothalamus)	↑	(169–171)
Urocortin	↓	Brain (hypothalamus)	↑	(172)
Corticotrophin-releasing hormone	↓	Brain (hypothalamus)	↑	(172)
Neuromedin U	↓	GI, Brain	↑	(74)
Galanin-like peptide	↑	Brain (hypothalamus)	↓	(174, 175)
Calcitonin gene-related peptide	↓	Brain (hypothalamus)	↓	(101, 103, 104, 176)
Brain-derived neurotrophic factor	↓	Brain (hypothalamus)	↓	(179)

NEAT, Non-exercise activity thermogenesis.

olfactory and taste stimuli are able to trigger appetite but not able to effectively activate compensatory mechanisms to adjust for excess caloric intake.

Here, we consider internal metabolic sensory cues, signals that convey long- or short-term energy stores or availability. Many of these hormones and peptides alter different aspects of activity, including locomotion and behaviours such as grooming or rearing. First, we focus on general locomotion rather than running wheel activity in order to try and capture non-exercise activity (42), and attempt to describe activity in similar terms as the authors do (e.g. all activity, locomotion, vertical movement, or specific behaviours such as grooming). NEAT in animals encompasses the energy expenditure of all of these activities (Fig. 1). Second, although all animal movements contribute to NEAT by definition, we highlight how metabolic cues alter movement specifically 'in the service of energy balance', or to promote positive or negative energy balance. These cues can be separated into hunger and satiety signals, and can be separated further into indicators of energy intake, energy stores, or circulating fuels. To begin with, many of the hormones released from the gastrointestinal tract that increase appetite also affect NEAT in animals (66). Ghrelin, which is synthesised in

stomach and other tissues (67), is a well-characterised and robust hunger signal and appears to be important in long-term control of appetite (68, 69). The ability of ghrelin to increase food intake is well known (70). More recently, however, it has been demonstrated that a single central intracerebroventricular (i.c.v.) microinjections of ghrelin can decrease 24-h locomotor activity in rats (69, 71). It is interesting that ghrelin appears to modulate long-term NEAT in rats, whereas many of the other hormones and neuropeptides investigated alter short-term NEAT (72–74). A peptide encoded on the ghrelin gene, obestatin, has the opposite effects of ghrelin on food intake (75, 76). Obestatin was not able to alter physical activity or energy expenditure, however, at least when administered alone (77).

Cholecystokinin (CCK) is a peptide synthesised throughout the gastrointestinal tract that also acts on the brain to decrease appetite (78). CCK has also been found to decrease locomotion when given both centrally and systemically (79–82). More specifically, CCK induces a 'satiety sequence' that includes decreased locomotor activity and rearing (81). Like many other hormones and neuropeptides reviewed here, CCK modulates anxiety-related behaviours and differentially affects locomotor activity depending on the animal's

environment and previous stressful experience (83). On the other hand, rats that are deficient in the CCK-A receptor gene show significantly less spontaneous activity compared to control rats, specifically fewer large, exploratory movements during the dark phase (84). Two other intestinal peptides secreted in response to ingestion and decrease food intake, glucagon-like peptide-1 and peptide YY (GLP-1 and PYY), also decrease appetite (85). However, no significant changes in locomotion or NEAT have been found in association with these peptides.

Other peripheral signals relay information to the brain regarding long-term energy store surplus and originate from sources such as adipose tissue. The adiposity signal leptin was first characterised in 1995 (86), when it was shown that it decreases food intake and body weight in leptin-deficient *ob/ob* mice without changing lean body mass. After 21 days of treatment with leptin, these mice also showed increased oxygen consumption, implicating a change in energy expenditure as well as energy intake. When examining changes in physical activity and energy expenditure, comparing animals of differing body mass can prove difficult. Body size in itself can affect physical activity (87) and influence oxygen consumption, energy expenditure, and their calculations, which are often stated in terms of body weight (88, 89). Earlier investigations of *ob/ob* mice, however, described well-controlled studies demonstrating that *ob/ob* and control mice have different levels of physical activity (90). Dauncey and Brown (90) investigated *ob/ob* mice and their lean littermates when they were at the same body weight. The young leptin-deficient mice expended less energy and showed less motor activity than their lean littermates. In adulthood, TDEE was similar in lean and obese mice, so the energy expended relative to metabolic size was greater in the lean mice, which were more active than the *ob/ob* mice. Dauncey and Brown (90) calculated that the energy expenditure of activity accounted for at least part of the altered body weight between *ob/ob* and wild-type mice. Clark and Gay (91) also found that the *ob/ob* mice showed half of the locomotor/exploratory activity as their wild-type littermates after their body weight was reduced to normal range by dietary restriction. These studies suggest that leptin normally acts to increase energy expenditure, at least partially through increasing physical activity. More recently, Bagnaso *et al.* (92) microinjected the leptin transgene into hypothalamic sites and found that the rats showed weight loss and increased nonshivering thermogenesis; physical activity was not examined. When leptin signalling is selectively restored in the arcuate nucleus of rats, there is a decrease in body weight and food intake and a marked increase in locomotor activity (93). Lastly, in humans, fasting leptin levels were significantly associated with the energy expenditure of physical activity (i.e. NEAT) (94). These results suggest that leptin acts not only to decrease feeding, but also to increase energy expenditure through promoting NEAT. The potential role of leptin in human NEAT is especially intriguing given the role of leptin insensitivity in obesity (95, 96).

Insulin is also considered to be a signal of long-term stores as its secretion is correlated with body fat stores. The pancreatic hormone insulin acts as an adiposity signal in the brain to reduce appetite and body weight in the regulation of energy balance,

separate from its peripheral effects on glucose uptake (97). Insulin given i.c.v. decreases food intake and body weight (98). The development of insulin and leptin resistance is one proposed mechanism of obesity (95). Insulin has also been found to influence energy expenditure (99). Specifically, insulin microinjected into the hypothalamic paraventricular nucleus (PVN) increases energy expenditure, but no differences in locomotor activity were detected (99). The possibility remains, however, that central insulin increases NEAT through altering the efficiency of physical activity. Amylin is a peptide cosecreted with insulin that affects food intake (100). Amylin induces satiety, reduces meal size, and decreases body weight and adiposity, and amylin antagonists increase food intake when given i.c.v (101, 102). Amylin appears to act in the brain to decrease locomotor activity: central administration of amylin decreases spontaneous locomotor activity after 1 h (101, 103), and open field activity after 3 and 6 h (104). Reports differ on the ability of amylin to affect stationary physical activity, with one report showing increases in rearing and grooming (104) and another that utilised lower doses of amylin showing no change in grooming (103). Lastly, glucagon also decreases meal size, with hepatic vagal communication to the brain important in this process (105). Intra-PVN glucagon increases thermogenesis but does not affect the amount of locomotor activity (106). In summary, the only pancreatic hormone that reliably affects physical activity levels is amylin, which acts in the hindbrain to decrease locomotion.

Metabolic fuels and macronutrients affect appetite and have the potential to affect NEAT as well. Decreases in glucose utilisation trigger increased appetite (107, 108). Although the idea that dietary sucrose increases locomotor activity is an attractive hypothesis, the preponderance of evidence from studies conducted mostly on children does not support a role for dietary glucose in the modulation of physical activity (109). Similar to glucose sensing, inhibition of fatty acid oxidation also produces hunger so that fatty acids act as signal of metabolic fuels (110). Specifically, within hypothalamic cells, fatty acid metabolism or triglyceride utilisation indicates macronutrient availability and subsequently alters appetite (111). In this way, long-chain fatty acids may act as a signal for signal energy surplus (110). As for NEAT, it has also been hypothesised that lipids decrease physical activity (112).

Other peripheral hormones have the capacity to affect both energy intake and expenditure. It is known that thyroid hormone increases energy expenditure, and rats rendered hyperthyroid increase their physical activity and NEAT (113). Thyroid hormone can also alter the energy efficiency of activity, as described below. Corticosterone plays an important role in feeding and energy balance. Corticosterone increases food intake by acting on receptors in the PVN (114). Adrenalectomy in mice or obese Zucker rats causes them to reduce food intake and, in general, fat intake and body fat is correlated with circulating glucocorticoids (115). Could a decrease in NEAT account for the increased body weight due to corticosterone? Very little information is available concerning glucocorticoids and NEAT. One study found that spontaneous locomotion was increased in the male offspring of corticosterone-treated pregnant dams (116), and another found that lack of central nervous system glucocorticoid receptors led to decreased food intake and energy

efficiency (117). The ability to examine the role of glucocorticoids in NEAT is hampered by the effects of stressors on both adrenal steroid levels and locomotion, as well as the independent effects of glucocorticoids on energy balance.

Lastly, gonadal hormones affect locomotor activity in male and female rodents. In general, female rodents show higher levels of activity than do males (87). Increases in activity are associated with testosterone as well as oestradiol, which also has anorectic effects; ovariectomy results in increased body weight, decreased activity, and increased energy intake (118–121). The ability of oestradiol to increase activity is counteracted by progesterone (122). Activity, food intake, and body weight also vary with the oestrous cycle, with activity peaking at times when circulating oestradiol levels are highest and food intake and body weight are lowest (123, 124). Interpretation is made more difficult because locomotion is most often measured using running wheels, which have effects on activity and energy expenditure independent of energy balance. One study examining nonwheel locomotor activity in female rats showed decreases in ambulatory activity in ovariectomised rats (125), and limited evidence supports the idea that cyclic changes in spontaneous activity seen in female rats are associated with increased energy expenditure (124). More recently, oestrogen has been linked to general arousal in females, encompassing sexual arousal, activity, and anxiety (126) through its actions on oestrogen receptors (127). The type of activity (i.e. wheel running versus home cage activity) may also be differentially mediated by oestrogen receptors α and β (127). The brain sites and mechanisms through which oestradiol affects different types of activity may not be the same (127, 128) and alterations in anxiety affect movement in an open field (129), further complicating investigations of this subject. Unlike many of the endocrine factors described here, oestradiol is not necessarily a signal of peripheral energy balance, but rather adjusts energy balance according to the current needs of the organism based on reproductive status (130), and thereby has a potentially large impact on body weight and NEAT.

Integration of energy balance signals

Peripheral hormones that indicate energy balance status act on the brain, which then integrates the information to affect energy intake and expenditure. Both peripheral sensor mechanisms and central neuropeptide systems that regulate feeding are notoriously redundant. Again, many of the neuropeptides and brain regions important in modulating appetite also affect energy balance through their modulation of NEAT. Some brain regions and neuropeptides associated with the control of food intake are more important than others when considering the control of NEAT, and the brain circuits controlling each function appear to diverge. The arcuate nucleus of the hypothalamus is critical for the integration of central and peripheral signals that modulate energy balance (93, 131, 132). The mechanism through which the arcuate nucleus affects appetite centres on the actions of two cell types. One cell type is orexigenic and contains neuropeptide Y (NPY) and agouti-related peptide (AgRP), whereas the other set of neurones is anorexigenic and contains cocaine- and amphetamine-regulated transcript (CART) and

pro-opiomelanocortin (POMC) (93, 131, 133). Many peripheral signals that affect appetite are thought to do so through their actions on these groups of arcuate neurones. For example, leptin stimulates the CART/POMC neurones and inhibits the NPY/AgRP neurones to decrease appetite (134). The arcuate nucleus also integrates information conveyed by different peripheral signals of energy balance (132). Central CART peptide reduces appetite as well as NPY-induced feeding, indicating an interaction between the two classes of arcuate neurones involved in feeding behaviour (135). Others have found that CART is orexigenic, however (136). Might these same cells have actions on locomotor activity and NEAT? Restoring leptin signalling in the arcuate nucleus of leptin receptor-null mice dramatically increases physical activity in these mice (93), implying that the effects of leptin on locomotor activity are mediated through the arcuate nucleus. The arcuate nucleus and the peptides contained in its neurones appear to be central to the integration of energy balance cues for the regulation of not only energy intake, but also NEAT.

If the neuropeptides contained within the arcuate nucleus alter NEAT to reach energy balance homeostasis, then one would expect feeding signals (NPY and AgRP) to decrease NEAT and satiety signals (CART and POMC) to increase it. First, microinjections of CART(55–102) into the arcuate induce increases in locomotion (136), supporting the hypothesis outlined above. The regulation of energy balance by POMC occurs primarily through the actions of α -melanocyte-stimulating hormone (α -MSH) on brain melanocortin receptors (MCR, specifically MC3R and MC4R), tonically inhibiting food intake (137). Serotonin also appears to mediate its effects on food intake through melanocortin signalling pathways (138, 139). But what about energy expenditure and NEAT? Male mice deficient in the MC4R show decreased locomotor activity, and mice deficient in MC4R also showed decreased oxygen consumption compared to wild-types (140). α -MSH given i.c.v., as well as both α -MSH and MC1R agonist infused into the ventral tegmental area (VTA), increased grooming and locomotor activity in rats, both NEAT-promoting activities (141). Another study found no effect of α -MSH on locomotion, however (142). Clues to the role of melanocortins in NEAT can also be garnered from studies in AgRP, which acts primarily as a melanocortin receptor inverse agonist but also has other actions (143). AgRP when given i.c.v. increases appetite (144). Similar to ghrelin, i.c.v. AgRP also induces significant long-term (up to 72 h) decreases in locomotor activity (69). Mice deficient in AgRP also show age-related decreases in body weight as well as increased locomotor activity (145). In general, agents that block the AgRP increase NEAT, suggesting that melanocortins may tonically activate NEAT.

NPY is perhaps the most widely recognised hunger signal (146, 147). There is no consensus regarding the effect of NPY on NEAT, however. Early studies reported that central application of NPY induced sedation, decreased physical activity, and decreased body temperature (148, 149). NPY has also been associated with decreased thermogenesis, and the promotion of positive energy balance (147). Others have reported no change in physical activity after NPY or even increases in activity or wakefulness with decreased sleep (69, 150, 151). It has been suggested that NPY is

related to the behaviours associated with dark onset in nocturnal animals, such as increased feeding and activity (150). An early idea suggested that NPY was associated with foraging as well as feeding (i.e. both the appetitive and consummatory aspects of appetite) because NPY increased activity in rats, but only when food was not readily available (151). NPY and NPY receptor agonists increase appetitive as well as consummatory aspects of feeding because they increase foraging and food hoarding in Siberian hamsters (152). Like orexin, the teleological role of NPY may not be assigned strictly to either positive or negative energy balance regulation, and this is reflected in its varied effects on physical activity in different situations.

Further integration of NEAT signals

Other hypothalamic and forebrain nuclei also play an important role in sensing hunger signals and integrating information regarding energy balance, including the PVN, lateral hypothalamic area (LHA), ventromedial nucleus (VMN), dorsomedial nucleus, periventricular nucleus, septum, and preoptic nucleus (131, 153). For example, glucose can be directly sensed by the hypothalamus, specifically by neurones in the LHA, arcuate nucleus, and VMN, as well as by brainstem neurones (107, 154). These cells act as sensor-integrator-effector neurones that may be important in the development of obesity (107, 155). Lastly, sensory innervation of adipose tissue also helps to regulate adiposity (156), indicating that the brain also senses energy balance through this route. The PVN is likely to be especially important in the ability of neural signals to alter NEAT. The PVN is known to mediate the locomotor-activating effect of several neuropeptides (74, 157, 158), affect energy expenditure (99, 106) through sympathetic activation, and have indirect projections to adipose tissue (159). Altogether, these features imply that the PVN is integral to the neuropeptidergic control of physical activity and energy expenditure. One example of the importance of the PVN in the modulation of NEAT is the increase in physical activity induced by neuromedin U (NMU) (74). NMU is a gastrointestinal peptide synthesised in hypothalamic nuclei including the arcuate and VMN (160). Central NMU decreases food intake and increases NEAT in rats, partially through its actions on the PVN (74, 157, 161). This may be one pathway through which leptin acts to alter physical activity and energy expenditure as leptin induces hypothalamic release of NMU, and leptin-induced hypophagia is attenuated after pretreatment with anti-NMU antisera (157, 161).

Neurones containing melanin-concentrating hormone (MCH) reside in similar brain regions as those containing orexin, but the two peptides are not colocalised (162). But, like orexin, MCH stimulates appetite (163), and mice lacking MCH eat less and display a lean phenotype (162, 164). MCH may also have a role in the regulation of NEAT because MCH-deficient mice show an increased metabolic rate (164). Mice deficient in the MCH 1 receptor (MCH1r) have reduced body fat, are hyperactive, and have higher night time energy expenditure than their wild-type counterparts (165, 166). The decreased susceptibility to diet-induced obesity in these mice is likely to be due to their increased NEAT (165, 166), and mice

deficient in MCH1r are more active than wild-type mice (167). These studies suggest that MCH normally acts to decrease physical activity and NEAT, and that MCH may be another route through which leptin affects NEAT and obesity. Leptin-deficient mice that are also lacking the gene encoding MCH are lean compared to *ob/ob* mice, but are also hyperphagic. The lean phenotype is due in part to their increased physical activity and therefore NEAT (168). Several lines of evidence suggest that one role of MCH is to increase NEAT in response to positive energy balance as signalled by increased circulating leptin; this may be one route through which MCH decreases obesity (165, 166, 168).

Several other brain neuropeptides that affect appetite also alter NEAT. Thyrotrophin-releasing hormone (TRH) given centrally decreases food intake, indicating that its effects on appetite are separate from its role as a releasing hormone (169). TRH similarly enhances physical activity levels (169–171). Urocortin is involved in satiety and also increases grooming when microinjected centrally; its effects on locomotor activity may be suppressive and are more complicated, however, due to its interaction with corticotrophin-releasing hormone (CRH) (172). Galanin-like peptide (GALP) also alters energy intake (173), although its effects on NEAT and energy expenditure are less certain. GALP has also been found to decrease food intake, body weight, and locomotion, although these results are difficult to interpret given that GALP also induced a conditioned taste aversion (174, 175). Central administration of calcitonin gene-related peptide (CGRP), also associated with parasympathetic sensory fibres and found in the nucleus of the solitary tract (NTS), decreases spontaneous locomotor activity and open field activity (101, 103, 104, 176). This peptide also suppresses food intake (177). Similar results were found with calcitonin receptor-stimulating peptide (178). Alterations in brain-derived neurotrophic factor (BDNF) did not follow the same pattern. Mice heterozygous for the gene encoding a BDNF mutation also show differences in body weight and locomotor activity (179). Of the heterozygous mice, half of the mice develop obesity. Compared to wild-types, as well as the one-half of heterozygotes that show increased body fat composition, the nonfat heterozygotes display almost twice the amount of physical activity (179). This implies that the increased NEAT seen in this portion of the heterozygotes contributed to their ability to fend off weight gain.

The brain peptide orexin belongs in its own category due to its ability to increase both appetite and NEAT. Orexin A and B (also called hypocretin 1 and 2) act on orexin receptors in areas of the brain important in the sleep-wake cycle as well as hypothalamic nuclei central to metabolism and feeding. Unlike many hunger signals described above, orexin consistently and robustly increases both locomotor activity and energy expenditure (180). This may reflect the role of orexin in relaying circadian information related to vigilance and metabolism (181). Accordingly, orexin is associated with activities and states that normally occur during the active phase such as physical activity, wakefulness, and increases in both energy intake and expenditure. The importance of rhythmic orexin is reflected in its ability to increase feeding during the light (inactive) phase in nocturnal rodents, a time when orexin release is

normally low (182). Whereas central orexins increase feeding at only certain times of day, locomotion and energy expenditure are increased regardless of the time of administration (183, 184), further underlining the importance of orexin in the regulation of locomotion, energy expenditure, and NEAT. Moreover, the effects of orexin on energy expenditure appear to outweigh (so to speak) its effects on food intake because orexin knockout mice show increases in body weight despite decreased food intake (185). Ironically, it is precisely because of its ability to induce coordinated increases in appetite and energy expenditure that makes orexin an attractive target for the increase in energy expenditure through NEAT. Insensitivity to satiety or adiposity signals such as leptin may result in heightened appetite along with decreased NEAT, a one-way road toward obesity. Increased appetite due to orexin, on the other hand, is likely to be associated with increased physical activity and energy expenditure to offset the weight gain incurred.

Several of the hormones addressed in this review exert their effects on appetite through their actions on hindbrain nuclei. The area postrema (AP) is a circumventricular organ important in the integration of several circulating signals (186). The adjacent NTS receives input from the AP, further integrates the information with other incoming neural signals, and sends projections to other brain regions important in appetite (186). These caudal brainstem nuclei are important in sensing and integrating information received from signals indicating digestion and sensation of food, as well as circulating and stored fuels as indicated by receptor content, direct actions of hormones and peptides on these brain regions, and lesion studies (56, 186). The gastrointestinal peptides that induce satiety and decrease meal size include CCK, glucagon, glucagon-like peptides, somatostatin, enterostatin, peptide YY, oxyntomodulin, and peptides of the bombesin family; many of these peptides are also synthesised in brain regions that regulate energy balance (187, 188). The NTS is responsive to satiety factors secreted from the gut as well as to hypothalamic neuropeptides, leptin, and insulin, and have reciprocal connections with hypothalamic nuclei important in energy balance (186, 187). Importantly, the vagus nerve also relates gastrointestinal information to the NTS (186).

How might the gastrointestinal satiety signals, hindbrain nuclei, and vagal afferent nerves influence NEAT? First, CCK does not fit the usual pattern of satiety signals of increasing NEAT. Through its actions on the NTS, CCK initiates a behavioural satiety sequence that includes decreased locomotor activity, probably through activation of CCK-A receptors that affect dopaminergic mechanisms (78, 79, 81, 84, 189). The vagus nerve is also important in mediating the ability of CCK to induce satiety (190). For its inhibitory effects on locomotion, the effects of peripheral but not central administration of CCK were reliant on an intact vagal pathway (189). Amylin, a pancreatic peptide, acts primarily in the AP to induce satiety and specifically to control meal size, and also acts as an adiposity signal; similar effects are found with CGRP (100, 191, 192). Similar to CCK, amylin does not increase NEAT; indeed, amylin inhibits open field locomotion (101, 103, 104), possibly acting through dopaminergic mechanisms (193) or through orexin and MCH (100). Another circulating signal sensed by the AP is glucose; decreased glucose utilisation triggers hunger, but evidence does support the

idea that sugar intake alters NEAT, as described above. Lastly, hindbrain areas are more sensitive to the appetite-suppressive actions of bombesin, whereas bombesin is likely to act in forebrain areas to increase locomotion (194, 195). Generally speaking, when compared to satiety signals that act primarily in the hypothalamus, hindbrain satiety mechanisms are associated with decreased locomotor activity rather than increasing NEAT to adjust energy balance. This could be due to the triggering of a satiety sequence rather than a signal of excess energy intake. The different effects of hindbrain vs hypothalamic peptides in NEAT and energy balance may have phylogenetic implications.

The limbic system, including the amygdala and the mesolimbic dopamine pathway, is also important for the integration of NEAT-related neural and hormonal signals. This system includes the VTA and nucleus accumbens (NAC) and is important in reward and addiction (196). Along with its importance in addiction, the mesolimbic pathway also mediates the rewarding value associated with food (197). Several of the hormones and neuropeptides discussed here act on these brain structures to alter physical activity (170, 189, 193, 194, 198–200). Orexin A acts in the shell region of the NAC to induce both food intake and locomotor activity (198). Ghrelin also acts in the VTA to increase feeding, presumably through increased dopaminergic neurotransmission (199). Although ghrelin decreases long-term physical activity in rats (69, 71), the effects of intra-VTA ghrelin on activity were not examined (199). In addition, the locomotor-activating and -sensitizing effects of amphetamine were decreased in leptin-deficient *ob/ob* mice compared to wild-types, and this effect was reversed by infusing leptin peripherally (201), suggesting that leptin interacts with the mesolimbic dopamine system. Moreover, leptin decreases the firing rate of VTA neurons, and intra-VTA leptin decreases food intake (202). Although intra-VTA leptin did not affect locomotion, activity was decreased when leptin receptor expression was suppressed in the VTA; this was accompanied by increased food intake (202). Both CCK (189) and amylin (193) act in the NAC to decrease locomotor activity. It is interesting that CCK only decreased open field locomotion if the environment was novel (203). It should also be noted that unilateral (right) intra-NAC CCK infusions actually increased locomotion, as did intra-amygdala infusions of low doses (but not higher doses, which, again, decreased activity) (200). Lastly, intra-VTA administration of melanocortin agonists increase vertical activity (204). The limbic system integrates information from several neuropeptides that affect NEAT.

Divergence of mechanisms controlling appetite and NEAT

Many of the hypothalamic factors that increase appetite also decrease NEAT, and other factors that decrease appetite increase NEAT. Although the neural mechanisms mediating food intake have been studied intensely, few efforts have been made to determine how these hormones and peptides affect physical activity and NEAT (71). The brain mechanisms mediating the ability of neuroendocrine factors to alter energy intake and expenditure appear to diverge at some point. For example, the CRH receptor 2 was found to be more

critical for the effects of CRH on appetite (205), which suggests a divergence in the central mechanisms of CRH on appetite versus physical activity. Bombesin, which decreases food intake and increases locomotor activity, acts on different brain sites to initiate these effects (194, 195, 206). Similarly, the pathways mediating thermogenic and feeding responses to NPY also diverge (207). Results such as these also imply that changes in locomotor activity are not dependent on changes in food intake and vice-versa.

Effector mechanisms

Once the peripheral and hormonal cues conveying information regarding energy balance have been integrated by central networks, effector systems can then alter NEAT appropriately. Within an individual, NEAT can be amplified in two different ways, either through increasing the amount of physical activity or through decreasing the efficiency of movement so that the energy expenditure required for the movement is increased. Because potential mechanisms that modulate the amount of physical activity are more apparent, relatively more information is known regarding the integration of these systems compared to the mechanisms underlying changes in skeletal muscle work efficiency. How might signals increase the amount of physical activity? Many of the brain systems controlling appetite and metabolism interact with brain monoaminergic arousal systems such as the noradrenergic neurones in the locus coeruleus (LC), serotonergic neurones in the raphe nuclei, dopaminergic neurones in several brain systems including the mesolimbic pathway, and the histaminergic neurones in the tuberomammillary nucleus, as well as cholinergic nuclei in the brainstem. All of these systems are important in arousal, vigilance, and physical activity and interact with striatal, thalamic, or cortical systems, as well as descending motor and autonomic systems (208). For example, orexin modulates these monoaminergic arousal systems and stimulates widespread cortical activation (209). It is possible that these arousal systems are utilised as an effector mechanism to alter NEAT and reach energy balance homeostasis.

Many factors also act through the mesolimbic dopamine system to alter activity levels. Although the NAC core region has indirect projections to premotor cortex via the basal ganglia (210), the NAC shell region has been the focus of studies examining neuropeptide-induced locomotion (198, 201). For the locomotion mediated by the mesolimbic pathway, the NAC output pathway involving the ventral pallidum (VP), mediodorsal thalamus, and prefrontal cortex is a likely candidate for affecting the amount of physical activity (210, 211). Although this pathway has been investigated mostly with respect to the hedonic aspects of rewarding stimuli, the VTA–NAC–VP circuit also mediates the increased locomotion induced by a novel environment (211). It could well be that some of the energy balance signals reviewed here tap into this circuit to affect locomotor activity. This is an especially enticing possibility given that, for many of the hormones and neuropeptides discussed here, locomotor activity is altered depending on the novelty of the environment, possibly due to interactions between brain mechanisms underlying physical activity and fear or anxiety (200, 212–214).

NEAT can also be altered through descending neural systems that modulate autonomic systems or muscle tone. For example, ghrelin alters thermogenesis, probably by affecting the sympathetic nervous system (215). Also, orexin acting on the LC increases skeletal muscle tone (216). Orexin has descending projections to spinal cord regions that increase sympathetic preganglionic neurone excitation and modulate autonomic tone, thereby affecting energy homeostasis (217). There are also descending serotonergic systems that modulate autonomic tone. Importantly, one brain region that projects to autonomic systems is the PVN, also shown to mediate increases in NEAT from many neuropeptides (72, 74). The PVN increases sympathetic drive (218) and controls sympathetic projections, including those to both white and brown adipose tissue (219). Another hypothalamic nucleus, the VMN, plays a role in increasing glucose uptake in tissues that use energy but not tissues that store energy (220). In this way, the brain can activate autonomic projections and increase the energy utilisation associated with physical activity.

Lastly, the energy efficiency of activity can be modulated by endocrine systems activated by the same ascending and descending neural systems described above. The work efficiency of skeletal muscle is decreased and the energy expenditure of physical activity is increased during weight loss or caloric restriction, reflecting a resistance to the maintenance of altered body weight that is supported by the autonomic nervous system (35, 36, 49, 221, 222), especially sympathetic drive (223). Moreover, the energy expenditure of activity appears to contribute the most to the changes in energy expenditure after experimental weight change (35). In humans, weight loss results in increased skeletal muscle work efficiency, whereas weight gain has the opposite effect, both opposing the change in body weight and adiposity by altering NEAT (221). Low-dose leptin treatment reverses the effects of weight loss on efficiency, sympathetic tone, and thyroid hormone levels, all of which presumably contributed to the increased energy expenditure shown by these subjects (221, 224, 225). It is especially intriguing that changes in skeletal muscle work efficiency were only detectable at low levels of physical activity, hinting that the work efficiency of NEAT, rather than exercise or resting thermogenesis, may be the critical to weight loss in this scenario (225). Within skeletal myocytes, the mechanism for this change may be the alteration of uncoupling proteins, some of which show decreased expression after weight loss (226); this might contribute to the increased efficiency and decreased energy expenditure of movement. Adrenal hormones associated with energy utilisation such as glucocorticoids and epinephrine increase the fuels available to tissues that utilise energy during increased activity, such as cardiac and skeletal muscle. Thyroid hormone also affects metabolism (227), and possibly skeletal muscle thermogenesis (228), but increases work efficiency and decreases skeletal muscle glycogen utilisation (229). It is interesting to note that the brain systems involved in the release of these hormones, such as CRH and TRH, also increase NEAT, highlighting the redundant coordination of NEAT and energy balance.

Differences in NEAT are seen in association with obesity and obesity propensity, as well as with alterations in diet and body weight (37, 38). As an example, variations in NEAT could be due to individual differences in brain orexin. Increased orexin release or

sensitivity in lean individuals would lead to increased levels of activity through the activation of brain arousal systems. Orexin is also sensitive to changes in energy balance through, among other mechanisms, circulating leptin and triglycerides (230, 231), implying that these signals have the potential to alter NEAT through their actions on orexin. Several of the sensor and integrator systems discussed here, including orexin, have direct or indirect access to CRH-containing cells within the PVN and have the ability to induce CRH release (232–234). In addition to decreasing food intake, CRH (i.c.v.) dose-dependently increases motor activity in rats, promoting negative energy balance through both its anorectic actions and increasing physical activity and thermogenesis (158, 205, 235). CRH neurones from the PVN contact both orexin-containing neurones (236) and noradrenergic neurones in the LC (237), which then activate cortical arousal and motor systems. Lastly, CRH activates the hypothalamic-pituitary-adrenal axis causing the release of corticosteroids, which can affect the work efficiency of skeletal muscle. CRH has been found to increase brown adipose tissue thermogenesis and heart rate through its actions on the DMH (238). This is one possible route through which alterations in energy balance might result in changes in NEAT.

Implications for energy balance and NEAT

Reward

As described above, the neural systems involved in addiction also mediate the rewarding aspects of food and interact with cognitive systems impacted by environmental influences (64, 65, 199, 201, 202, 239). Moreover, this same circuit mediates changes in locomotor activity initiated by several metabolic hormones and neuropeptides. For example, CCK acts in the NAC to decrease locomotion (240), and the suppressive actions of amylin on locomotion may be through the NAC (193). Also, orexin A changes synaptic efficacy in the VTA, suggesting a role for orexin in neural plasticity and addiction (241). Activation of orexin neurones is associated with both food and drug reward, as demonstrated using conditioned place preferences, as well as the reinstatement of drug-seeking (242). This mechanistic overlap implies that these peptides may also modulate the rewarding or addictive properties of stimuli, and further suggests the possibility that changes in NEAT may be linked to not only the novelty or rewarding aspects of foods, but also to other rewarding stimuli, both endogenous and exogenous. Drugs of abuse that increase locomotion through this brain reward system may override endogenous NEAT signals, thereby altering energy balance homeostasis. Moreover, other alterations in brain hedonics associated with, for example, depression or eating disorders might also be linked to changes in NEAT through this same pathway. Such data raise the question of whether the actions of neuropeptides such as orexin on NEAT might interact with the brain mechanisms that mediate addiction. First, there are individual differences in NEAT that relate to obesity as well as to changes in hypothalamic orexin levels and central orexin sensitivity (72, 180). Is there a relationship between obesity or the propensity for obesity, decreased NEAT, decreased orexin sensitivity, and the addictive or rewarding

properties of drugs or food? These two processes (reward and NEAT) may or may not interact with respect to orexin signalling, depending on whether the same subset of orexin neurones is involved in NEAT and the modulation of reward (242). Conversely, if environmental cues related to rewarding stimuli activate brain orexin neurones (242), how might this affect NEAT and the ability of NEAT to combat obesity? Similar to orexin, leptin may also affect the brain reward system: leptin-deficient *ob/ob* mice demonstrate decreased dopamine synthesis, content, and release after NAC stimulation compared to wild-type mice (201). Lastly, chronic amphetamine treatment affects the ability of intra-NAC CCK antagonists to alter amphetamine-induced locomotion in rats (243). These data suggest that endogenous and exogenous rewarding stimuli may not only alter NEAT, also but might modify the way energy balance signals modulate NEAT.

Stress

Systems and peptides that are important for the modulation of NEAT are also integral to the stress response (244). It has been suggested that CRH and related peptides such as urocortin mimic the stress response by increasing arousal and decreasing appetite (245). On the other hand, Heinrichs and Joppa (246) argued that the locomotor-activating and anxiety-like effects of CRH are mechanistically separable and, furthermore, that the arousal induced by CRH is the more physiologically relevant action of the peptide (235, 246). The PVN, a brain region important of the integration of NEAT-promoting signals, is central to the stress response because of both the action of CRH cells on peripheral hormone levels as well as the central actions of CRH. Moreover, many of the NEAT signals described here appear to interact with brain mechanisms controlling stress- and anxiety-related behaviours. For example, even though CCK is known to act in the NAC to decrease locomotion, the effect is only seen in a novel environment and not in a familiar one (240). The ability of CCK to modulate activity is also dependent on the stress response of the animal because a CCK receptor antagonist given in the NAC only affected activity in a novel environment if the rats were previously exposed to chronic restraint stress (243). Rotziner and Vaccarino (83) suggested that CCK-B receptors in the amygdala mediate the effects of CCK on anxiety-related behaviours, whereas CCK-A receptors in the NAC are important for reward-related behaviours. Other NEAT factors, including oestradiol (212) and galanin (244), also influence fear- and anxiety-related behaviours. Moreover, these processes are also altered by diet. For example, the ability of restraint stress to induce corticosterone secretion in rats was augmented by a high-fat diet (247), and rats exposed to a high-fat diet for only 4 days showed a similar enhancement of corticosterone secretion (248). Findings such as these are interesting in that they suggest that the mechanisms controlling NEAT are intertwined with those controlling anxiety and the stress response. This suggests the possibility that differences in anxiety or responsiveness to stress might be related to individual differences in obesity propensity due to variability in NEAT. Diet-induced obese rats have been shown to be hyporesponsive to chronic stress and show

reduced amygdala CRH expression (249, 250), both of which may contribute to their decreased NEAT compared to lean rats. It might be assumed that brain mechanisms controlling the behavioural response to stress have been co-opted to alter NEAT in the service of energy balance. The use of running wheels or the a novel environment can modulate how neuropeptides and hormones affect NEAT (42, 83). Altogether, studies such as these underscore the importance of closely examining the methods used to measure locomotion.

Sleep

Most of the research on the interaction between obesity and sleep has appropriately focused on the effects of weight gain on sleep apnoea and the resulting increase in sleepiness, although other interactions between sleep and energy balance systems are beginning to be recognised (251, 252). Many of the systems that potentially affect energy balance and NEAT are also involved in the modulation of the sleep-wake cycle to different degrees. The most salient example of this is the orexin system. The two functions of orexin that were first recognised were to induce feeding and modulate the onset of rapid eye movement sleep (253, 254). Subsequently, the role of orexin in the modulation of energy balance has risen to prominence, as has its role in the induction of NEAT (255). More recently, other systems that primarily modulate energy balance have also been shown to affect sleep. Ghrelin promotes slow-wave sleep; obestatin, which is derived from the same gene as ghrelin, has the opposite effect (256, 257). Leptin increases sleep (258), and NPY decreases it (150). Overall, the mechanisms of these actions are not as well understood as are the mechanisms mediating the effects of orexin on sleep (259, 260). In the case of orexin, although it is likely that the neural substrates through which orexin modulates sleep and NEAT overlap, it is unlikely that orexin increases NEAT exclusively through decreasing sleep. Intra-PVN orexin increases NEAT during both the active and inactive phases of the cycle in rats (184), and orexin increases activity to levels far above those predicted if it simply inhibited sleep (73, 184). Again, it might be suggested that the orexin system and its downstream mechanisms were co-opted to serve energy balance by modulating NEAT.

Summary

A multitude of neural and endocrine factors that affect appetite also have a demonstrable and predictable effect on NEAT. Compared to the neural mechanisms of appetite, only a modest amount of information is available describing the neural mechanisms that give rise to changes in NEAT in the service of energy balance. This is especially true regarding the effects of metabolic cues on human energy balance with respect to NEAT or energy expenditure in general (261). As the importance of NEAT in human energy balance is increasingly being recognised, the influences of diet and genetic background on physical activity and NEAT are being examined more

closely. Because of the importance of the modern sedentary environment on obesity trends, and especially because of the failure of current treatments to effectively combat obesity in the majority of individuals, we should recognise that potential environmental, behavioural, and pharmacological manipulations of NEAT open up many avenues for the treatment of obesity.

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References

- 1 James WP, Rigby N, Leach R. Obesity and the metabolic syndrome: the stress on society. *Ann NY Acad Sci* 2006; **1083**: 1–10.
- 2 Gregg EW, Cheng YJ, Cadwell BL, Imperatore G, Williams DE, Flegal KM, Narayan KM, Williamson DF. Secular trends in cardiovascular disease risk factors according to body mass index in US adults. *JAMA* 2005; **293**: 1868–1874.
- 3 Ogden CL, Carroll MD, Curtin LR, McDowell MA, Tabak CJ, Flegal KM. Prevalence of overweight and obesity in the United States, 1999–2004. *JAMA* 2006; **295**: 1549–1555.
- 4 James PT. Obesity: the worldwide epidemic. *Clin Dermatol* 2004; **22**: 276–280.
- 5 Ogden CL, Fryar CD, Carroll MD, Flegal KM. Mean body weight, height, and body mass index, United States 1960–2002. *Adv Data* 2004; **347**: 1–17.
- 6 Mokdad AH, Ford ES, Bowman BA, Dietz WH, Vinicor F, Bales VS, Marks JS. Prevalence of obesity, diabetes, and obesity-related health risk factors, 2001. *JAMA* 2003; **289**: 76–79.
- 7 Dietz WH. Health consequences of obesity in youth: childhood predictors of adult disease. *Pediatrics* 1998; **101**: 518–525.
- 8 Ogden CL, Flegal KM, Carroll MD, Johnson CL. Prevalence and trends in overweight among US children and adolescents, 1999–2000. *JAMA* 2002; **288**: 1728–1732.
- 9 Hensrud DD, Weinsier RL, Darnell BE, Hunter GR. Relationship of co-morbidities of obesity to weight loss and four-year weight maintenance/rebound. *Obes Res* 1995; **3** (Suppl. 2): 217S–222S.
- 10 Lawrence VJ, Kopelman PG. Medical consequences of obesity. *Clin Dermatol* 2004; **22**: 296–302.
- 11 Calle EE, Rodriguez C, Walker-Thurmond K, Thun MJ. Overweight, obesity, and mortality from cancer in a prospectively studied cohort of US adults. *N Engl J Med* 2003; **348**: 1625–1638.
- 12 Hubert HB, Feinleib M, McNamara PM, Castelli WP. Obesity as an independent risk factor for cardiovascular disease: a 26-year follow-up of participants in the Framingham Heart Study. *Circulation* 1983; **67**: 968–977.
- 13 Young T, Palta M, Dempsey J, Skatrud J, Weber S, Badr S. The occurrence of sleep-disordered breathing among middle-aged adults. *N Engl J Med* 1993; **328**: 1230–1235.
- 14 Olshansky SJ, Passaro DJ, Hershow RC, Layden J, Carnes BA, Brody J, Hayflick L, Butler RN, Allison DB, Ludwig DS. A potential decline in life expectancy in the United States in the 21st century. *N Engl J Med* 2005; **352**: 1138–1145.

- 15 Vita AJ, Terry RB, Hubert HB, Fries JF. Aging, health risks, and cumulative disability. *N Engl J Med* 1998; **338**: 1035–1041.
- 16 Heo M, Allison DB, Faith MS, Zhu S, Fontaine KR. Obesity and quality of life: mediating effects of pain and comorbidities. *Obes Res* 2003; **11**: 209–216.
- 17 Popkin BM, Siega-Riz AM, Haines PS. A comparison of dietary trends among racial and socioeconomic groups in the United States. *N Engl J Med* 1996; **335**: 716–720.
- 18 Prentice AM, Jebb SA. Obesity in Britain: gluttony or sloth? *BMJ* 1995; **311**: 437–439.
- 19 Ravussin E, Lillioja S, Knowler WC, Christin L, Freymond D, Abbott WG, Boyce V, Howard BV, Bogardus C. Reduced rate of energy expenditure as a risk factor for body-weight gain. *N Engl J Med* 1988; **318**: 467–472.
- 20 Saltzman E, Roberts SB. The role of energy expenditure in energy regulation: findings from a decade of research. *Nutr Rev* 1995; **53**: 209–220.
- 21 Roberts SB, Savage J, Coward WA, Chew B, Lucas A. Energy expenditure and intake in infants born to lean and overweight mothers. *N Engl J Med* 1988; **318**: 461–466.
- 22 Weinsier RL, Nelson KM, Hensrud DD, Darnell BE, Hunter GR, Schutz Y. Metabolic predictors of obesity. Contribution of resting energy expenditure, thermic effect of food, and fuel utilization to four-year weight gain of post-obese and never-obese women. *J Clin Invest* 1995; **95**: 980–985.
- 23 Weinsier RL, Hunter GR, Zuckerman PA, Darnell BE. Low resting and sleeping energy expenditure and fat use do not contribute to obesity in women. *Obes Res* 2003; **11**: 937–944.
- 24 Arluk SL, Branch JD, Swain DP, Dowling EA. Childhood obesity's relationship to time spent in sedentary behavior. *Mil Med* 2003; **168**: 583–586.
- 25 Wyatt HR, Peters JC, Reed GW, Barry M, Hill JO. A Colorado statewide survey of walking and its relation to excessive weight. *Med Sci Sports Exerc* 2005; **37**: 724–730.
- 26 Rissanen AM, Heliövaara M, Knekt P, Reunanen A, Aromaa A. Determinants of weight gain and overweight in adult Finns. *Eur J Clin Nutr* 1991; **45**: 419–430.
- 27 Weinsier RL, Hunter GR, Heini AF, Goran MI, Sell SM. The etiology of obesity: relative contribution of metabolic factors, diet, and physical activity. *Am J Med* 1998; **105**: 145–150.
- 28 Hu FB, Li TY, Colditz GA, Willett WC, Manson JE. Television watching and other sedentary behaviors in relation to risk of obesity and type 2 diabetes mellitus in women. *JAMA* 2003; **289**: 1785–1791.
- 29 Bassett DR, Schneider PL, Huntington GE. Physical activity in an Old Order Amish community. *Med Sci Sports Exerc* 2004; **36**: 79–85.
- 30 Heini AF, Weinsier RL. Divergent trends in obesity and fat intake patterns: the American paradox. *Am J Med* 1997; **102**: 259–264.
- 31 Donahoo WT, Levine JA, Melanson EL. Variability in energy expenditure and its components. *Curr Opin Clin Nutr Metab Care* 2004; **7**: 599–605.
- 32 Ravussin E, Lillioja S, Anderson TE, Christin L, Bogardus C. Determinants of 24-hour energy expenditure in man. Methods and results using a respiratory chamber. *J Clin Invest* 1986; **78**: 1568–1578.
- 33 Dauncey MJ. Activity and energy expenditure. *Can J Physiol Pharmacol* 1990; **68**: 17–27.
- 34 Livingstone MB, Strain JJ, Prentice AM, Coward WA, Nevin GB, Barker ME, Hickey RJ, McKenna PG, Whitehead RG. Potential contribution of leisure activity to the energy expenditure patterns of sedentary populations. *Br J Nutr* 1991; **65**: 145–155.
- 35 Leibel RL, Rosenbaum M, Hirsch J. Changes in energy expenditure resulting from altered body weight. *N Engl J Med* 1995; **332**: 621–628.
- 36 Hirsch J, Hudgins LC, Leibel RL, Rosenbaum M. Diet composition and energy balance in humans. *Am J Clin Nutr* 1998; **67**: 551S–555S.
- 37 Levine JA, Eberhardt NL, Jensen MD. Role of nonexercise activity thermogenesis in resistance to fat gain in humans. *Science* 1999; **283**: 212–214.
- 38 Levine JA, Lanningham-Foster LM, McCrady SK, Krizan AC, Olson LR, Kane PH, Jensen MD, Clark MM. Interindividual variation in posture allocation: possible role in human obesity. *Science* 2005; **307**: 584–586.
- 39 Zurlo F, Ferraro RT, Fontvielle AM, Rising R, Bogardus C, Ravussin E. Spontaneous physical activity and obesity: cross-sectional and longitudinal studies in Pima Indians. *Am J Physiol* 1992; **263**: E296–E300.
- 40 Fruhbeck G. Does a NEAT difference in energy expenditure lead to obesity? *Lancet* 2005; **366**: 615–616.
- 41 Levine JA. Nonexercise activity thermogenesis (NEAT): environment and biology. *Am J Physiol Endocrinol Metab* 2004; **286**: E675–E685.
- 42 Sherwin CM. Voluntary wheel running: a review and novel interpretation. *Anim Behav* 1998; **56**: 11–27.
- 43 Burghardt PR, Fulk LJ, Hand GA, Wilson MA. The effects of chronic treadmill and wheel running on behavior in rats. *Brain Res* 2004; **1019**: 84–96.
- 44 Blanchong JA, McElhinny TL, Mahoney MM, Smale L. Nocturnal and diurnal rhythms in the unstriped Nile rat, *Arvicanthis niloticus*. *J Biol Rhythms* 1999; **14**: 364–377.
- 45 Dixon DP, Ackert AM, Eckel LA. Development of, and recovery from, activity-based anorexia in female rats. *Physiol Behav* 2003; **80**: 273–279.
- 46 Kas MJ, Edgar DM. A nonphotic stimulus inverts the diurnal-nocturnal phase preference in *Octodon degus*. *J Neurosci* 1999; **19**: 328–333.
- 47 Belke TW, Wagner JP. The reinforcing property and the rewarding aftereffect of wheel running in rats: a combination of two paradigms. *Behav Processes* 2005; **68**: 165–172.
- 48 Patterson CM, Levin BE. Role of exercise in the central regulation of energy homeostasis and in the prevention of obesity. *Neuroendocrinology* 2007; Mar 19; [Epub ahead of print].
- 49 Civitarese AE, Carling S, Heilbronn LK, Hulver MH, Ukropcova B, Deutsch WA, Smith SR, Ravussin E. Calorie restriction increases muscle mitochondrial biogenesis in healthy humans. *PLoS Med* 2007; **3**: E76.
- 50 Weed JL, Lane MA, Roth GS, Speer DL, Ingram DK. Activity measures in rhesus monkeys on long-term calorie restriction. *Physiol Behav* 1997; **62**: 97–103.
- 51 Overton JM, Williams TD. Behavioral and physiologic responses to caloric restriction in mice. *Physiol Behav* 2004; **81**: 749–754.
- 52 Lynn SE, Breuner CW, Wingfield JC. Short-term fasting affects locomotor activity, corticosterone, and corticosterone binding globulin in a migratory songbird. *Horm Behav* 2003; **43**: 150–157.
- 53 Novak CM, Jiang X, Wang C, Teske JA, Kotz CM, Levine JA. Caloric restriction and physical activity in zebrafish (*Danio rerio*). *Neurosci Lett* 2005; **383**: 99–104.
- 54 Overton JM, Williams TD, Chambers JB, Rashotte ME. Cardiovascular and metabolic responses to fasting and thermoneutrality are conserved in obese Zucker rats. *Am J Physiol Regul Integr Comp Physiol* 2001; **280**: R1007–R1015.
- 55 Powley TL. Vagal circuitry mediating cephalic-phase responses to food. *Appetite* 2000; **34**: 184–188.
- 56 Rolls ET. Taste and olfactory processing in the brain and its relation to the control of eating. *Crit Rev Neurobiol* 1997; **11**: 263–287.
- 57 Baly C, Aioun J, Badonnel K, Lacroix MC, Durieux D, Schlegel C, Salesse R, Caillol M. Leptin and its receptors are present in the rat olfactory mucosa and modulated by the nutritional status. *Brain Res* 2007; **1129**: 130–141.

- 58 Kawai K, Sugimoto K, Nakashima K, Miura H, Ninomiya Y. Leptin as a modulator of sweet taste sensitivities in mice. *Proc Natl Acad Sci USA* 2000; **97**: 11044–11049.
- 59 Hardy AB, Aioun J, Baly C, Julliard KA, Caillol M, Salesse R, Duchamp-Viret P. Orexin A modulates mitral cell activity in the rat olfactory bulb: patch-clamp study on slices and immunocytochemical localization of orexin receptors. *Endocrinology* 2005; **146**: 4042–4053.
- 60 Caillol M, Aioun J, Baly C, Persuy MA, Salesse R. Localization of orexins and their receptors in the rat olfactory system: possible modulation of olfactory perception by a neuropeptide synthesized centrally or locally. *Brain Res* 2003; **960**: 48–61.
- 61 Aime P, Duchamp-Viret P, Chaput MA, Savigner A, Mahfouz M, Julliard AK. Fasting increases and satiation decreases olfactory detection for a neutral odor in rats. *Behav Brain Res* 2007; **179**: 258–264.
- 62 Primeaux SD, Barnes MJ, Bray GA. Olfactory bulbectomy increases food intake and hypothalamic neuropeptide Y in obesity-prone but not obesity-resistant rats. *Behav Brain Res* 2007; **180**: 190–196.
- 63 Levine JA, Kotz CM. NEAT – non-exercise activity thermogenesis – egocentric and geocentric environmental factors vs. biological regulation. *Acta Physiol Scand* 2005; **184**: 309–318.
- 64 Berthoud HR. Interactions between the 'cognitive' and 'metabolic' brain in the control of food intake. *Physiol Behav* 2007; Jan 12; [Epub ahead of print].
- 65 Berthoud HR. Homeostatic and non-homeostatic pathways involved in the control of food intake and energy balance. *Obesity (Silver Spring)* 2006; **14** (Suppl. 5): 1975–2005.
- 66 Murphy KG, Dhillon WS, Bloom SR. Gut peptides in the regulation of food intake and energy homeostasis. *Endocr Rev* 2006; **27**: 719–727.
- 67 Kojima M, Hosoda H, Date Y, Nakazato M, Matsuo H, Kangawa K. Ghrelin is a growth-hormone-releasing acylated peptide from stomach. *Nature* 1999; **402**: 656–660.
- 68 Tschöp M, Smiley DL, Heiman ML. Ghrelin induces adiposity in rodents. *Nature* 2000; **407**: 908–913.
- 69 Tang-Christensen M, Vrang N, Ortmann S, Bidlingmaier M, Horvath TL, Tschöp M. Central administration of ghrelin and agouti-related protein (83–132) increases food intake and decreases spontaneous locomotor activity in rats. *Endocrinology* 2004; **145**: 4645–4652.
- 70 Horvath TL, Diano S, Sotonyi P, Heiman M, Tschöp M. Minireview: ghrelin and the regulation of energy balance – a hypothalamic perspective. *Endocrinology* 2001; **142**: 4163–4169.
- 71 Castaneda TR, Jurgens H, Wiedmer P, Pfluger P, Diano S, Horvath TL, Tang-Christensen M, Tschöp MH. Obesity and the neuroendocrine control of energy homeostasis: the role of spontaneous locomotor activity. *J Nutr* 2005; **135**: 1314–1319.
- 72 Novak CM, Kotz CM, Levine JA. Central orexin sensitivity, physical activity, and obesity in diet-induced obese and diet-resistant rats. *Am J Physiol Endocrinol Metab* 2006; **290**: E396–E403.
- 73 Kotz CM, Wang C, Teske JA, Thorpe AJ, Novak CM, Kiwaki K, Levine JA. Orexin a mediation of time spent moving in rats: neural mechanisms. *Neuroscience* 2006; **142**: 29–36.
- 74 Novak CM, Zhang M, Levine JA. Neuromedin U in the paraventricular and arcuate hypothalamic nuclei increases non-exercise activity thermogenesis. *J Neuroendocrinol* 2006; **18**: 594–601.
- 75 Zhang JV, Ren PG, Avsian-Kretschmer O, Luo CW, Rauch R, Klein C, Hsueh AJ. Obestatin, a peptide encoded by the ghrelin gene, opposes ghrelin's effects on food intake. *Science* 2005; **310**: 996–999.
- 76 Gourcerol G, St-Pierre DH, Tache Y. Lack of obestatin effects on food intake: should obestatin be renamed ghrelin-associated peptide (GAP)? *Regul Pept* 2007; **141**: 1–7.
- 77 Nogueiras R, Pfluger P, Tovar S, Arnold M, Mitchell S, Morris A, Perez-Tilve D, Vazquez MJ, Wiedmer P, Castaneda TR, DiMarchi R, Tschöp M, Schürmann A, Joost HG, Williams LM, Langhans W, Diéguez C. Effects of obestatin on energy balance and growth hormone secretion in rodents. *Endocrinology* 2007; **148**: 21–26.
- 78 Little TJ, Horowitz M, Feinle-Bisset C. Role of cholecystokinin in appetite control and body weight regulation. *Obes Rev* 2005; **6**: 297–306.
- 79 Takeda Y, Takano Y, Kamiya H. Effects of cholecystokinin tetra and octa peptides on locomotor activity in mice. *Jpn J Pharmacol* 1986; **42**: 145–149.
- 80 Kapas L, Obal F Jr, Farkas I, Payne LC, Sary G, Rubicsek G, Krueger JM. Cholecystokinin promotes sleep and reduces food intake in diabetic rats. *Physiol Behav* 1991; **50**: 417–420.
- 81 Kapas L, Obal F Jr, Alfoldi P, Rubicsek G, Penke B, Obal F. Effects of nocturnal intraperitoneal administration of cholecystokinin in rats: simultaneous increase in sleep, increase in EEG slow-wave activity, reduction of motor activity, suppression of eating, and decrease in brain temperature. *Brain Res* 1988; **438**: 155–164.
- 82 Antin J, Gibbs J, Holt J, Young RC, Smith GP. Cholecystokinin elicits the complete behavioral sequence of satiety in rats. *J Comp Physiol Psychol* 1975; **89**: 784–790.
- 83 Rotzinger S, Vaccarino FJ. Cholecystokinin receptor subtypes: role in the modulation of anxiety-related and reward-related behaviours in animal models. *J Psychiatry Neurosci* 2003; **28**: 171–181.
- 84 Sei M, Sei H, Shima K. Spontaneous activity, sleep, and body temperature in rats lacking the CCK-A receptor. *Physiol Behav* 1999; **68**: 25–29.
- 85 Talsania T, Anini Y, Siu S, Drucker DJ, Brubaker PL. Peripheral exendin-4 and peptide YY(3–36) synergistically reduce food intake through different mechanisms in mice. *Endocrinology* 2005; **146**: 3748–3756.
- 86 Pellemounter MA, Cullen MJ, Baker MB, Hecht R, Winters D, Boone T, Collins F. Effects of the obese gene product on body weight regulation in ob/ob mice. *Science* 1995; **269**: 540–543.
- 87 Tou JC, Wade CE. Determinants affecting physical activity levels in animal models. *Exp Biol Med (Maywood)* 2002; **227**: 587–600.
- 88 Arch JR, Hislop D, Wang SJ, Speakman JR. Some mathematical and technical issues in the measurement and interpretation of open-circuit indirect calorimetry in small animals. *Int J Obes (Lond)* 2006; **30**: 1322–1331.
- 89 James WP, Davies HL, Bailes J, Dauncey MJ. Elevated metabolic rates in obesity. *Lancet* 1978; **1**: 1122–1125.
- 90 Dauncey MJ, Brown D. Role of activity-induced thermogenesis in twenty-four hour energy expenditure of lean and genetically obese (ob/ob) mice. *Q J Exp Physiol* 1987; **72**: 549–559.
- 91 Clark LD, Gay PE. Activity and body-weight relationships in genetically obese animals. *Biol Psychiatry* 1972; **4**: 247–250.
- 92 Bagnasco M, Dube MG, Kalra PS, Kalra SP. Evidence for the existence of distinct central appetite, energy expenditure, and ghrelin stimulation pathways as revealed by hypothalamic site-specific leptin gene therapy. *Endocrinology* 2002; **143**: 4409–4421.
- 93 Coppari R, Ichinose M, Lee CE, Pullen AE, Kenny CD, McGovern RA, Tang V, Liu SM, Ludwig T, Chua SC Jr, Lowell BB, Elmquist JK. The hypothalamic arcuate nucleus: a key site for mediating leptin's effects on glucose homeostasis and locomotor activity. *Cell Metab* 2005; **1**: 63–72.
- 94 Franks PW, Farooqi IS, Luan J, Wong MY, Halsall I, O'Rahilly S, Wareham NJ. Does physical activity energy expenditure explain the between-individual variation in plasma leptin concentrations after adjusting for differences in body composition? *J Clin Endocrinol Metab* 2003; **88**: 3258–3263.
- 95 Wang J, Obici S, Morgan K, Barzilai N, Feng Z, Rossetti L. Overfeeding rapidly induces leptin and insulin resistance. *Diabetes* 2001; **50**: 2786–2791.
- 96 Levin BE, Dunn-Meynell AA. Reduced central leptin sensitivity in rats with diet-induced obesity. *Am J Physiol Regul Integr Comp Physiol* 2002; **283**: R941–R948.

- 97 Niswender KD, Schwartz MW. Insulin and leptin revisited: adiposity signals with overlapping physiological and intracellular signaling capabilities. *Front Neuroendocrinol* 2003; **24**: 1–10.
- 98 Air EL, Strowski MZ, Benoit SC, Conarello SL, Salituro GM, Guan XM, Liu K, Woods SC, Zhang BB. Small molecule insulin mimetics reduce food intake and body weight and prevent development of obesity. *Nat Med* 2002; **8**: 179–183.
- 99 Menendez JA, Atrens DM. Insulin and the paraventricular hypothalamus: modulation of energy balance. *Brain Res* 1991; **555**: 193–201.
- 100 Lutz TA. Amylinergic control of food intake. *Physiol Behav* 2006; **89**: 465–471.
- 101 Bouali SM, Wimalawansa SJ, Jolicoeur FB. In vivo central actions of rat amylin. *Regul Pept* 1995; **56**: 167–174.
- 102 Rushing PA, Hagan MM, Seeley RJ, Lutz TA, D'Alessio DA, Air EL, Woods SC. Inhibition of central amylin signaling increases food intake and body adiposity in rats. *Endocrinology* 2001; **142**: 5035.
- 103 Clementi G, Valerio C, Emmi I, Prato A, Drago F. Behavioral effects of amylin injected intracerebroventricularly in the rat. *Peptides* 1996; **17**: 589–591.
- 104 Kovacs AM, Telegdy G. The effects of amylin on motivated behavior in rats. *Physiol Behav* 1996; **60**: 183–186.
- 105 Woods SC, Lutz TA, Geary N, Langhans W. Pancreatic signals controlling food intake; insulin, glucagon and amylin. *Philos Trans R Soc Lond B Biol Sci* 2006; **361**: 1219–1235.
- 106 Atrens DM, Menendez JA. Glucagon and the paraventricular hypothalamus: modulation of energy balance. *Brain Res* 1993; **630**: 245–251.
- 107 Levin BE, Dunn-Meynell AA, Routh VH. Brain glucose sensing and body energy homeostasis: role in obesity and diabetes. *Am J Physiol* 1999; **276**: R1223–R1231.
- 108 Miselis RR, Epstein AN. Feeding induced by intracerebroventricular 2-deoxy-D-glucose in the rat. *Am J Physiol* 1975; **229**: 1438–1447.
- 109 Wolraich ML, Wilson DB, White JW. The effect of sugar on behavior or cognition in children. A meta-analysis. *JAMA* 1995; **274**: 1617–1621.
- 110 Lam TK, Schwartz GJ, Rossetti L. Hypothalamic sensing of fatty acids. *Nat Neurosci* 2005; **8**: 579–584.
- 111 Pocai A, Lam TK, Obici S, Gutierrez-Juarez R, Muse ED, Arduini A, Rossetti L. Restoration of hypothalamic lipid sensing normalizes energy and glucose homeostasis in overfed rats. *J Clin Invest* 2006; **116**: 1081–1091.
- 112 Proulx K, Cota D, Castaneda TR, Tschop MH, D'Alessio DA, Tso P, Woods SC, Seeley RJ. Mechanisms of oleoylethanolamide (OEA) - induced changes in feeding behavior and motor activity. *Am J Physiol Regul Integr Comp Physiol* 2005; **289**: R729–R737.
- 113 Levine JA, Nygren J, Short KR, Nair KS. Effect of hyperthyroidism on spontaneous physical activity and energy expenditure in rats. *J Appl Physiol* 2003; **94**: 165–170.
- 114 Tempel DL, Leibowitz SF. Glucocorticoid receptors in PVN: interactions with NE, NPY, and Gal in relation to feeding. *Am J Physiol* 1993; **265**: E794–E800.
- 115 Castonguay TW. Glucocorticoids as modulators in the control of feeding. *Brain Res Bull* 1991; **27**: 423–428.
- 116 Diaz R, Fuxe K, Ogren SO. Prenatal corticosterone treatment induces long-term changes in spontaneous and apomorphine-mediated motor activity in male and female rats. *Neuroscience* 1997; **81**: 129–140.
- 117 Kellendonk C, Eiden S, Kretz O, Schutz G, Schmidt I, Tronche F, Simon E. Inactivation of the GR in the nervous system affects energy accumulation. *Endocrinology* 2002; **143**: 2333–2340.
- 118 Asarian L, Geary N. Modulation of appetite by gonadal steroid hormones. *Philos Trans R Soc Lond B Biol Sci* 2006; **361**: 1251–1263.
- 119 Wood RI. Oral testosterone self-administration in male hamsters: dose-response, voluntary exercise, and individual differences. *Horm Behav* 2002; **41**: 247–258.
- 120 Morgan MA, Pfaff DW. Estrogen's effects on activity, anxiety, and fear in two mouse strains. *Behav Brain Res* 2002; **132**: 85–93.
- 121 Wade GN, Gray JM. Gonadal effects on food intake and adiposity: a metabolic hypothesis. *Physiol Behav* 1979; **22**: 583–593.
- 122 Rodier WI III, Segal S. The effect of progesterone on the activity-wheel running of ovariectomized rats. *Horm Behav* 1977; **9**: 214–221.
- 123 Shaw MA, Whitaker EM, Hervey E, Hervey GR. The effects of ovarian hormones on regulation of energy balance in Zucker rats. *J Endocrinol* 1983; **98**: 165–171.
- 124 Anantharaman-Barr HG, Decombaz J. The effect of wheel running and the estrous cycle on energy expenditure in female rats. *Physiol Behav* 1989; **46**: 259–263.
- 125 Shimomura Y, Shimizu H, Takahashi M, Sato N, Uehara Y, Fukatsu A, Negishi M, Kobayashi I, Kobayashi S. The significance of decreased ambulatory activity during the generation by long-term observation of obesity in ovariectomized rats. *Physiol Behav* 1990; **47**: 155–159.
- 126 Frohlich J, Morgan M, Ogawa S, Burton L, Pfaff D. Statistical analysis of measures of arousal in ovariectomized female mice. *Horm Behav* 2001; **39**: 39–47.
- 127 Ogawa S, Chan J, Gustafsson JA, Korach KS, Pfaff DW. Estrogen increases locomotor activity in mice through estrogen receptor alpha: specificity for the type of activity. *Endocrinology* 2003; **144**: 230–239.
- 128 Fahrbach SE, Meisel RL, Pfaff DW. Preoptic implants of estradiol increase wheel running but not the open field activity of female rats. *Physiol Behav* 1985; **35**: 985–992.
- 129 Frohlich J, Morgan M, Ogawa S, Burton L, Pfaff D. Statistical analysis of hormonal influences on arousal measures in ovariectomized female mice. *Horm Behav* 2002; **42**: 414–423.
- 130 Wade GN, Schneider JE. Metabolic fuels and reproduction in female mammals. *Neurosci Biobehav Rev* 1992; **16**: 235–272.
- 131 Cone RD, Cowley MA, Butler AA, Fan W, Marks DL, Low MJ. The arcuate nucleus as a conduit for diverse signals relevant to energy homeostasis. *Int J Obes Relat Metab Disord* 2001; **25** (Suppl. 5): S63–S67.
- 132 Hewson AK, Tung LY, Connell DW, Tookman L, Dickson SL. The rat arcuate nucleus integrates peripheral signals provided by leptin, insulin, and a ghrelin mimetic. *Diabetes* 2002; **51**: 3412–3419.
- 133 Cone RD. Anatomy and regulation of the central melanocortin system. *Nat Neurosci* 2005; **8**: 571–578.
- 134 van den Top M, Spanswick D. Integration of metabolic stimuli in the hypothalamic arcuate nucleus. *Prog Brain Res* 2006; **153**: 141–154.
- 135 Kristensen P, Judge ME, Thim L, Ribel U, Christjansen KN, Wulff BS, Clausen JT, Jensen PB, Madsen OD, Vrang N, Larsen PJ, Hastrup S. Hypothalamic CART is a new anorectic peptide regulated by leptin. *Nature* 1998; **393**: 72–76.
- 136 Abbott CR, Rossi M, Wren AM, Murphy KG, Kennedy AR, Stanley SA, Zollner AN, Morgan DG, Morgan I, Ghatei MA, Small CJ, Bloom SR. Evidence of an orexigenic role for cocaine- and amphetamine-regulated transcript after administration into discrete hypothalamic nuclei. *Endocrinology* 2001; **142**: 3457–3463.
- 137 Ellacott KL, Cone RD. The central melanocortin system and the integration of short- and long-term regulators of energy homeostasis. *Recent Prog Horm Res* 2004; **59**: 395–408.
- 138 Zhou L, Williams T, Lachey JL, Kishi T, Cowley MA, Heisler LK. Serotonergic pathways converge upon central melanocortin systems to regulate energy balance. *Peptides* 2005; **26**: 1728–1732.
- 139 Heisler LK, Jobst EE, Sutton GM, Zhou L, Borok E, Thornton-Jones Z, Liu HY, Zigman JM, Balthasar N, Kishi T, Lee CE, Aschkenasi CJ, Zhang CY, Yu J, Boss O, Mountjoy KG, Clifton PG, Lowell BB, Friedman JM, Horvath T, Butler AA, Elmquist JK, Cowley MA. Serotonin reciprocally regulates melanocortin neurons to modulate food intake. *Neuron* 2006; **51**: 239–249.

- 140 Ste Marie L, Miura GI, Marsh DJ, Yagaloff K, Palmiter RD. A metabolic defect promotes obesity in mice lacking melanocortin-4 receptors. *Proc Natl Acad Sci USA* 2000; **97**: 12339–12344.
- 141 Watts AG, Khan AM, Sanchez-Watts G, Salter D, Neuner CM. Activation in neural networks controlling ingestive behaviors: What does it mean, and how do we map and measure it? *Physiol Behav* 2006; **89**: 501–510.
- 142 Vergoni AV, Bertolini A, Mutulis F, Wikberg JE, Schiöth HB. Differential influence of a selective melanocortin MC4 receptor antagonist (HS014) on melanocortin-induced behavioral effects in rats. *Eur J Pharmacol* 1998; **362**: 95–101.
- 143 Pritchard LE, White A. Agouti-related protein: more than a melanocortin-4 receptor antagonist? *Peptides* 2005; **26**: 1759–1770.
- 144 Stutz AM, Morrison CD, Argypoulos G. The Agouti-related protein and its role in energy homeostasis. *Peptides* 2005; **26**: 1771–1781.
- 145 Wortley KE, Anderson KD, Yasenchak J, Murphy A, Valenzuela D, Diano S, Yancopoulos GD, Wiegand SJ, Sleeman MW. Agouti-related protein-deficient mice display an age-related lean phenotype. *Cell Metab* 2005; **2**: 421–427.
- 146 Chronwall BM, Zukowska Z. Neuropeptide Y, ubiquitous and elusive. *Peptides* 2004; **25**: 359–363.
- 147 Levine AS, Jewett DC, Cleary JP, Kotz CM, Billington CJ. Our journey with neuropeptide Y: effects on ingestive behaviors and energy expenditure. *Peptides* 2004; **25**: 505–510.
- 148 Heilig M, Wahlestedt C, Widerlov E. Neuropeptide Y (NPY)-induced suppression of activity in the rat: evidence for NPY receptor heterogeneity and for interaction with alpha-adrenoceptors. *Eur J Pharmacol* 1988; **157**: 205–213.
- 149 Jolicœur FB, Michaud JN, Rivest R, Menard D, Gaudin D, Fournier A, St-Pierre S. Neurobehavioral profile of neuropeptide Y. *Brain Res Bull* 1991; **26**: 265–268.
- 150 Szentirmai E, Krueger JM. Central administration of neuropeptide Y induces wakefulness in rats. *Am J Physiol Regul Integr Comp Physiol* 2006; **291**: R473–R480.
- 151 Stanley BG, Leibowitz SF. Neuropeptide Y injected in the paraventricular hypothalamus: a powerful stimulant of feeding behavior. *Proc Natl Acad Sci USA* 1985; **82**: 3940–3943.
- 152 Day DE, Keen-Rhinehart E, Bartness TJ. Role of NPY and its receptor subtypes in foraging, food hoarding, and food intake by Siberian hamsters. *Am J Physiol Regul Integr Comp Physiol* 2005; **289**: R29–R36.
- 153 King BM. The rise, fall, and resurrection of the ventromedial hypothalamus in the regulation of feeding behavior and body weight. *Physiol Behav* 2006; **87**: 221–244.
- 154 Burdakov D, Luckman SM, Verkhatsky A. Glucose-sensing neurons of the hypothalamus. *Philos Trans R Soc Lond B Biol Sci* 2005; **360**: 2227–2235.
- 155 Levin BE, Govek EK, Dunn-Meynell AA. Reduced glucose-induced neuronal activation in the hypothalamus of diet-induced obese rats. *Brain Res* 1998; **808**: 317–319.
- 156 Bartness TJ, Kay Song C, Shi H, Bowers RR, Foster MT. Brain-adipose tissue cross talk. *Proc Nutr Soc* 2005; **64**: 53–64.
- 157 Wren AM, Small CJ, Abbott CR, Jethwa PH, Kennedy AR, Murphy KG, Stanley SA, Zollner AN, Ghatei MA, Bloom SR. Hypothalamic actions of neuromedin U. *Endocrinology* 2002; **143**: 4227–4234.
- 158 Sutton RE, Koob GF, Le Moal M, Rivier J, Vale W. Corticotropin releasing factor produces behavioural activation in rats. *Nature* 1982; **297**: 331–333.
- 159 Bowers RR, Festuccia WT, Song CK, Shi H, Migliorini RH, Bartness TJ. Sympathetic innervation of white adipose tissue and its regulation of fat cell number. *Am J Physiol Regul Integr Comp Physiol* 2004; **286**: R1167–R1175.
- 160 Graham ES, Turnbull Y, Fotheringham P, Nilaweera K, Mercer JG, Morgan PJ, Barrett P. Neuromedin U and neuromedin U receptor-2 expression in the mouse and rat hypothalamus: effects of nutritional status. *J Neurochem* 2003; **87**: 1165–1173.
- 161 Jethwa PH, Small CJ, Smith KL, Seth A, Darch SJ, Abbott CR, Murphy KG, Todd JF, Ghatei MA, Bloom SR. Neuromedin U has a physiological role in the regulation of food intake and partially mediates the effects of leptin. *Am J Physiol Endocrinol Metab* 2005; **289**: E301–E305.
- 162 Chambers J, Ames RS, Bergsma D, Muir A, Fitzgerald LR, Hervieu G, Dytko GM, Foley JJ, Martin J, Liu WS, Park J, Ellis C, Ganguly S, Konchar S, Cluderay J, Leslie R, Wilson S, Sarau HM. Melanin-concentrating hormone is the cognate ligand for the orphan G-protein-coupled receptor SLC-1. *Nature* 1999; **400**: 261–265.
- 163 Qu D, Ludwig DS, Gammeltoft S, Piper M, Pellemounter MA, Cullen MJ, Mathes WF, Przypek R, Kanarek R, Maratos-Flier E. A role for melanin-concentrating hormone in the central regulation of feeding behaviour. *Nature* 1996; **380**: 243–247.
- 164 Shimada M, Tritos NA, Lowell BB, Flier JS, Maratos-Flier E. Mice lacking melanin-concentrating hormone are hypophagic and lean. *Nature* 1998; **396**: 670–674.
- 165 Marsh DJ, Weingarh DT, Novi DE, Chen HY, Trumbauer ME, Chen AS, Guan XM, Jiang MM, Feng Y, Camacho RE, Shen Z, Frazier EG, Yu H, Metzger JM, Kuca SJ, Shearman LP, Gopal-Truter S, MacNeil DJ, Strack AM, MacIntyre DE, Van der Ploeg LH, Qian S. Melanin-concentrating hormone 1 receptor-deficient mice are lean, hyperactive, and hyperphagic and have altered metabolism. *Proc Natl Acad Sci USA* 2002; **99**: 3240–3245.
- 166 Chen Y, Hu C, Hsu CK, Zhang Q, Bi C, Asnicar M, Hsiung HM, Fox N, Sliker LJ, Yang DD, Heiman ML, Shi Y. Targeted disruption of the melanin-concentrating hormone receptor-1 results in hyperphagia and resistance to diet-induced obesity. *Endocrinology* 2002; **143**: 2469–2477.
- 167 Lalonde R, Qian S. Exploratory activity, motor coordination, and spatial learning in Mchr1 knockout mice. *Behav Brain Res* 2007; **178**: 293–304.
- 168 Segal-Lieberman G, Bradley RL, Kokkotou E, Carlson M, Trombly DJ, Wang X, Bates S, Myers MG Jr, Flier JS, Maratos-Flier E. Melanin-concentrating hormone is a critical mediator of the leptin-deficient phenotype. *Proc Natl Acad Sci USA* 2003; **100**: 10085–10090.
- 169 Lechan RM, Fekete C. The TRH neuron: a hypothalamic integrator of energy metabolism. *Prog Brain Res* 2006; **153C**: 209–235.
- 170 Kalivas PW, Stanley D, Prange AJ Jr. Interaction between thyrotropin-releasing hormone and the mesolimbic dopamine system. *Neuropharmacology* 1987; **26**: 33–38.
- 171 Yarbrough GG. On the neuropharmacology of thyrotropin releasing hormone (TRH). *Prog Neurobiol* 1979; **12**: 291–312.
- 172 Ohata H, Shibasaki T. Effects of urocortin 2 and 3 on motor activity and food intake in rats. *Peptides* 2004; **25**: 1703–1709.
- 173 Kageyama H, Takenoya F, Kita T, Hori T, Guan JL, Shioda S. Galanin-like peptide in the brain: effects on feeding, energy metabolism and reproduction. *Regul Pept* 2005; **126**: 21–26.
- 174 Krasnow SM, Fraley GS, Schuh SM, Baumgartner JW, Clifton DK, Steiner RA. A role for galanin-like peptide in the integration of feeding, body weight regulation, and reproduction in the mouse. *Endocrinology* 2003; **144**: 813–822.
- 175 Kauffman AS, Buenzle J, Fraley GS, Rissman EF. Effects of galanin-like peptide (GALP) on locomotion, reproduction, and body weight in female and male mice. *Horm Behav* 2005; **48**: 141–151.
- 176 Jolicœur FB, Menard D, Fournier A, St-Pierre S. Structure-activity analysis of CGRP's neurobehavioral effects. *Ann NY Acad Sci* 1992; **657**: 155–163.

- 177 Krahn DD, Gosnell BA, Levine AS, Morley JE. Effects of calcitonin gene-related peptide on food intake. *Peptides* 1984; **5**: 861–864.
- 178 Sawada H, Yamaguchi H, Shimbara T, Toshinai K, Mondal MS, Date Y, Murakami N, Katafuchi T, Minamino N, Nunoi H, Nakazato M. Central effects of calcitonin receptor-stimulating peptide-1 on energy homeostasis in rats. *Endocrinology* 2006; **147**: 2043–2050.
- 179 Kernie SG, Liebl DJ, Parada LF. BDNF regulates eating behavior and locomotor activity in mice. *EMBO J* 2000; **19**: 1290–1300.
- 180 Teske JA, Levine AS, Kuskowski M, Levine JA, Kotz CM. Elevated hypothalamic orexin signaling, sensitivity to orexin A and spontaneous physical activity in obesity resistant rats. *Am J Physiol Regul Integr Comp Physiol* 2006; **291**: R889–R899.
- 181 Saper CB. Staying awake for dinner: hypothalamic integration of sleep, feeding, and circadian rhythms. *Prog Brain Res* 2006; **153**: 243–252.
- 182 Yamanaka A, Sakurai T, Katsumoto T, Yanagisawa M, Goto K. Chronic intracerebroventricular administration of orexin-A to rats increases food intake in daytime, but has no effect on body weight. *Brain Res* 1999; **849**: 248–252.
- 183 Lubkin M, Stricker-Krongrad A. Independent feeding and metabolic actions of orexins in mice. *Biochem Biophys Res Commun* 1998; **253**: 241–245.
- 184 Kiwaki K, Kotz CM, Wang C, Lanningham-Foster L, Levine JA. Orexin A (hypocretin 1) injected into hypothalamic paraventricular nucleus and spontaneous physical activity in rats. *Am J Physiol Endocrinol Metab* 2004; **286**: E551–E559.
- 185 Hara J, Yanagisawa M, Sakurai T. Difference in obesity phenotype between orexin-knockout mice and orexin neuron-deficient mice with same genetic background and environmental conditions. *Neurosci Lett* 2005; **380**: 239–242.
- 186 Luckman SM, Lawrence CB. Anorectic brainstem peptides: more pieces to the puzzle. *Trends Endocrinol Metab* 2003; **14**: 60–65.
- 187 Woods SC, Benoit SC, Clegg DJ, Seeley RJ. Clinical endocrinology and metabolism. Regulation of energy homeostasis by peripheral signals. *Best Pract Res Clin Endocrinol Metab* 2004; **18**: 497–515.
- 188 Woods SC. Gastrointestinal satiety signals I. An overview of gastrointestinal signals that influence food intake. *Am J Physiol Gastrointest Liver Physiol* 2004; **286**: G7–G13.
- 189 Katsuura G, Itoh S. Effects of cholecystokinin and vasoactive intestinal peptide on locomotion and rearing in neofrontal decorticated rats. *Prog Clin Biol Res* 1985; **192**: 147–150.
- 190 Joyner K, Smith GP, Gibbs J. Abdominal vagotomy decreases the satiating potency of CCK-8 in sham and real feeding. *Am J Physiol* 1993; **264**: R912–R916.
- 191 Rushing PA. Central amylin signaling and the regulation of energy homeostasis. *Curr Pharm Des* 2003; **9**: 819–825.
- 192 Lutz TA, Senn M, Althaus J, Del Prete E, Ehrensperger F, Scharer E. Lesion of the area postrema/nucleus of the solitary tract (AP/NTS) attenuates the anorectic effects of amylin and calcitonin gene-related peptide (CGRP) in rats. *Peptides* 1998; **19**: 309–317.
- 193 Baldo BA, Kelley AE. Amylin infusion into rat nucleus accumbens potently depresses motor activity and ingestive behavior. *Am J Physiol Regul Integr Comp Physiol* 2001; **281**: R1232–R1242.
- 194 Schulz DW, Kalivas PW, Nemeroff CB, Prange AJ Jr. Bombesin-induced locomotor hyperactivity: evaluation of the involvement of the mesolimbic dopamine system. *Brain Res* 1984; **304**: 377–382.
- 195 Ladenheim EE, Ritter RC. Low-dose fourth ventricular bombesin selectively suppresses food intake. *Am J Physiol* 1988; **255**: R988–R993.
- 196 Le Moal M, Simon H. Mesocorticolimbic dopaminergic network: functional and regulatory roles. *Physiol Rev* 1991; **71**: 155–234.
- 197 Broberger C. Brain regulation of food intake and appetite: molecules and networks. *J Intern Med* 2005; **258**: 301–327.
- 198 Thorpe AJ, Kotz CM. Orexin A in the nucleus accumbens stimulates feeding and locomotor activity. *Brain Res* 2005; **1050**: 156–162.
- 199 Abizaid A, Liu ZW, Andrews ZB, Shanabrough M, Borok E, Elsworth JD, Roth RH, Sleeman MW, Picciotto MR, Tschöp MH, Gao XB, Horvath TL. Ghrelin modulates the activity and synaptic input organization of mid-brain dopamine neurons while promoting appetite. *J Clin Invest* 2006; **116**: 3229–3239.
- 200 Belcheva I, Belcheva S, Petkov VV, Petkov VD. Asymmetry in behavioral responses to cholecystokinin microinjected into rat nucleus accumbens and amygdala. *Neuropharmacology* 1994; **33**: 995–1002.
- 201 Fulton S, Pissios P, Manchon RP, Stiles L, Frank L, Pothos EN, Maratos-Flier E, Flier JS. Leptin regulation of the mesoaccumbens dopamine pathway. *Neuron* 2006; **51**: 811–822.
- 202 Hommel JD, Trinko R, Sears RM, Georgescu D, Liu ZW, Gao XB, Thurmon JJ, Marinelli M, DiLeone RJ. Leptin receptor signaling in midbrain dopamine neurons regulates feeding. *Neuron* 2006; **51**: 801–810.
- 203 Rotzinger S, Bush DE, Vaccarino FJ. Cholecystokinin modulation of mesolimbic dopamine function: regulation of motivated behaviour. *Pharmacol Toxicol* 2002; **91**: 404–413.
- 204 Klusa V, Svirskis S, Opmane B, Muceniece R, Skujins A, Mutulis F, Wikberg JE, Schioth HB. Evaluation of behavioural effects of neural melanocortin receptor antagonists injected ICV and in VTA in rats. *Neuropeptides* 1998; **32**: 573–580.
- 205 Contarino A, Delle F, Koob GF, Smith GW, Lee KF, Vale WW, Gold LH. Dissociation of locomotor activation and suppression of food intake induced by CRF in CRFR1-deficient mice. *Endocrinology* 2000; **141**: 2698–2702.
- 206 Merali Z, McIntosh J, Anisman H. Role of bombesin-related peptides in the control of food intake. *Neuropeptides* 1999; **33**: 376–386.
- 207 Kotz CM, Briggs JE, Grace MK, Levine AS, Billington CJ. Divergence of the feeding and thermogenic pathways influenced by NPY in the hypothalamic PVN of the rat. *Am J Physiol* 1998; **275**: R471–R477.
- 208 Jones BE. Arousal systems. *Front Biosci* 2003; **8**: s438–s451.
- 209 Peyron C, Tighe DK, van den Pol AN, de Lecea L, Heller HC, Sutcliffe JG, Kilduff TS. Neurons containing hypocretin (orexin) project to multiple neuronal systems. *J Neurosci* 1998; **18**: 9996–10015.
- 210 Zahm DS, Brog JS. On the significance of subterritories in the 'accumbens' part of the rat ventral striatum. *Neuroscience* 1992; **50**: 751–767.
- 211 Hooks MS, Kalivas PW. The role of mesoaccumbens – pallidal circuitry in novelty-induced behavioral activation. *Neuroscience* 1995; **64**: 587–597.
- 212 Morgan MA, Schulkin J, Pfaff DW. Estrogens and non-reproductive behaviors related to activity and fear. *Neurosci Biobehav Rev* 2004; **28**: 55–63.
- 213 Sell SL, Dillon AM, Cunningham KA, Thomas ML. Estrous cycle influence on individual differences in the response to novelty and cocaine in female rats. *Behav Brain Res* 2005; **161**: 69–74.
- 214 Miller CC, Holmes PV, Edwards GL. Area postrema lesions elevate NPY levels and decrease anxiety-related behavior in rats. *Physiol Behav* 2002; **77**: 135–140.
- 215 Theander-Carrillo C, Wiedmer P, Cettour-Rose P, Nogueiras R, Perez-Tilve D, Pfluger P, Castaneda TR, Muzzin P, Schürmann A, Szanto I, Tschöp MH, Rohner-Jeanrenaud F. Ghrelin action in the brain controls adipocyte metabolism. *J Clin Invest* 2006; **116**: 1983–1993.
- 216 Kiyashchenko LI, Mileykovskiy BY, Lai YY, Siegel JM. Increased and decreased muscle tone with orexin (hypocretin) microinjections in the locus coeruleus and pontine inhibitory area. *J Neurophysiol* 2001; **85**: 2008–2016.
- 217 van den Top M, Nolan MF, Lee K, Richardson PJ, Buijs RM, Davies CH, Spanswick D. Orexins induce increased excitability and synchronization of rat sympathetic preganglionic neurones. *J Physiol* 2003; **549**: 809–821.

- 218 Kannan H, Hayashida Y, Yamashita H. Increase in sympathetic outflow by paraventricular nucleus stimulation in awake rats. *Am J Physiol* 1989; **256**: R1325–R1330.
- 219 Bartness TJ, Bamshad M. Innervation of mammalian white adipose tissue: implications for the regulation of total body fat. *Am J Physiol* 1998; **275**: R1399–R1411.
- 220 Haque MS, Minokoshi Y, Hamai M, Iwai M, Horiuchi M, Shimazu T. Role of the sympathetic nervous system and insulin in enhancing glucose uptake in peripheral tissues after intrahypothalamic injection of leptin in rats. *Diabetes* 1999; **48**: 1706–1712.
- 221 Rosenbaum M, Vandenborne K, Goldsmith R, Simoneau JA, Heymsfield S, Joannise DR, Hirsch J, Murphy E, Matthews D, Segal KR, Leibel RL. Effects of experimental weight perturbation on skeletal muscle work efficiency in human subjects. *Am J Physiol Regul Integr Comp Physiol* 2003; **285**: R183–R192.
- 222 Aronne LJ, Mackintosh R, Rosenbaum M, Leibel RL, Hirsch J. Autonomic nervous system activity in weight gain and weight loss. *Am J Physiol* 1995; **269**: R222–R225.
- 223 Tentolouris N, Liatis S, Katsilambros N. Sympathetic system activity in obesity and metabolic syndrome. *Ann NY Acad Sci* 2006; **1083**: 129–152.
- 224 Rosenbaum M, Murphy EM, Heymsfield SB, Matthews DE, Leibel RL. Low dose leptin administration reverses effects of sustained weight-reduction on energy expenditure and circulating concentrations of thyroid hormones. *J Clin Endocrinol Metab* 2002; **87**: 2391–2394.
- 225 Rosenbaum M, Goldsmith R, Bloomfield D, Magnano A, Weimer L, Heymsfield S, Gallagher D, Mayer L, Murphy E, Leibel RL. Low-dose leptin reverses skeletal muscle, autonomic, and neuroendocrine adaptations to maintenance of reduced weight. *J Clin Invest* 2005; **115**: 3579–3586.
- 226 Vidal-Puig A, Rosenbaum M, Considine RC, Leibel RL, Dohm GL, Lowell BB. Effects of obesity and stable weight reduction on UCP2 and UCP3 gene expression in humans. *Obes Res* 1999; **7**: 133–140.
- 227 Silva JE. Thyroid hormone control of thermogenesis and energy balance. *Thyroid* 1995; **5**: 481–492.
- 228 Biron R, Burger A, Chinet A, Clausen T, Dubois-Ferriere R. Thyroid hormones and the energetics of active sodium-potassium transport in mammalian skeletal muscles. *J Physiol* 1979; **297**: 47–60.
- 229 Kaciuba-Uscilko H, Jessen C, Feistkorn G, Brzezinska Z. Work performance, thermoregulation and muscle metabolism in thyroidectomized goats (*Capra hircus*). *Comp Biochem Physiol A* 1987; **87**: 915–921.
- 230 Chang GQ, Karatayev O, Davydova Z, Leibowitz SF. Circulating triglycerides impact on orexigenic peptides and neuronal activity in hypothalamus. *Endocrinology* 2004; **145**: 3904–3912.
- 231 Yamanaka A, Beuckmann CT, Willie JT, Hara J, Tsujino N, Mieda M, Tominaga M, Yagami K, Sugiyama F, Goto K, Yanagisawa M, Sakurai T. Hypothalamic orexin neurons regulate arousal according to energy balance in mice. *Neuron* 2003; **38**: 701–713.
- 232 Yokota M, Ozaki Y, Sakamoto F, Yamada S, Saito J, Fujihara H, Ueta Y. Fos expression in CRF-containing neurons in the rat paraventricular nucleus after central administration of neuromedin U. *Stress* 2004; **7**: 109–112.
- 233 Proulx K, Richard D, Walker CD. Leptin regulates appetite-related neuropeptides in the hypothalamus of developing rats without affecting food intake. *Endocrinology* 2002; **143**: 4683–4692.
- 234 Heinrichs SC, Cole BJ, Pich EM, Menzaghi F, Koob GF, Hauger RL. Endogenous corticotropin-releasing factor modulates feeding induced by neuropeptide Y or a tail-pinch stressor. *Peptides* 1992; **13**: 879–884.
- 235 Heinrichs SC, Koob GF. Corticotropin-releasing factor in brain: a role in activation, arousal, and affect regulation. *J Pharmacol Exp Ther* 2004; **311**: 427–440.
- 236 Winsky-Sommerer R, Yamanaka A, Diano S, Borok E, Roberts AJ, Sakurai T, Kilduff TS, Horvath TL, de Lecea L. Interaction between the corticotropin-releasing factor system and hypocretins (orexins): a novel circuit mediating stress response. *J Neurosci* 2004; **24**: 11439–11448.
- 237 Reyes BA, Valentino RJ, Xu G, Van Bockstaele EJ. Hypothalamic projections to locus coeruleus neurons in rat brain. *Eur J Neurosci* 2005; **22**: 93–106.
- 238 Cerri M, Morrison SF. Corticotropin releasing factor increases in brown adipose tissue thermogenesis and heart rate through dorsomedial hypothalamus and medullary raphe pallidus. *Neuroscience* 2006; **140**: 711–721.
- 239 Bushnik T, Bielajew C, Konkle AT, Merali Z. Influence of bombesin on threshold for feeding and reward in the rat. *Acta Neurobiol Exp (Wars)* 1999; **59**: 295–302.
- 240 Dauge V, Dor A, Feger J, Roques BP. The behavioral effects of CCK8 injected into the medial nucleus accumbens are dependent on the motivational state of the rat. *Eur J Pharmacol* 1989; **163**: 25–32.
- 241 Borgland SL, Taha SA, Sarti F, Fields HL, Bonci A. Orexin A in the VTA is critical for the induction of synaptic plasticity and behavioral sensitization to cocaine. *Neuron* 2006; **49**: 589–601.
- 242 Harris GC, Wimmer M, Aston-Jones G. A role for lateral hypothalamic orexin neurons in reward seeking. *Nature* 2005; **437**: 556–559.
- 243 Wunderlich GR, Rotzinger S, Bush DE, DeSousa NJ, Vaccarino FJ. Cholecystokinin modulation of locomotor behavior in rats is sensitized by chronic amphetamine and chronic restraint stress exposure. *Brain Res* 2004; **1001**: 95–107.
- 244 Holmes A, Picciotto MR. Galanin: a novel therapeutic target for depression, anxiety disorders and drug addiction? *CNS Neurol Disord Drug Targets* 2006; **5**: 225–232.
- 245 Koob GF, Heinrichs SC. A role for corticotropin releasing factor and urocortin in behavioral responses to stressors. *Brain Res* 1999; **848**: 141–152.
- 246 Heinrichs SC, Joppa M. Dissociation of arousal-like from anxiogenic-like actions of brain corticotropin-releasing factor receptor ligands in rats. *Behav Brain Res* 2001; **122**: 43–50.
- 247 Kamara K, Eskay R, Castonguay T. High-fat diets and stress responsiveness. *Physiol Behav* 1998; **64**: 1–6.
- 248 Legendre A, Harris RB. Exaggerated response to mild stress in rats fed high-fat diet. *Am J Physiol Regul Integr Comp Physiol* 2006; **291**: R1288–R1294.
- 249 Michel C, Dunn-Meynell A, Levin BE. Reduced brain CRH and GR mRNA expression precedes obesity in juvenile rats bred for diet-induced obesity. *Behav Brain Res* 2004; **154**: 511–517.
- 250 Levin BE, Richard D, Michel C, Servatius R. Differential stress responsiveness in diet-induced obese and resistant rats. *Am J Physiol Regul Integr Comp Physiol* 2000; **279**: R1357–R1364.
- 251 Taheri S, Lin L, Austin D, Young T, Mignot E. Short sleep duration is associated with reduced leptin, elevated ghrelin, and increased body mass index. *PLoS Med* 2004; **1**: E62.
- 252 Taheri S. The link between short sleep duration and obesity: we should recommend more sleep to prevent obesity. *Arch Dis Child* 2006; **91**: 881–884.
- 253 Sakurai T, Amemiya A, Ishii M, Matsuzaki I, Chemelli RM, Tanaka H, Williams SC, Richardson JA, Kozlowski GP, Wilson S, Arch JR, Buckingham RE, Haynes AC, Carr SA, Annan RS, McNulty DE, Liu WS, Terrett JA, Elshourbagy NA, Bergsma DJ, Yanagisawa M. Orexins and orexin receptors: a family of hypothalamic neuropeptides and G protein-coupled receptors that regulate feeding behavior. *Cell* 1998; **92**: 573–585.
- 254 de Lecea L, Kilduff TS, Peyron C, Gao X, Foye PE, Danielson PE, Fukuhara C, Battenberg EL, Gautvik VT, Bartlett FS, Frankel WN, van

- den Pol AN, Bloom FE, Gautvik KM, Sutcliffe JG. The hypocretins: hypothalamus-specific peptides with neuroexcitatory activity. *Proc Natl Acad Sci USA* 1998; **95**: 322–327.
- 255 Kotz CM. Integration of feeding and spontaneous physical activity: role for orexin. *Physiol Behav* 2006; **88**: 294–301.
- 256 Szentirmai E, Kapas L, Krueger JM. Ghrelin microinjection into forebrain sites induces wakefulness and feeding in rats. *Am J Physiol Regul Integr Comp Physiol* 2007; **292**: R575–R585.
- 257 Szentirmai E, Krueger JM. Obestatin alters sleep in rats. *Neurosci Lett* 2006; **404**: 222–226.
- 258 Laposky AD, Shelton J, Bass J, Dugovic C, Perrino N, Turek FW. Altered sleep regulation in leptin-deficient mice. *Am J Physiol Regul Integr Comp Physiol* 2006; **290**: R894–R903.
- 259 Prinz P. Sleep, appetite, and obesity – what is the link? *PLoS Med* 2004; **1**: E61.
- 260 Sakurai T. Roles of orexin/hypocretin in regulation of sleep/wakefulness and energy homeostasis. *Sleep Med Rev* 2005; **9**: 231–241.
- 261 Sloth B, Holst JJ, Flint A, Gregersen NT, Astrup A. Effects of PYY1–36 and PYY3–36 on appetite, energy intake, energy expenditure, glucose and fat metabolism in obese and lean subjects. *Am J Physiol Endocrinol Metab* 2007; **292**: E1062–E1068.
- 262 McMahon LR, Wellman PJ. PVN infusion of GLP-1-(7–36) amide suppresses feeding but does not induce aversion or alter locomotion in rats. *Am J Physiol* 1998; **274**: R23–R29.
- 263 Paxinos G, Watson C. *The Rat Brain in Stereotaxic Coordinates*, 5th edn. New York, NY: Elsevier Academic Press, 2005.