MINIREVIEW

Neurochemical Alterations During Age-Related Anorexia (44398)

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Abstract. **Unexplained weight loss during the latter stages of aging is commonly preceded by a spontaneous diminution in food intake. Multiple etiologies of age-related anorexia in humans, ranging from social isolation to impaired gastrointestinal function, have been proposed. The observation of this phenomenon in older laboratory animals suggests that physiological changes play a significant causal role. A continually expanding body of information on the neurochemical control of food intake supports a contribution of altered neurochemistry to dysregulated feeding behavior. This review provides an update on the relationship between declining food intake during advanced age and physiological (specifically neurochemical) function. The complexity of the control of food intake as well as the variety of investigative methods used in this field of study render the identification of definitive causes difficult. Evidence presented here is evaluated and possible etiologic factors are suggested.**

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Aging, especially in its advanced stages, is associated with declines in food intake and body weight in both humans and laboratory animals (1–3). Longitudinal studies, including the third National Health and Nutrition Examination Survey (NHANES III), the New Mexico Aging Process Study, and the Baltimore Longitudinal Study of Aging show a progressive decrease in caloric intake with age (4–6). Data from NHANES III are summarized in Table I. This decline in energy intake is more than offset by reduced energy expenditure during middle age, and the adiposity percentage actually rises through the fifth and sixth decades of life (3). After age 70, energy balance often becomes negative, and losses in lean body mass and body fat occur (7,8). Dietary intake studies involving elderly subjects frequently report inadequate caloric intake (9–12), and unintentional weight loss has been described in up to 35% of aged community-dwellers (13) and 70% of nursing home residents (14). This decrease in eating, known as the "anorexia of aging," appears to develop without definitive cause in otherwise healthy elderly persons and has been proposed to constitute a prelude to "natural" death (15).

The concept that normal aging is associated with alterations in the homeostatic mechanisms influencing hunger, satiety, and food intake is supported by several studies. Roberts (16) demonstrated that older (mean age 68 years) versus younger (22.7 years) men were less able to appropriately adjust their energy intake in response to dietary manipulation (i.e., the elderly did not return to energy balance after under- or over-feeding as did the younger subjects). Consistent with this finding, Rolls (17) used a preloading method to show that elderly men (60–84 years) compensated less precisely for changes in caloric intake than did younger men (18–35 years). The older subjects in this study also reported less hunger and greater fullness compared to the younger group. Similarly, desire to eat after

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Table I. Mean Daily Kilocalorie (kcal)[a] Intake by Age Group (Third National Health and Nutrition Examination Survey, Phase 1, 1988–1991)

Age group (yrs)	Sample size	Daily kcal intake	
		No.	(SE[b])
20–29	1,682	2484	(± 44.4)
30–39	1,526	2372	(± 43.4)
40–49	1,228	2146	(± 44.5)
50–59	929	1967	(± 30.7)
60–69	1,106	1822	(± 39.0)
70–79	851	1624	(± 25.3)
≥80	609	1484	(± 27.4)

[a] Kilocalories derived from consumption of foods and beverages.
[b] Standard error.

a test meal is diminished more in aged (70–84-year-old) versus younger (23–50-year-old) men and women (18).

Although the phenomenon of age-related anorexia may be a physiologically "normal" manifestation of the aging process, it often results in undernutrition and secondary problems such as sarcopenia (19), impaired immunity (20), and physical frailty (21), all of which can substantially impact quality of life. Despite continued research in clinical and laboratory settings, the etiology of the anorexia of aging has been difficult to ascertain because declines in food intake are often subtle and not recognized until confounding illnesses develop. The purpose of this review is to evaluate the evidence for a physiological (specifically neurochemical) contribution to the anorexia of aging and to suggest possible etiologic factor(s). The influence of psychosocial state is beyond the scope of this paper and is addressed elsewhere (2, 3, 22, 23). The term senescence will be used to refer to the terminal stage of aging, when the rate of functional deterioration increases significantly and death follows shortly thereafter. Prior to addressing individual neurochemicals and briefly, chemosensory and gastric function, a synopsis of the control of food intake will be discussed. Mean or median life spans are indicated for those rodent strains for which sufficient longevity data exist. Concluding remarks include a description of the approach to research that our laboratories have found effective for elucidating potential processes underlying age-related anorexia.

Overview of Neurochemical Control of Food Intake

Food intake involves an extensive array of effectors and proposed mechanisms that are not completely understood. Neural and chemical signals originating in the brain and periphery are thought to interact within the brain and result in feeding behavior and regulatory energy expenditure that maintain energy balance. Although early lesion experiments identified the importance of the hypothalamus in the control of food intake (24), other brain areas, such as the nucleus of the solitary tract (25–27), nigrostriatal tract (28), and limbic system (27) have since become recognized for their role in feeding. As investigations continue to elucidate the central integration of sensory input from the vagus nerve, endocrine messages from peripheral tissues, information on nutrient metabolite concentrations, and neurotransmitter signals from various brain regions, considerable data have accumulated regarding a number of chemical modulators of hunger and satiety. Many neurochemicals have been implicated in the control of food intake. These include neuropeptide Y (NPY) (29), neurotensin (30), cholecystokinin (CCK) (31), leptin (32), galanin (33), the orexins (34), melanocyte-stimulating hormone (35), melanin-concentrating hormone (36), agouti-related peptide (37), and corticotropin-releasing hormone (38). However, few of these factors have been studied in relation to the aging process, and still fewer have been investigated for their potential role in the age-related decline in food intake. Included in this latter group are NPY, leptin, CCK, and endogenous opioids. An examination of the roles of each of these in the control of food intake and what, if any, age-related changes in their function are observed is helpful in narrowing the list of candidate effectors in the anorexia of aging.

Neuropeptide Y: Neurotransmitter Pathways

One function of NPY is to facilitate energy balance by mediating food intake–stimulating signals within a complex interconnected neural pathway (39). Neuroanatomical mapping studies have shown that NPY-producing neurons are located in the brain stem, cortex, and arcuate nucleus (ARC) of the basal hypothalamus and innervate various hypothalamic sites, including the paraventricular nucleus (PVN) (40). Experiments indicate that NPY synthesis in the ARC and release in the PVN increase when feeding occurs either normally or after food restriction (41, 42). Among the signals that regulate NPY synthesis and release are insulin, leptin, and glucocorticoids (43–47). Neuropeptide Y exerts its feeding-stimulatory effects most potently through Y_1 and Y_5 receptors, which belong to the family of G protein-coupled receptors (48, 49). Intrahypothalamic injection of NPY (particularly into the paraventricular and perifornical areas) elicits a marked increase in eating (50–53), whereas immunoneutralization of hypothalamic NPY or blocking its receptor suppresses the hyperphagic response to food deprivation (29, 54).

Neuropeptide Y also acts within the brain to influence peripheral metabolic systems in ways that favor energy storage. That is, NPY modulates activity of the sympathetic nervous system and hypothalamic-pituitary axis to stimulate lipogenic enzymes in adipose tissue and the liver, enhance glucose uptake at adipose, and reduce brown adipose tissue thermogenesis (46, 55, 56). Because of its powerful effects to reestablish energy balance after reductions in food intake and body weight, NPY has been investigated as a potential factor in age-related anorexia. Due to the invasive experimental procedures currently required to determine hypothalamic NPY concentrations, research has relied mostly upon animal models rather than human subjects. Rats provide an

excellent model for investigating the anorexia of aging because they undergo a similar syndrome of age-related decreases in food intake and body weight (1, 57–62).

Effects of Age on Feeding Responses to Neuropeptide Y. One of the first investigations demonstrating an age-related decline in hypothalamic NPY function that was associated with reduced food intake was conducted by Pich *et al.* (63). Using two groups of male Sprague-Dawley rats (3-month-old and 24-month-old), and two feeding states (24-hr food-deprived and satiated), the effects of intraparaventricular NPY injection were measured. The feeding response elicited by NPY at 30, 90, and 240 min postinjection was attenuated in the older compared to younger satiated and food-deprived rats, but no age difference was seen at 22 hr after injection. Lower levels of NPY immunoreactivity within the PVN were found in older versus younger animals. Pich's group proposed that the impaired sensitivity to exogenous NPY and the reduction of NPY in the same nucleus in aged rats suggested that NPY deficiency in the PVN may be one factor responsible for age-related anorexia. However, a limitation in interpreting results from studies such as these that use only two age groups, one of which may still be in its rapid growth state, is the inability to determine if differences between the groups develop early in adulthood or arise abruptly upon advanced age. Comparing responsiveness to NPY between animals in late adulthood and senescence would help clarify the role of NPY in age-related anorexia.

Additional investigations have sought to determine an effect of age on NPY-related functions. Although initially focusing on reproductive senescence and the role of decreased positive testicular feedback on NPY synthesis, the experimental results of Gruenewald *et al.* (64) led them to propose a connection between decreased NPY levels and the age-associated decline in food intake. More recently, these investigators tested the hypothesis that the NPY-mediated ability to increase food intake and maintain body weight after short-term fasting is impaired with aging (58). Fasting and fed levels of preproNPY (ppNPY) mRNA were compared in 3-, 12-, and 24-month-old male Brown Norway (BN) rats. Whereas fasting was associated with increases in ppNPY mRNA in all groups, this response was attenuated with progressive age, with significant differences demonstrated between the two older groups and the youngest rats. The 24-month-old rats exhibited significantly slower rates of body weight recovery to prefasting levels during refeeding compared to the 12- and 3-month-old rats. A causal relationship was suggested between the age-associated reduction in ppNPY mRNA response to fasting and the blunted food intake response to fasting. The authors addressed the possibility that lower energy requirements in aged versus younger rats may have contributed to the reduced levels of NPY in the former: less food intake would be required to maintain stable body weight. The significantly slower recovery of body weight after fasting in the oldest versus middle-aged versus young rats, and the inability of many of the 24-month-old animals to fully recover prefasting weight suggest an impairment in homeostasis, rather than reduced caloric needs with aging. Yet the question of whether or not reduced NPY gene transcription is related to the anorexia of advanced age is difficult to answer from this study since the mean life span, a commonly used benchmark for senescence, of the BN rat (29 months) (65, 66) is considerably greater than the oldest experimental group. Moreover, the reduction in ppNPY mRNA began in middle-aged rats and did not show significant change thereafter. It is likely that the results of this investigation are more applicable to developmental aspects of aging rather than to senescence.

Effects of Age on Levels of Neuropeptide Y and Related Neuromodulators. A number of other researchers have demonstrated age-related changes in NPY concentrations within the rat brain. Kowalski *et al.* (67) found that while changes in corticotropin-releasing hormone, methionine (met)-enkephalin, and β-endorphin concentrations varied with age depending on brain area, NPY levels were more consistently decreased throughout the brain (frontal cortex, hypothalamus, and hippocampus) in male Wistar rats (26 < 18 < 4-months-old). These investigators concluded that the NPY pepidergic system is one of the most vulnerable to aging and that physiological functions that involve NPY, such as the sensori-motor axis and cognition, are preferentially impaired during aging. Kowalski *et al.* suggested that, given the close relationship between NPY and dopamine (DA), as well as their synaptic and biochemical similarities (68, 69), the alterations in NPY may be linked to the decreased dopaminergic activity seen in the aged rat brain (70–72). This proposed correlation is supported by studies showing that intrahypothalamic NPY administration results in perturbations in the turnover of dopamine (73, 74). Similar to Kowalski's group, Cha *et al.* (75) found severe loss of cerebral cortex NPY neurons in aged (20–29-month-old) compared to young (4–6-month-old) Sprague-Dawley rats (gender not specified) and suggested a connection with age-related impairments in dopaminergic activity.

The role and function of dopamine in the central control of food intake continues to be defined (76). Yet common age-related alterations in dopamine and NPY could be expected to have a profound impact on feeding. If dopamine suppresses feeding, as studies have shown (76–78), the release of inhibitory constraint on dopamine activity exerted by NPY (secondary to reduced levels of NPY) may contribute to age-related anorexia. This would be particularly important if the effects of age on the dopaminergic system are relatively mild compared to those on the NPY pathway.

Neuropeptide Y has been found to coexist with norepinephrine (NE) in neurons of the hypothalamus, brainstem (79), and tracts of the locus coeruleus innervating the hypothalamus (80). Various experiments have demonstrated a relationship between NPY and α_2-adrenoceptors in these brain areas, including the alteration of NPY action by α_2-

adrenoceptor antagonists (81, 82) and the modulation of NE activity and noradrenoreceptor number by NPY (83–86). These findings have led to the suggestion that NPY acts in concert with or through NE to influence food intake (79, 80, 85). Huguet *et al.* (87) found a reduction in hypothalamic NPY concentrations accompanied by decreased density of α_2-adrenoceptors coupled with increased binding affinity of ^{3}H-rauwolscine in 34- versus 4-month-old Wistar rats (gender not specified). No difference was found in NE concentrations between the younger and older rats. Considering the data demonstrating that both NPY and α_2-adrenoceptor agonists/antagonists act through a G protein–adenylate cyclase system (49), Huguet's group proposed that age-related alterations in functional activities that involve the actions of NPY and NE, such as feeding, reflect disorganization in signal transduction (87). Because the functions of NPY include the presynaptic modulation of release of neurotransmitters, including catecholamines, age-related changes in NPY may result in modified synaptic transmission, especially catecholaminergic transmission. The homeostatic control that NPY and catecholamines exert upon food intake may thus be altered.

Serotonin (5-HT) is another neurotransmitter that appears to be integrated into the NPY network of food intake control. Serotonin administration directly into the PVN suppresses feeding with a selective reduction in carbohydrate consumption (88, 89), an action contrary to NPY's effects (90). Neurons containing NPY have close anatomical associations with neurons containing 5-HT, and a functional interaction has been demonstrated (91, 92). Dryden *et al.* (93) treated rats with acute and chronic administrations of methysergide, a 5-HT receptor antagonist, and found significant elevations of NPY in the ARC and PVN with corresponding increased food intake. It was concluded that NPYergic and serotoninergic innervations in the hypothalamus interact to regulate food intake, and that increased NPY release may mediate the hyperphagic effect of 5-HT receptor blockade. A logical extension of these results is that a physiological (versus pharmacological) reduction of 5-HT receptor signaling, secondary to impaired receptor integrity or reduced ligand concentrations, would allow NPY levels to rise and induce food intake. Yet the anorexia of aging occurs during a period in life when 5-HT signaling is decreased. Meal-induced serotonin release within the PVN of the hypothalamus is reduced in old (12-month-old) versus young (4-month-old) lean and obese male Zucker rats (12-month mean life span for obese Zucker) (71). Additionally, Slotkin *et al.* (94) found that 5-HT transporter expression was attenuated in aged rat brain, and Woods *et al.* (72) reported increased 5-HT turnover in various brain areas of aged (24-month-old) male Fischer 344 (F344) rats (mean life span of 24–26 months) (66). These experiments suggest that aging is associated with decreased serotonergic signal transduction. The reciprocal increase in hypothalamic NPY levels that occurs in young rats when 5-HT signal is low may be attenuated in older rats and result in impaired feeding.

Not all investigators have observed age-related changes in NPY function. Li *et al.* (95) found no significant differences in hypothalamic NPY mRNA levels in male F344×BN rats (median life span of 33 months) aged 3, 24, and 31 months. Yet the fact that the two older groups displayed similar levels of food intake per $kg^{.67}$ body weight, adiposity, leptin mRNA, and body weight indicates that the oldest animals were not exhibiting signs of age-related anorexia. In providing possible explanations for the disagreement between their findings and those of Gruenewald and colleagues, Li's group noted the differences in the times at which the rats were sacrificed. Since NPY mRNA displays a circadian rhythm (41, 43), and aging is known to be associated with altered rhythmicity (96–98), age-related changes in NPY mRNA levels may be observable only during specific points in the rhythm (e.g., the predark peak). Such considerations should also be applied when comparing data from experiments measuring other peptides that are synthesized and/or released according to a circadian rhythm (e.g., leptin, CCK, opioids) (99–101).

Results from research on other forms of anorexia can offer insights into the potential role of NPY in the decline in food intake during advanced age. Using mice homozygous for the anorexia mutation (*anx*), Broberger *et al.* (102) investigated if hypothalamic neurochemical changes could be found in this genetic model of starvation. Although immunohistochemistry revealed no significant differences in concentrations of CCK, galanin, and 5-HT between *anx/anx* and wild-type mice, significant alterations in NPY were seen: high levels of NPY were found within perikarya in the ARC of the *anx/anx* mouse compared with low levels in the control mice. In contrast, NPY within nerve terminals of the PVN, ARC, and other hypothalamic nuclei showed a concomitant decrease in concentration in *anx/anx* mice compared with control animals. Interestingly, *in situ* hybridization demonstrated no change between the groups in the levels of ppNPY mRNA within the ARC. The authors concluded that NPY synthesis in the ARC is normal in the *anx/anx* mouse, but that impaired axonal transport of NPY causes an accumulation in perikarya and a deficit in terminals. This then prevents the generation of a signal to increase food intake in response to negative energy balance and results in death by starvation. Given the selective disruption in the NPY neuropathway in this model for anorexia, and the absence of change in levels of other modulators of food intake (including leptin), it is reasonable to believe that alterations in NPY may play a role in anorexia near the end of life.

It is important to consider the contrary findings of apparently normal food intake and energy balance in NPY knockout (–/–) mice (103, 104). In contrast to the acute intervention studies described earlier, NPY knockout mice exhibit fasting-induced hyperphagia and leptin-induced hypophagia. These observations imply that the current understanding of NPY's contribution to the control of food intake may be less than previously proposed, or that alternate path-

ways can effectively compensate for the absence of NPY. Targeted gene mutation has revealed that processes of gene transcription and intracellular signal transduction are able to substitute certain protein factors for others that are absent (105, 106). It should be recognized that this compensatory action is displayed by animals who have from conception lost the ability to synthesize the gene product. For vital systems, such as feeding and energy homeostasis, there are often redundant circuits that act as safeguards. An embryo could develop these substitute pathways to remain viable in terms of energy regulation. Perhaps in NPY –/– mice, the effectors of food intake that normally function in parallel with or downstream from NPY (e.g., NE, DA, and dynorphin) have developed the ability to respond directly to those signals that influence NPY. This adaptation may not be possible beyond the animal's developmental period, or even less so during advanced ages.

Although the preponderance of data provides evidence for an age-related decline in the NPY feeding system, a clear correlation between a significant loss in NPY function and anorexia near the end of life has yet to be elucidated. Critical to resolving this issue are future studies comparing endogenous NPY secretion and potency in animals before and after they have entered senescence, when food intake is significantly reduced.

Leptin

Leptin is secreted from adipose tissue and acts on the brain to inhibit feeding and stimulate thermogenesis (44). The leptin receptor (*Ob-R*) gene encodes at least five splice variants, including *Ob-Rb,* which is expressed at a high level in the hypothalamus (107). Most evidence suggests that at least some of the effects of leptin are mediated by inhibition of NPY gene transcription. Wang *et al.* (108) injected leptin into the lateral ventricle of fasted rats and noted inhibition of food intake accompanied by reduced NPY concentrations in the ARC, PVN, and dorsomedial hypothalamus. Similar findings of leptin's suppression of NPY levels have been reported by several investigators (109–112). When functional leptin or its *Ob-Rb* receptor is lacking, as in the obese (*ob/ob*) or diabetic (*db/db*) mouse, respectively, NPY signaling is enhanced (107, 113). Wang *et al.* (108) also showed that chronic intracerebroventricular NPY administration in rats resulted in elevated serum leptin levels, although independent effects of feeding (hyperphagia) and increased insulin levels on leptin were not determined.

Because circulating leptin levels are positively correlated with adiposity (114, 115), and the percentage of body fat tends to increase through middle age (95, 116, 117), an independent primary effect of age on serum leptin concentration has been investigated. At least two studies in humans demonstrated that after adjustment for body fat, a significant inverse relationship between serum leptin and age exists (118, 119). Experiments in humans and rodents have shown serum leptin concentrations to increase with age in parallel with body fat, and to fall with the decline in adiposity during advanced age (1, 117, 120). Findings from our laboratories indicate that near the end of their life spans, male and female F344 rats exhibit decreased serum leptin concentrations concomitant with lower carcass fat (121). Conversely, in studying the strength of the relationship between leptin and adiposity with advancing age, Moller's group (122) showed that beyond age 45 years, increased body fat is *not* paralleled by elevated serum leptin levels. These data, which led to the authors' suggestion that leptin deficiency results in obesity in middle-aged and older adults, could also indicate that the suppressed appetite near the end of life is not associated with elevated serum leptin levels. Similarly, Cederholm *et al.* (123) found no correlation between serum leptin and adiposity in malnourished elderly (mean age 74 years) patients with chronic nonmalignant illnesses. These investigators proposed that leptin levels were altered in the patients because their adipose stores had become depleted to such a degree that leptin secretion was impaired or, that their poor nutritional status had caused a lack of serum leptin carrier protein (124, 125).

The discrepancy among these results involving aging and leptin may reflect variation in the time of sample collection. Leptin secretion displays a circadian rhythm in humans and rodents that is entrained to meal timing (99, 109, 126, 127). Both the known and unknown (due to lack of specification) differences among the studies with respect to times of blood draw render comparisons difficult. Although the question of whether or not enhanced leptin potency contributes to age-related anorexia remains to be answered, the available data indicate that circulating leptin levels in older animals and humans are relatively low and presumably should allow NPY levels to rise and stimulate food intake. The absence of an appropriate increase in food intake in these subjects suggests dysregulation in the leptin-NPY feeding pathway. In conjunction with the suggested NPY investigations, it will be important to determine if sensitivity to leptin at the site of feeding control is altered in the anorectic elderly.

Cholecystokinin (CCK)

The satiety hormone CCK is thought to act centrally on the brain feeding system and peripherally by slowing gastric emptying and activating visceral sensory nerves (128). Protein and fat ingestion causes CCK to be secreted from the intestine as well as the hypothalamus, although the mechanisms of the latter are not fully understood (129, 130). Cholecystokinin also aids in coordinating digestion by causing the simultaneous contraction of the gallbladder and relaxation of the sphincter of Oddi to release bile into the proximal intestine and by stimulating pancreatic enzyme secretion (131). At least two types of CCK receptors are known to exist: CCK_A, which is predominantly in the gastrointestinal tract, and CCK_B, which is predominantly in the brain (128). A number of investigators have sought to reveal alterations in this relatively well-established modulator of

food intake in an attempt to explain changes in feeding. Theoretically, a greater level of or sensitivity to CCK may inhibit food intake, such as observed in the anorexia of aging. However, studies of the relationship of age to food intake are not in agreement regarding peripheral and central concentrations and potency of CCK in laboratory animals and humans. Basal and postprandial plasma CCK levels were similar in young (mean age 29 years) and aged (mean age 80 years) healthy subjects (132). Using cerebral cortex synaptosomes and perfused intestine to study central and peripheral CCK release, respectively, Miyasaka *et al.* (133) examined basal and stimulated CCK release in older (25–29-month-old) and younger (5–8-month-old) male and female Wistar rats and found no significant effect of age except a decreased CCK release in response to the highest dose of stimulant in the older males. In other experiments, Miyasaka *et al.* (134) and Ohta *et al.* (135) reported significantly lower levels of CCK mRNA in cerebral cortex and small intestine of older (24–29-month-old) versus younger (3–8-month-old) male, but not female, Wistar rats. This, along with the finding of higher CCK levels in the cerebral cortex, led the authors to conclude that aging impairs central and peripheral CCK release, resulting in accumulation of CCK within tissue and subsequent inhibition of further synthesis. The authors acknowledge that because middle-aged animals were not included in the study, it is possible that the changes in CCK release begin earlier in adulthood. Poston *et al.* (136) similarly found increased intestinal CCK concentrations in 3-year-old versus 1-year-old and 1-month-old guinea pigs, but these elevated levels were accompanied by decreased pancreatic and gallbladder CCK receptor sensitivity. Similarly, Khalil *et al.* (137) observed that comparable gallbladder contraction in response to fat ingestion in younger (22–42-year-old) and older (60–84-year-old) men and women was associated with significantly higher plasma CCK levels in the aged group, suggesting attenuated responsiveness to CCK. A comparable study by Masclee *et al.* (138) revealed corresponding results. These findings suggest an age-related enhancement, rather than impairment, in CCK release in conjunction with receptor downregulation. When comparing CCK fractions between elderly (65–86-year-old) men and women with idiopathic senile anorexia (in the absence of recognizable organic or mental disorder) and control subjects, Martinez *et al.* (139) found that the anorectics had higher plasma levels of one of two CCK prohormone products measured. However, no significant differences were observed in CCK measurements taken from cerebral spinal fluid. Although plasma CCK probably does not cross the blood-brain barrier to act on central nervous system receptors, it can act upon the vagus nerve that transmits signals from the periphery to brain neurons involved in the control of feeding (140). Given this, and experimental results showing that peripheral administration of CCK into rats suppresses food intake (141, 142), elevated plasma CCK could induce premature satiation.

Unlike the data supporting an age-related decrease in CCK potency, other studies demonstrate increased peripheral and central sensitivity to CCK in the aged. Voigt *et al.* (143) investigated the effects of intraperitoneal CCK on food intake under two different feeding conditions in younger (2-month-old) and older (23-month-old) male Wistar rats. In food-deprived animals, CCK reduced feeding similarly in both age groups, whereas under fixed (meal) feeding conditions, the highest dose of CCK significantly reduced food intake only in the oldest rats. An explanation offered by the authors for the age difference is that age-related deficits in learning and memory, which presumably are needed to adapt to a fixed feeding schedule, render the older rats more susceptible to the hypophagic effect of CCK. That an age effect was not observed in both experimental feeding conditions is consistent with the complexity of the control of food intake and suggests that mechanisms were able to compensate for enhanced sensitivity to CCK in these aged rats. Silver *et al.* (144) also demonstrated a greater sensitivity to CCK in 25- versus 8-month-old male C57BL/6Nnia mice (median life span of 27 months) (66). Younger mice ate significantly more than baseline whereas older mice showed a nonsignificant tendency toward enhanced feeding in response to intraperitoneal injection of an antagonist to peripheral CCK receptors.

These data do not show advanced age to be consistently associated with higher levels of and/or greater sensitivity to CCK. Under experimental conditions of chronic CCK administration, food intake and body weight are not significantly changed (145), apparently because the resulting reduction in meal size is compensated for by increased meal frequency (146). Although satiety factors such as CCK are powerful effectors of individual meal size, they are thought to have limited independent influence on long-term energy balance (147). Although we have begun experiments on the role of CCK potency in aged anorectic rats, these studies have not progressed enough to determine if CCK plays a significant role in the etiology of this anorexia.

Endogenous Opioids

The sensory pleasure response to food, particularly fats and sweets, is thought to be mediated, in part, by endogenous opiates, including dynorphin, β-endorphin, and the enkephalins (57, 148). Opioids enhance feeding by direct action on central nervous system structures that are known to contribute to the control of food intake (hypothalamus, amygdala, and nucleus accumbens) (149). Injection of opioids into the cerebroventricles of animals results in increased food intake, and blocking opioid receptors causes reduced feeding in animals and humans (150–152). Interactions occur between opioids and other neuropeptides (e.g., NPY) and neurotransmitters (e.g., DA) during the modulation of food intake behavior (153, 154).

It has been proposed that the anorexia of aging involves impairments in the opioid feeding system. Gosnell *et al.* (57) evaluated the feeding responses to naloxone (an opioid antagonist) and butorphanol (an opioid agonist) in F344 rats

aged 2, 12, 22, and 28 months. They found that low doses of naloxone resulted in decreased food intake in 2- and 12-month-old rats, but not the 22- and 28-month-old animals. Although the two older age groups responded similarly to all antagonist treatments, after injection of agonist, the 28-month-old animals ate significantly more compared to baseline than did the 22-month-old group. The role of opioids on long-term energy balance is obscured by studies showing that chronic opioid antagonist administration in rats has no effect on food intake or body weight within days of beginning treatment (155, 156). Evidence for an age-related decline in hypothalamic opioid concentrations is provided by Forman *et al.* (157), who reported decreased β-endorphin content in 19–23-month- versus 3–5-month-old male Sprague-Dawley rats, and by Dupont *et al.* (158), who demonstrated lower levels of enkephalins in male Sprague-Dawley rats aged 24–26 months compared to 4–5 months. Plasma and CSF β-endorphin concentrations are lower in humans with idiopathic senile anorexia (aged 70–86 years) compared to normal-weight controls (139). However, following treatment with megestrol acetate (an appetite stimulant) in the anorectic individuals, significant increases in CSF β-endorphin levels were observed without improvements in food intake or body weight.

Several investigators have found no significant age-related changes in opioid concentrations within brain areas associated with the control of feeding. Levels of hypothalamic met-enkephalin and β-endorphin were not significantly different among 3-, 8-, and 23-month-old male Sprague-Dawley rats (153). In male Sprague-Dawley rats of ages 3, 12, and 22 months, Wang *et al.* (159) examined the effect of aging and light/dark cycle on levels of brain neuropeptides and found no differences in hypothalamic concentrations of β-endorphin, leucine (leu)-enkephalin, or met-enkephalin with the exception of lower daytime met-enkephalin levels in the oldest rats. Additional reports exist indicating an absence of an age effect on levels of hypothalamic β-endorphin and met-enkephalin in male Sprague-Dawley rats beyond the age of sexual maturity (160–162). Furthermore, recent results from our laboratories provide indirect evidence for a functional feeding reward system during advanced aging. When offered a choice of a basal semipurified diet [65% total kcal carbohydrate (32.5% sucrose, 32.5% cornstarch); 22% fat; 13% protein] and a high-fat, high-sucrose (palatable) diet [51% total kcal fat; 36% carbohydrate (100% sucrose), 13% protein], older (25–29+ months) male F344 rats maintained a preference for the palatable diet throughtout their natural life spans (1). The contribution of opioid system dysfunction to the anorexia of aging remains to be clearly defined.

Chemosensory and Gastrointestinal Function

Because the actions of certain neurochemicals, such as opioids and CCK, involve the transmission of signals generated by oronasal and gastrointestinal stimuli, respectively, a brief discussion of these processes as they relate to feeding and aging is warranted. Substantial attention has been directed toward elucidating sensory and gastrointestinal signals that could explain reductions in appetite and body weight during advanced age. Diminutions in these systems have the potential to adversely affect energy balance by reducing food intake. Research has tended to focus upon age-related changes in olfaction, taste, and gastric function. Many early reports showing significant declines in sensory and gastric function with age used tissue from persons with unknown medical histories or patients with diagnosed pathologies (163–166); however, more recent investigations have attempted to exclude diseased subjects from study. Even so, in researching taste and smell, the methodologies employed include a large subjective component that could complicate direct comparisons of perceived sensations among various age groups.

Many investigations have demonstrated that the density and basic structure of taste buds and papillae are maintained in healthy old humans and animals (167–169). Whereas evidence supports the conclusion that taste and smell thresholds are elevated with age (170, 171), the impact of these deficits on food intake is uncertain. Changes in thresholds do not reliably predict changes in perception of more concentrated stimuli, such as those experienced during daily eating (172, 173). Furthermore, we are unaware of investigations that directly demonstrate a causal relationship between diminished sensory function, reduced hedonic response, and changes in feeding in older individuals (148, 174). Studies on independently living elderly are not consistent: Griep *et al.* (175) observed a correlation between lower energy intake and poor odor perception, whereas Duffy *et al.* (176) showed no impairment in appetite or food enjoyment as a result of significant olfactory losses. Even flavor enhancement of foods for elderly (average age 85 years) subjects with deficits in taste and smell does not improve food intake enough to affect body weight or body mass index (177). Until longitudinal data characterizing sensory changes and their relationship to food intake across the life span become available, there is insufficient evidence to support a major role of smell or taste deficits in contributing to anorexia at advanced ages.

The existence of age-related changes in gastric function is controversial, largely due to the confounding influence of atrophic gastritis, an acquired disease that is prevalent in the elderly (178). Atrophic gastritis results in hypochlorhydria or achlorhydria that impairs gastric emptying (179). Numerous studies showing reduced gastric secretion and emptying with age did not exclude the presence of atrophic gastritis or its causative bacterium, *Helicobacter pylori* (*H. pylori*) (165, 166, 180). At least two investigations using human subjects in whom gastric disease was strictly excluded demonstrated no age-related decrease in gastric acid secretion (181, 182). Interpreting the contrary findings of Clarkston *et al.* (18) and Horowitz *et al.* (183), showing gastric emptying rates to be slower in elderly versus younger humans, is not straightforward since neither study indicates that the ab-

sence of gastric disease was confirmed (e.g., by endoscopy or biopsy). In addition, Horowitz concluded that the changes in gastric emptying were small, within the normal range found in younger controls, and were unlikely to be of clinical significance.

Studies on aging rats have produced similarly inconsistent results. Smits *et al.* (184) compared gastrointestinal transit time in 3-, 12-, and 24-month-old male Wistar rats and demonstrated delayed gastric emptying in the oldest group. In contrast, McDougal *et al.* (185) and Varga (186) observed no significant age differences in rates of gastric emptying.

In general, there is a lack of conclusive data indicating that gastric secretion or emptying decreases significantly as a function of normal aging. Without such an association established, it is premature to implicate stomach dysfunction in age-related anorexia. Although a causal relationship has been suggested between the anorexia of aging and *H. pylori* infection (187), this represents a correlation of decreased food intake with age-associated disease rather than with normal aging processes.

Conclusions Regarding the Effects of Age on Control of Food Intake

Despite the caveats mentioned throughout this paper, there are general conclusions that can be drawn. The control of food intake is composed of an intricate network of many neurochemicals acting in serial and parallel fashion to coordinate feeding behavior and overall energy balance. Both short-term modulators, such as CCK and opioids, which are released and act within individual meals, and long-term modulators of energy intake and output, such as leptin and NPY, interact to maintain energy homeostasis. A disruption in the energy balance system, as seen in age-related anorexia, most likely reflects loss in function of more than one component. Nonetheless, as complex systems often employ a hierarchy where central effectors exert a greater degree of control compared to others, it is reasonable to suggest a contribution by a particular neurochemical to the anorexia of aging. A large body of evidence suggests that NPY plays an important role in the compensatory response to negative energy balance. Many factors (e.g., adipose stores, blood glucose concentration) mediated through different signaling molecules (e.g., leptin, insulin) influence NPY synthesis, and once NPY is released, it initiates a cascade of events that work to restore and maintain energy homeostasis. Age-related impairments are observed more consistently in NPY function than in other known mediators of feeding behavior. It would seem unlikely that changes could occur in this important component of the food intake system without causing overt effects. Furthermore, the changes in food intake observed in humans and animals of advanced age (e.g., decreased food intake due to smaller meal size) (1, 188) are consistent with a disruption in NPY prolongation of eating (189, 190). The progressive weight loss associated with age-related anorexia implicates dysfunction of an effector of long-term energy balance, such as NPY.

There remains much to be revealed about the underlying causes of declining food intake near the end of life. A more thorough understanding of this phenomenon can contribute to optimizing quality of life during its final stages. Clarifying the mechanisms of the anorexia of aging can also offer clues to the more comprehensive investigation of the aging process. However, a challenge to research on aging is to verify an index of biological age that can be used widely by gerontologists. Such a biomarker would provide a reliable predictor of mortality and therefore facilitate the direct comparison of research results. Because no biomarker has been broadly accepted, most experiments have relied on chronological age as an indicator of the extent of aging. This review illustrates the difficulties encountered when interpreting data from studies using chronological age. The following is a description of an animal model that our laboratories use in an attempt to overcome this problem.

New Animal Model for the Anorexia of Aging

Our laboratories study biological aging by examining changes in physiological function occurring near the end of life that are not strictly related to chronological age or specific pathology. Within an individual, age-related changes occur at different rates and manifest at different degrees of severity. During the early and middle periods of postmaturational aging, physiological systems undergo gradual alterations that generally do not result in dramatic changes in function. However, as death becomes imminent, compensatory mechanisms fail, and the rate of physiological deterioration rapidly increases. This terminal phase of aging, which we call senescence, is a major focus of our investigations.

The animal model used by our laboratories developed from our finding that among old F344 rats of the same chronological age, some were much more susceptible to cold exposure than others. We found that these more susceptible rats had entered a phase in which they were spontaneously losing weight (senescence). During acute cold exposure, the senescent rats developed severe hypothermia, whereas age-matched weight-stable and age-matched rats with comparable weight loss due to food restriction maintained relatively stable core body temperature (59). Further investigations demonstrated that senescent rats display marked alterations in circadian rhythmicity of body temperature (96) and significant changes in feeding behavior, including reduced food intake (1). The onset of senescence, which cannot be explained by specific disease, is followed by death within 2–6 weeks. The similarity in symptoms and duration (relative to life span) of senescence in rats and age-related anorexia in humans (191) has allowed us to use the senescent rat as a model to study the characteristics of and physiological events associated with decreased food intake near the end of life. Because energy balance (body weight and food intake), body temperature, and circadian rhythm are controlled, in part, by the hypothalamus (147,

Table II. Feeding Patterns of Old Rats

	Age period				
	Early weight-stable	Mid weight-stable	Late weight-stable	Immediate preweight-loss	Weight-loss
Grams eaten/day	14.94 ± 0.27^{a}	13.82 ± 0.29^{b}	13.59 ± 0.35^{b}	13.27 ± 0.29^{b}	10.97 ± 0.33^{c}
Meals/day	9.25 ± 0.33^{a}	9.24 ± 0.27^{a}	$9.90 \pm 0.30^{a,b}$	$9.91 \pm 0.32^{a,b}$	10.56 ± 0.45^{b}
Grams/meal	1.60 ± 0.04^{a}	1.49 ± 0.04^{a}	$1.39 \pm 0.03^{a,b}$	1.35 ± 0.04^{b}	1.04 ± 0.03^{c}
Meal duration (min)	19.17 ± 0.96^{a}	16.92 ± 0.85^{b}	15.16 ± 0.65^{b}	15.81 ± 0.74^{b}	11.92 ± 0.59^{c}

Note. Means ± standard error of feeding pattern variables of old (25–32.5-month-old; $n = 9$) male F344 rats across age periods. Within a row, values sharing superscripts do not differ significantly. Data adapted from Blanton, *et al.* (1).

192), the working hypothesis of our group implicates the hypothalamus in the onset of senescence. More specific to energy homeostasis, we propose that the anorexia of aging reflects altered hypothalamic function. Thus, as we investigate the possible mechanisms involved in the altered control of food intake during senescence, we have concentrated on neurochemical mediation.

Clues to the nature of the proposed neurochemical involvement in reduced food intake during advanced age have been provided by the examination of feeding patterns of senescent rats. In a recent study, we monitored food intake continuously in 24-month-old F344 rats until 7 days after the onset of senescence (1). Regardless of age at death (25.5–32.5 months), all rats ate significantly smaller meals of shorter duration during this initial stage of senescence compared to their period of weight stability (presenescence) (Table II). These findings, combined with the fact that meal frequency did not differ between senescence and presenescence, is suggestive of early satiation in the senescent rats. Furthermore, the consumption of shorter, smaller meals despite progressively declining body weight (which averaged 5.2% in these senescent rats) implicates dysfunction in a mediator of energy balance that acts by attenuating satiety. Both CCK and NPY are strong candidates for this failure in energy homeostasis of the senescent rats. Whereas leptin works to reduce meal size (32, 193), we have shown that circulating leptin is not elevated in senescent rats (1, 121). We have also demonstrated a hedonic response to palatable food in senescent rats, which implies a functional opioid system. Ongoing research is designed to better describe the neurochemical changes that occur during senescence. With the resulting data, we seek to propose more definitively a mechanism underlying the anorexia of aging.

1. Blanton CA, Geitzen DW, Griffey SM, Horwitz BA, McDonald RB, Murtagh-Mark C. Meal patterns associated with the age-related decline in food intake in the Fischer 344 rat. Am J Physiol **275:**R1494–R1502, 1998.
2. Fischer J, Johnson MA. Low body weight and weight loss in the aged. J Am Diet Assoc **90:**1697–1706, 1990.
3. Morley JE. Anorexia of aging: Physiologic and pathologic. Am J Clin Nutr **66:**760–773, 1997.
4. Anonymous. Daily dietary fat and total food energy intakes. Third National Health and Nutrition Examination Survey, phase III. 1988–1991. Morbidity and Mortality Weekly Rep **43:**116–125, 1994.
5. Hallfrisch J, Muller D, Drinkwater D. Continuing trends in men: The Baltimore Longitudinal Study of Aging (1961–1987). J Gerontol **45:**M186–M191, 1990.
6. Koehler KM. The New Mexico Aging Process Study. Nutr Rev **8**(Suppl):S34–S37, 1994.
7. Chumlea WC, Garry PJ, Hunt WC, Rhyne RL. Distributions of serial changes in stature and weight in a healthy elderly population. Hum Biol **60:**917–925, 1988.
8. Steen B, Isaksson B, Svanborg A. Body composition at 70 and 75 years of age: A longitudinal population study. J Clin Exp Gerontol **1:**185–192, 1979.
9. McGandy RB, Russell RM, Hartz SC, Jacob RA, Tannenbaum S, Peters H, Sahyoun N, Otradovec CL. Nutritional status survey of healthy noninstitutionalized elderly: Energy and nutrient intakes from three-day diet records and nutrient supplements. Nutr Res **6:**785, 1986.
10. Mowe M, Bohmer T, Kindt E. Reduced nutritional status in an elderly population (>70 yr) is probable before disease and possibly contributes to the development of disease. Am J Clin Nutr **59:**317–324, 1994.
11. O'Hanlon P, Kohrs MB. Dietary studies of older Americans. Am J Clin Nutr **31:**1257, 1978.
12. Posner BEM, Smigelski CG, Krachenfels MM. Dietary characteristics and nutrient intake in an urban homebound population. J Am Diet Assoc **87:**452, 1987.
13. Launer LJ, Harris T, Rumpel C, Madans J. Body mass index, weight change, and risk of mobility disability in middle-aged and older women: The epidemiologic follow-up study of NHANES I. JAMA **271:**1093–1098, 1994.
14. Silver AJ, Morley JE, Strome SS, Jones D, Vickers L. Nutritional status in an academic nursing home. J Am Geriatr Soc **36:**487, 1988.
15. McCue JD. The naturalness of dying. JAMA **273:**1039–1043, 1995.
16. Roberts S. Effects of aging on energy requirements and the control of food intake in men. J Gerontol **50:**101–106, 1995.
17. Rolls B, Dimeo K, Shide D. Age-related impairments in the regulation of food intake. Am J Clin Nutr **62:**923–931, 1995.
18. Clarkston WK, Pantano MM, Morley JE, Horowitz M, Littlefield JM, Burton FR. Evidence for the anorexia of aging: Gastrointestinal transit and hunger in healthy elderly vs. young adults. Am J Physiol **272:**R243–R248, 1997.
19. Evans WJ, Campbell WW. Sarcopenia and age-related changes in body composition and functional capacity. J Nutr **123:**465–468, 1993.
20. Thompson JS, Robbins J, Cooper JK. Nutritional and immune function in the geriatric population. Clin Geriatr Med **3:**309, 1987.
21. Folstein MF, Folstein SE, McHugh PR. Mini-mental state. J Psychiatr Res **12:**189–198, 1975.
22. Egbert A. The Dwindles: Failure to thrive in older patients. Nutr Rev **54:**S25–S30, 1996.
23. Marcus E-L, Berry EM. Refusal to eat in the elderly. Nutr Rev **56:**163–171, 1998.
24. Anand BK, Brobeck JR. Hypothalamic control of food intake in rats and cats. Yale J Biol Med **24:**123–140, 1951.
25. Hyde TM, Miselis RR. Effects of area postrema/caudal medial nucleus of the solitary tract lesions on food intake and body weight. Am J Physiol **244:**R577–R587, 1983.
26. Ricardo JA, Koh ET. Anatomical evidence of direct projections from the nucleus of the solitary tract to the hypothalamus, amygdala and other forebrain structures in the rat. Brain Res **153:**1–26, 1978.

27. Sawchenko PE. Central connections of the sensory and motor nuclei of the vagus nerve. J Auton Nerv System **9:**13–26, 1983.
28. Fibiger HC, Zis AP, McGreer EG. Feeding and drinking deficits after 6-hydroxytryptamine administration in the rat: Similarities to the lateral hypothalamic syndrome. Brain Res **55:**135–148, 1973.
29. Dube MG, Xu B, Crowley WR, Kalra PS, Kalra SP. Evidence that neuropeptide Y is a physiological signal for normal food intake. Brain Res **646:**341–344, 1994.
30. Rovere C, Viale A, Nahon J, Kitabgi P. Impaired processing of brain proneurotensin and promelanin-concentrating hormone in obese fat/fat mice. Endocrinology **137:**2954–2958, 1996.
31. Antin J, Gibbs J, Holt J, Young RC, Smith GP. Cholecystokinin elicits the complete behavioral sequence of satiety in rats. J Comp Physiol Psychol **89:**784–790, 1975.
32. Flynn MC, Scott TR, Pritchard TC, Plata-Salaman CR. Mode of action of OB protein, (leptin) on feeding. Am J Physiol **275:**R174–R179, 1998.
33. Schick R, Samsami S, Zimmermann J. Effect of galanin on food intake in rats: Involvement of lateral and ventromedial hypothalamic sites. Am J Physiol **264:**R355–R361, 1993.
34. Sakurai T, Amemiya A, Ishii M, Matsuzaki I, Chumelli RM, Tanaka H, Williams SC, Richardson JA, Kozlowski GP, Wilson S. Orexins and Orexin receptors: A family of hypothalamic neuropeptides and G protein-coupled receptors that regulate feeding behavior. Cell **92:**573–585, 1998.
35. Fan W, Boston BA, Desterson RA, Hruby VJ, Cone RD. Role of melanocortinergic neurons in feeding and agouti obesity syndrome. Nature **385:**165–168, 1997.
36. Prese F, Sorokovsky I, Max J, Nicolaidis S, Nahon J. Melanin-concentrating hormone is a potent anorectic peptide regulated by food deprivation and glucopenia in the rat. Neurosci **71:**735–745, 1996.
37. Ollmann M, Wilson BD, Yang YK, Kerns JA, Chen Y, Gantz I, Barsh GS. Antagonism of central melanocortin receptors *in vitro* and *in vivo* by agouti-related protein. Science **278:**135–138, 1997.
38. Arase K, York DA, Shimizu H, Shargill N, Bray GA. Effects of corticotropin-releasing factor on food intake and brown adipose tissue thermogenesis in rats. Am J Physiol **235:**E255–E259, 1988.
39. Kalra SP, Kalra PS. Is neuropeptide Y (NPY) a naturally occurring appetite transducer? Curr Op Endocrinol Diabetes **3:**157–163, 1996.
40. Chronwall BM, DiMaggio DA, Massari VJ, Pickel VM, Ruggiecro DA, O'Donohue TL. The anatomy of neuropeptide-Y-containing neurons in rat brain. Neuroscience **15:**1159–1181, 1985.
41. Kalra SP, Dube MG, Sahu A, Phelps CP, Kalra P. Neuropeptide Y secretion increases in the paraventricular nucleus in association with increased appetite for food. Proc Natl Acad Sci U S A **88:**10931–10935, 1991.
42. Sahu A, Kalra PS, Kalra SP. Food deprivation and ingestion induce reciprocal changes in neuropeptide Y concentrations in the paraventricular nucleus. Peptides **9:**83–86, 1988.
43. Akabayashi A, Watanabe Y, Wahlestedt R, McEwan BS, Paez X, Leibowitz SF. Hypothalamic neuropeptide Y, its gene expression and receptor activity: Relation to circulating corticosterone in adrenalectomised rats. Brain Res **665:**201–212, 1994.
44. Mistry A, Swich A, Romse D. Leptin rapidly lowers food intake and elevates metabolic rates in lean and *ob/ob* mice. J Nutr **127:**2065–2072, 1997.
45. Schwartz MW, Sipols AJ, Marks JL, Sanacora G, White JD, Scheurnik A, Kahn SE, Baskin DG. Inhibition of hypothalamic neuropeptide Y gene expression by insulin. Endocrinology **130:**3608–3616, 1992.
46. Schwartz M, Dallman M, Woods S. Hypothalamic response to starvation: Implications for the study of wasting disorders. Am J Physiol **269:**R949–R957, 1995.
47. Strack AM, Sebastian RJ, Schwartz MW, Dallman MF. Glucocorticoids and insulin: Reciprocal signals for energy balance. Am J Physiol **268:**R142–R149, 1995.
48. Hu Y, Bloomquist BT, Cornfield LJ, DeCarr LB, Flores-Riveros JR, Friedman L, Jiang P, Lewis-Higgins L, Sadlowski Y, Schaifer J, Velazquez N, McCaleb ML. Identification of a novel hypothalamic neuropeptide Y receptor associated with feeding behavior. J Biol Chem **271:**26315–26319, 1996.
49. Michel M, Lewejohann K, Farke W, Bischoff A, Feth F, Rascher W. Regulation of NPY/NPY Y1 receptor/G protein system in rat brain cortex. Am J Physiol **268:**R192–R200, 1995.
50. Jolicoeur FB, Bouali SM, Fournier A, St-Pierre S. Mapping of hypothalamic sites involved in the effects of NPY on body temperature and food intake. Brain Res Bull **36:**125–129, 1995.
51. Kalra SP, Dube MG, Kalra PS. Continuous intraventricular infusion of neuropeptide Y evokes episodic food intake in satiated female rats: Effects of adrenalectomy and cholecystokinin. Peptides **9:**723–728, 1988.
52. Morley JE, Levine AS, Gosnell BA, Kneip J, Grace M. Effect of neuropeptide Y on ingestive behaviors in the rats. Am J Physiol **252:**R599–R609, 1987.
53. Stanley BG. Neuropeptide Y in multiple hypothalamic sites controls eating behavior, endocrine, and autonomic systems for energy balance. In: Colmers WF, Wahlestedt C, Ed. The Biology of Neuropeptide Y and Related Peptides. Totowa, NJ: Humana, pp457–509, 1993.
54. Stanley BG, Magdalin W, Seirafi A, Nguyen MM, Leibowitz SF. Evidence for neuropeptide Y mediation of eating produced by food deprivation and for a variant of the Y_1 receptor mediating this peptide's effect. Peptides **13:**581–587, 1992.
55. Billington CJ, Briggs JE, Grace M. Effects of intracerebroventricular injection of neuropeptide Y on energy metabolism. Am J Physiol **260:**R321–R327, 1991.
56. Egawa M, Yoshimatsu H, Bray GA. Neuropeptide Y suppresses sympathetic activity to interscapular brown adipose tissue in rats. Am J Physiol **260:**R328–R334, 1991.
57. Gosnell BA, Levine AS, Morley JE. The effects of aging on opioid modulation of feeding in rats. Life Sci **32:**2793–2799, 1983.
58. Gruenewald DA, Marck BT, Matsumoto AM. Fasting-induced increases in food intake and neuropeptide Y gene expression are attenuated in aging male Brown Norway rats. Endocrinology **137:**4460–4467, 1996.
59. McDonald RB, Florez-Duquet M, Murtagh-Mark C, Horwitz BA. Relationship between cold-induced thermoregulation and spontaneous rapid body weight loss of aging F344 rats. Am J Physiol **271:**R1115–R1122, 1996.
60. Peng MT, Jiang MJ, Hsu HK. Changes in running-wheel activity, eating and drinking and their day/night distributions throughout the life span of the rat. J Gerontol **35:**339–347, 1980.
61. Peng MT, Kang M. Circadian rhythms and patterns of running-wheel activity, feeding and drinking behaviors of old male rats. Physiol Behav **33:**615–620, 1984.
62. Veyrat-Durebex C, Alliot J. Changes in pattern of macronutrient intake during aging in male and female rats. Physiol Behav **62:**1273–1278, 1997.
63. Pich EM, Messori B, Zoli M, Ferraguti F, Marrama P, Biagini G, Fuxe K, Agnati LF. Feeding and drinking responses to neuropeptide Y injections in the paraventricular hypothalamic nucleus of aged rats. Brain Res **575:**265–271, 1992.
64. Gruenewald DA, Naai MA, Marck BT, Matsumoto AM. Age-related decrease in neuropeptide Y gene expression in the arcuate nucleus of the male rat brain is independent of testicular feedback. Endocrinology **134:**2383–2389, 1994.
65. Burek JD, Hollander CF. Incidence patterns of spontaneous tumors in BN/Bi rats. J Natl Cancer Inst **58:**99–105, 1977.
66. Schneider EL, Rowe JW. Handbook of the Biology of Aging. San Diego, Ca: Academic Press, p8, 1996.
67. Kowalski C, Micheau J, Corder R, Gaillard R, Conte-Devolx B. Age-related changes in cortico-releasing factor, somatostatin, neuropeptide Y, methionine enkephalin and β-endorphin in specific rat brain areas. Brain Res **582:**38–46, 1992.
68. Kerkerian L, Bosler O. Striatal neuropeptide Y neurons are under influence of the nigrostriatal dopaminergic pathway: Immunohistochemical evidence. Neurosci Lett **66:**106–112, 1986.
69. Kerkerian L, Salin P, Nieoullon A. Pharmacological characterization of dopaminergic influence on expression of neuropeptide Y immunoreactivity by rat striatal neurons. Neuroscience **26:**809–817, 1988.
70. Himi T, Cao M, Mori N. Reduced expression of the molecular markers of dopaminergic neuronal atrophy in the aging rat brain. J Gerontol **50:**B193–200, 1995.
71. Lemierre S, Rouch C, Nicolaidis S, Orosco M. Combined effect of obesity and aging on feeding-induced monoamine release in the rostromedial hypothalamus of the Zucker rat. Int J Obes Relat Metab Disord **22:**993–999, 1998.
72. Woods JM, Druse MJ. Effects of chronic ethanol consumption and aging on dopamine, serotonin, and metabolites. J Neurochem **66:**2168–2178, 1996.

73. Gillard ER, Dang DQ, Stanley BG. Evidence that neuropeptide Y and dopamine in the perifornical hypothalamus interact antagonistically in the control of food intake. Brain Res **628:**128–136, 1993.
74. Myers RD, Lankford MF, Roscoe AK. Neuropeptide Y perfused in the preoptic area of rats shifts extracellular efflux of dopamine, norepinephrine, and serotonin during hypothermia and feeding. Neurochem Res **21:**637–648, 1996.
75. Cha C, Young I, Lee E. Age-related changes of VIP, NPY and somatostatin-immunoreactive neurons in the cerebral cortex of aged rats. Brain Res **753:**235–244, 1997.
76. Meguid MM, Yang Z, Laviano A. Meal size and number: Relationship to dopamine levels in the ventromedial hypothalamic nucleus. Am J Physiol **272:**R1925–R1930, 1997.
77. Meguid MM, Yang ZJ, Koseki M. Eating-induced rise in LHA-dopamine correlates with meal size in normal and bulbectomized rats. Brain Res Bull **36:**487–490, 1995.
78. Yang ZJ, Meguid MM, Chai JK, Chen C, Oler A. Bilateral hypothalamic dopamine infusion in male Zucker rat suppresses feeding due to reduced meal size. Pharmacol Biochem Behav **58:**631–635, 1997.
79. Blundell J. Pharmacological approaches to appetite suppression. Trends Physiol Sci **12:**147–157, 1991.
80. Lee MC, Schiffman SS, Pappas TN. Role of neuropeptides in the regulation of feeding behavior: A review of cholecystokinin, bombesin, neuropeptide Y, and galanin. Neurosci Biobehav Rev **18:**313–323, 1994.
81. Lambert PD, Wilding JP. A role for neuropeptide Y, dynorphin, and noradrenaline in the central control of food intake after food deprivation. Endocrinology **133:**29–32, 1993.
82. Stanley BG, Leibowitz SF. Neuropeptide Y: Stimulation of feeding and drinking by injections into the paraventricular nucleus. Life Sci **33:**2635–2642, 1984.
83. Agnati L, Fuxe K. Neuropeptide Y *in vivo* selectively increases the number of α_2-adrenergic binding sites in membranes of the medulla oblongata of the rat. Acta Physiol Scand **118:**293–295, 1983.
84. Matos FF, Guss V, Korpinen C. Effects of neuropeptide Y (NPY) and [D-Trp32] NPY on monoamine and metabolite levels in dialysates from rat hypothalamus during feeding behavior. Neuropeptides **30:**391–398, 1996.
85. Myers RD, Lankford MF, Paez X. Norepinephrine, dopamine, and 5-HT release from perfused hypothalamus of the rat during feeding induced by neuropeptide Y. Neurochem Res **17:**1123–1132, 1992.
86. Shimizu H, Bray GA. Effects of neuropeptide Y on norepinephrine and serotonin metabolism in rat hypothalamus *in vivo*. Brain Res Bull **22:**945–950, 1989.
87. Huguet F, Comoy E, Piriou A, Bohuon C. Age-related changes of noradrenergic-neuropeptide Y (NPY) interactions in rat brain: Norepinephrine, NPY levels, and α-adrenoceptors. Brain Res **625:**256–260, 1993.
88. Dourish C. 5-HT receptor subtypes and feeding behavior. Adv Biosci **85:**179–202, 1992.
89. Leibowitz SF, Weiss GF, Shor-Posner G. Hypothalamic serotonin: Pharmacological, biochemical, and behavioral analyses of its feeding-suppressive action. Clin Neuropharmacol **11:**S51–S71, 1988.
90. Stanley BG, Daniel DR, Chen AS, Leibowitz SF. Paraventricular nucleus injection of peptide YY and neuropeptide Y preferentially enhance carbohydrate ingestion. Peptides **6:**1205–1211, 1985.
91. Rogers P, McKibbin PE, Williams G. Acute fenfluramine administration reduces neuropeptide Y concentrations in specific hypothalamic regions of the rat: Possible implications for the anorectic effect of fenfluramine. Peptides **12:**251–255, 1991.
92. Smialowska M, Legutko B. Influence of imipramine on neuropeptide Y immunoreactivity in the rat brain. Neuroscience **41:**767–771, 1991.
93. Dryden S, McCarthy HD, Malabu UH, Ware M, Williams G. Increased neuropeptide Y concentrations in specific hypothalamic nuclei of the rat following treatment with methysergide: Evidence that NPY may mediate serotonin's effects on food intake. Peptides **14:**791–796, 1993.
94. Slotkin TA, McCook EC, Ritchie JC, Carroll BJ, Seidler FJ. Serotonin transporter expression in rat brain regions and blood platelets: Aging and glucocorticoid effects. Biol Psychiatry **41:**172–183, 1997.
95. Li H, Matheny M, Turner N, Scarpace PJ. Aging and fasting regulation of leptin and hypothalamic neuropeptide Y gene expression. Am J Physiol **275:**E405–E411, 1998.
96. McDonald RB, Hoban-Higgins TM, Ruhe RC, Fuller CA, Horwitz BA. Alterations in endogenous circadian rhythm of core temperature in senescent Fischer 344 rats. Am J Physiol **276:**R824–R830, 1999.
97. Myers BL, Badia P. Changes in circadian rhythms and sleep quality with aging: Mechanisms and interventions. Neurosci Biobehav Rev **19**(4):553–571, 1995.
98. Satinoff E. Patterns of circadian body temperature rhythms in aged rats. Clin Exp Pharmacol Physiol **2:**135–140, 1998.
99. Kousta E, Chrisoulidou A, Lawrence NJ, al-Shoumer KA, Parker KH, McCarthy MI, Johnston DG. The circadian rhythm of leptin is preserved in growth hormone deficient hypopituitary adults. Clin Endocrinol **48:**685–690, 1998.
100. Lindow SW, Newham A, Hendricks MS, Thompson JW, van der Spuy ZM. The 24-hour rhythm of oxytocin and β-endorphin secretion in human pregnancy. Clin Endocrinol **45:**443–446, 1996.
101. Schade R, Vick K, Ott T, Sohr R, Pfister C, Bellach J, Golor G, Lemmer B. Circadian rhythms of dopamine and cholecystokinin in nucleus accumbens and striatum of rats: Influence of dopaminergic stimulation. Chronobiol Int **12:**87–99, 1995.
102. Broberger C, Johansen J, Schalling M, Hokfelt T. Hypothalamic neurohistochemistry of the murine anorexia (*anx/anx*) mutation: Altered processing of neuropeptide Y in the arcuate nucleus. J Comp Neurol **387:**124–135, 1997.
103. Erickson JC, Clegg KE, Palmiter RD. Sensitivity to leptin and susceptibility to seizures of mice lacking neuropeptide Y. Nature **381:**415–418, 1996.
104. Erickson JC, Ahima RS, Hollopeter G, Flier JS, Palmiter RD. Endocrine function of neuropeptide Y knockout mice. Regul Pept **70:**199–202, 1997.
105. Araki E, Lipes MA, Patti ME, Bruning JC, Haag B III, Johnson RS, Kahn CR. Alternative pathway of insulin signalling in mice with targeted disruption of the IRS-1 gene. Nature **372:**186–189, 1994.
106. Hummler E, Cole TJ, Blendy JA, Ganss R, Aguzzi A, Schmid W, Beerman F, Schutz G. Targeted mutation of the CREB gene: Compensation within the CREB/ATF family of transcription factors. Proc Natl Acad Sci U S A **91:**5647–5651, 1994.
107. Lee G, Proenca P, Montez J. Abnormal splicing of the leptin receptor in diabetic mice. Nature **379:**632–635, 1996.
108. Wang Q, Bing C, Al-Barazanji K, Mossakowaska DE, Wang XM, McBay DL, Neville WA, Taddayon M, Pickavance L, Dryden S. Interactions between leptin and hypothalamic neuropeptide Y neurons in the control of food intake and energy homeostasis in the rat. Diabetes **46:**335–341, 1997.
109. Ahima RS, Prabakaran D, Mantzoros C, Qu D, Lowell B, Maratos-Flier E, Flier JS. Role of leptin in the neuroendocrine response to fasting. Nature **382:**250–252, 1996.
110. Cusin I, Rohner-Jeanrenaud F, Stricker-Krongrad A, Jeanrenaud B. The weight-reducing effect of an intracerebroventricular bolus injection of leptin in genetically obese *fa/fa* rats: Reduced sensitivity compared with lean animals. Diabetes **45:**1446–1450, 1996.
111. Sahu A. Evidence suggesting that galanin, melanin-concentrating hormone, neurotensin, proopiomelanocortin and neuropeptide Y are targets of leptin signaling in the hypothalamus. Endocrinology **139:**795–798, 1998.
112. Schwartz MW, Baskin DG, Bukowshi TR, Kuijper JL, Fuster D, Lasser G, Prunkard DE, Porte D, Woods SC, Seeley RJ, Weigle DS. Specificity of leptin action on elevated blood glucose and hypothalamic neuropeptide Y gene expression in *ob/ob* mice. Diabetes **45:**531–535, 1996.
113. Campfield LA, Smith FJ. Overview: Neurobiology of OB protein (leptin). Proc Nutr Soc **57:**429–440, 1998.
114. Havel PJ, Kasim-Karakas S, Mueller W, Johnson PR, Gingerich RL, Stern JS. Relationship of plasma leptin to plasma insulin and adiposity in normal weight and overweight women: Effects of dietary fat content and sustained weight loss. J Clin Endocrinol Metab **81:**4406–4413, 1996.
115. Li H, Matheny M, Turner N, Scarpace P. Leptin gene expression increases with age independent of increasing adiposity in rats. Diabetes **46:**2035–2039, 1997.
116. Elahi D, McAloon Dyke M, Andres R. Aging, fat metabolism, and adiposity. In: Masoro EJ, Ed. Handbook of Physiology: Aging. New York: Oxford University Press, pp147–170, 1995.
117. Perry HM, Morley JE, Horowitz M, Kaiser FE, Miller DK, Willert G. Body composition and age in African-American and Caucasian

women: Relationship to plasma leptin levels. Metab Clin Exp **46:**1399–1405, 1997.
118. Ostlund RE, Yang JW, Klein S, Gingerich R. Relation between plasma leptin concentration and body fat, gender, diet, age, and metabolic covariates. J Clin Endocrinol Metab **81:**3909–3913, 1996.
119. Ryan AS, Elahi D. The effects of acute hyperglycemia and hyperinsulinemia on plasma leptin levels: Its relationships with body fat, visceral adiposity, and age in women. J Clin Endocrinol Metab **81:**4433–4438, 1996.
120. Ahren B, Mansson S, Gingerich RL, Havel PJ. Regulation of plasma leptin in mice: Influence of age, high-fat diet, and fasting. Am J Physiol **273:**R113–R120, 1997.
121. Gabaldon A, McDonald R, Horwitz B. Gender differences in the relationship of neuropeptide Y (NPY) to serum leptin and adiposity in senescent F344 rats. FASEB J (in press).
122. Moller N, O'Brien P, Sreekumaran Nair K. Disruption of the relationship between fat content and leptin levels with aging in humans. J Clin Endocrinol Metab **83:**931–934, 1998.
123. Cederholm T, Arner P, Palmblad J. Low circulating leptin levels in protein-energy malnourished chronically ill elderly patients. J Intern Med **97:**377–382, 1997.
124. Hale L, Becker GW, Bowsher RR. Evidence of free and bound leptin in human circulation: Studies in lean and obese subjects and during short-term fasting. J Clin Invest **98:**1277–1282, 1996.
125. Houseknecht KL, Mantzoros CS, Kuliawat R. Evidence for leptin binding to proteins in serum of rodents and humans: Modulation with obesity. Diabetes **45:**1638–1643, 1996.
126. Saad MF, Riad-Gabriel MG, Khan A, Sharma A, Michael R, Jinagouda SD, Boyadjian R, Steil GM. Diurnal and ultradian rhythmicity of plasma leptin: Effects of gender and adiposity. J Clin Endocrinol Metab **83:**453–459, 1998.
127. Schoeller DA, Cella LK, Sinha MK, Caro JF. Entrainment of the diurnal rhythm of plasma leptin to meal timing. J Clin Invest **100:**1882–1887, 1997.
128. Reidelberger RD. Cholecystokinin and control of food intake. J Nutr **124:**1327S–1333S, 1994.
129. Grider J. Role of cholecystokinin in regulation of gastrointestinal motility. J Nutr **124:**13345–13395, 1994.
130. Schick RR, Harty GJ, Yaksh TL. Sites in the brain at which cholecystokinin octapeptide acts to suppress feeding in rats: A mapping study. Neuropharmacology **29:**109–118, 1990.
131. Louie D, Liddle R, Owyang C, Reidelberger R, Grider J. Overview symposium: New research in the physiology of cholecystokinin: Nutrition issues. J Nutr **124:**1306S–1339S, 1994.
132. Bertelemy P, Bouisson M, Vellas B. Postprandial cholecystokinin secretion in elderly with protein-energy undernutrition. J Am Geriatr Society **40:**365–369, 1992.
133. Miyasaka K, Kanai S, Ohta M, Funakoshi A. Aging impairs release of central and peripheral cholecystokinin (CCK) in male but not in female rats. J Gerontol **52A:**M14–M18, 1997.
134. Miyasaka K, Kanai S, Masuda M. Gene expression of cholecystokinin (CCK) and CCK receptors, and its satiety effect in young and old male rats. Arch Gerontol Geriatr **21:**147–155, 1995.
135. Ohta M, Tanaka Y, Masuda M, Miyasaka K, Funakoshi A. Impaired release of cholecystokinin (CCK) from synaptosomes in old rats. Neurosic Lett **198:**161–164, 1995.
136. Poston GJ, Singh P, Maclellan DG. Age-related changes in gallbladder contractibility and gallbladder cholecystokinin receptor population in guinea pigs. Mech Aging Dev **46:**225–236, 1988.
137. Khalil T, Walker PJ, Wiener I, Fagan CJ, Townsend J, Courtney M, Greeley J, George H, Thompson JC. Effect of aging on gallbladder contraction and release of cholecystokinin-33 in humans. Surgery **98:**423–429, 1985.
138. Masclee AA, Geuskens LM, Driessen WM. Effect of aging on plasma cholecystokinin secretion and gallbladder emptying. Age **11:**136–140, 1988.
139. Martinez M, Hernanz A, Gomez-Cerezo J, Pena JM, Vazquez JJ, Arnalich F. Alterations in plasma and cerebrospinal fluid levels of neuropeptides in idiopathic senile anorexia. Reg Peptides **49:**109–117, 1993.
140. Baile CA, McLaughlin CL, Della-Fera MA. Role of cholecystokinin and opioid peptides in control of food intake. Physiol Rev **66:**172–234, 1986.
141. McCaleb M, Myers R. Cholecystokinin acts on the hypothalamic noradrenergic system: Involved in feeding. Peptides **1:**47–49, 1980.
142. Willis GL, Hansky J, Smith GC. Central and peripheral proglumide administration and CCK-induced satiety. Reg Peptides **15:**87–98, 1986.
143. Voigt JP, Huston JP, Voits M, Fink H. Effects of cholecystokinin octapeptide on food intake in adult and aged rats under different feeding conditions. Peptides **17:**1313–1315, 1996.
144. Silver AJ, Flood JF, Song AM. Evidence for a physiological role for CCK in the regulation of food intake in mice. Am J Physiol **256:**R646–R652, 1989.
145. Crawley J, Beinfeld M. Rapid development of tolerance to the behavioral actions of cholecystokinin. Nature London **302:**703–706, 1983.
146. West DB, Fey D, Woods SC. Cholecystokinin persistently suppresses meal size but not food intake in free-feeding rats. Am J Physiol **246:**R776–R787, 1984.
147. Woods SC, Seeley RJ, Porte D, Schwartz MW. Signals that regulate food intake and energy homeostasis. Science **280:**1378–1382, 1998.
148. Drewnowski A. Taste preferences and food intake. Annu Rev Nutr **17:**237–253, 1997.
149. Geiselman JP. Control of food intake: A physiologically complex, motivated behavioral system. Endocrinol Metab Clin North Am **25:**815–829, 1996.
150. Tepperman F, Hirst M. Effect of intrahypothalamic injections of D-Ala D-Leu enkephalin on feeding and temp in rat. Eur J Pharmacol **96:**243–249, 1983.
151. Trenchard ES, Silverstone T. Naloxane reduces the food intake of normal human volunteers. Appetite **4:**43, 1983.
152. Yeomans MR, Gray RW. Effects of naltrexone on food intake and changes in subjective appetite during eating: Evidence for opioid involvement in the appetizer effect. Physiol Behav **62:**15–21, 1997.
153. Lau SM, Tang F. The effect of haloperidol on met-enkephalin, β-endorphin, cholecystokinin and substance P in the pituitary, the hypothalamus and the striatum of rats during aging. Prog Neuropsychopharmacol Biol Psychiatry **19:**1163–1175, 1995.
154. Rudski JM, Grace M, Kuskowski MA, Billington CJ, Levine AS. Behavioral effects of naloxone on neuropeptide Y-induced feeding. Pharmacol Biochem Behav **54:**771–777, 1996.
155. Brands B, Thornhill J, Hirst M, Gowdey C. Suppression of food intake and body weight gain by naloxone in rats. Life Sci **24:**1773–1778, 1979.
156. Tseng L, Cheng D. Acute and chronic administration of β-endorphin and naltrexone on food and water intake in rats. Fed Proc **39:**606, 1980.
157. Forman LJ, Sonntag WE, Van Vugt DA, Meites J. Immunoreactive β-endorphin in the plasma, pituitary, and hypothalamus of young and old male rats. Neurobiol Aging **2:**281–284, 1981.
158. Dupont A, Savard P, Merand Y, Labrie F, Boissier JR. Age-related changes in central nervous system enkephalins and substance P. Life Sci **29:**2317–2322, 1981.
159. Wang ZP, Man SY, Tang F. Age-related changes in the contents of neuropeptides in the rat brain and pituitary. Neurobiol Aging **14:**529–534, 1993.
160. Gambert SR, Garthwaite TL, Pontzer CH, Hagen TC. Age-related changes in central nervous system β-endorphin and ACTH. Neuroendocrinology **31:**252–255, 1980.
161. Missale C, Govoni S, Croce L, Bosio A, Spano PF, Trabucchi M. Changes of β-endorphin and met-enkephalin content in the hypothalamus-pituitary axis induced by aging. J Neurochem **40:**20–24, 1983.
162. Tang F, Tang J, Chou J, Costa E. Age-related and diurnal changes in met5-enk-arg6-phe7 and met5-enkephalin contents of pituitary and rat brain structures. Life Sci **35:**1005–1014, 1984.
163. Arey LB, Tremaine MJ, Mozingo FL. The numerical and topographical relations of taste buds to human circumvallate papillae throughout the life span. Anat Rec **64:**9–25, 1935.
164. Mochizuki Y. Topographical relations of tastebuds to circumvallate papillae of Japanese. Okajimas Folia Anat Jpn **15:**595–608, 1937.
165. Vanzant F, Alvarez W, Essterman G, Dunn H, Berkson H. The normal range of gastric acidity from youth to old age. Arch Intern Med **49:**345–359, 1932.
166. Wormsley K, Grossman M. Maximal histalog test in control subjects and patients with peptic ulcer. Gut **6:**427–435, 1965.
167. Bradley R, Stedman H, Mistretta C. Age does not affect numbers of

taste buds and papillae in adult rhesus monkeys. Anat Rec **212:**246–249, 1985.
168. Miller IJ Jr. Human taste bud density across adult age groups. J Gerontol **43:**B26–B30, 1988.
169. Mistretta C. Anatomy and neurophysiology of the taste system in aged animals. Ann N Y Acad Sci **561:**277–290, 1989.
170. Doty R. Influence of age and age-related disease on olfactory function. Ann NY Acad Sci **561:**76–86, 1989.
171. Weiffenbach J, Bartoshuk L. Taste and smell. Clin Geriatr Med **8:**543–555, 1992.
172. Bartoshuk L, Duffy V. Taste and smell. In: Masoro EJ, Ed. Handbook of Physiology: Aging. New York: Oxford University Press, pp363–375, 1995.
173. De Simone J, Heck G, Bartoshuk L. Surface active taste modifiers: A comparison of the physical and psycho-physical properties of gymnemic acid and sodium lauryl sulfate. Chem Sens **5:**317–330, 1980.
174. Rolls BJ, Drewnowski A. Nutrition and aging. In: Encyclopedia of Gerontology. New York: Academic, pp429–440, 1997.
175. Greip M, Verleye G, Franck A, Collys K, Mets T, Massart D. Variation in nutrient intake with dental status, age and odour perception. Eur J Clin Nutr **50:**816–825, 1996.
176. Duffy V, Backstrand J, Ferris A. Olfactory dysfunction and related nutritional risk in free-living elderly women. J Am Diet Assoc **95:**879–884, 1995.
177. Schiffman SS, Warwick ZS. Effect of flavor enhancement of foods for the elderly on nutritional status: Food intake, biochemical indices, and anthrometric measures. Physiol Behav **53:**395–402, 1993.
178. Cave D. Transmission and epidemiology of the *Helicobacter pylori.* Am J Med **100**(Suppl):125–185, 1996.
179. Frank E, Lange R, McCallum R. Abnormal gastric emptying in patients with atrophic gastritis with or without pernicious anemia. Gastroenterology **80:**A1151, 1981.
180. Goldschmiedt M, Feldman M. Age-Related Changes in Gastric Acid Secretion. Boca Raton, FL: CRC Press, pp13–30, 1993.
181. Goldschmeidt M, Barnett C, Schwarz B, Karnes W, Redfern J, Feldman M. Effect of age on gastric acid secretion and serum gastrin concentrations in healthy men and women. Gastroenterology **101:**977–990, 1991.
182. Kekki M, Sipponen P. Age behavior of gastric acid secretion in males and females with a normal antral and body mucosa. Scand J Gastroenterol **18:**1009–1016, 1983.
183. Horowitz M, Guy M, Chjatterton B, Collins P, Harding P, Shearman D. Changes in gastric emptying rates with age. Clin Sci **67:**213–218, 1984.
184. Smits G, Lefebvre R. Influence of aging on gastric emptying of liquids, small intestinal transit and fecal output in rats. Exp Gerontol **31:**589–596, 1996.
185. McDougal JW, Miller MS, Burks TF. Intestinal transit and gastric emptying in young and senescent rats. Dig Dis Sci **25:**A–15, 1980.
186. Varga F. Transit time changes with age in the gastrointestinal tract of the rat. Digestion **14:**319–324, 1976.
187. Portnoi VA. *Helicobacter pylori* infection and anorexia of aging. Arch Intern Med **153:**269–272, 1997.
188. DeCastro JM. Age-related changes in spontaneous food intake and hunger in humans. Appetite **21:**255–272, 1993.
189. Leibowitz SF. Brain neuropeptide Y: An integrator of endocrine, metabolic and behavioural processes. Brain Res Bull **27:**333–337, 1991.
190. Lynch WC, Hart P, Babcock AM. Neuropeptide Y attenuates satiety: Evidence from a detailed analysis of patterns ingestion. Brain Res **636:**28–34, 1994.
191. Verdery RB. Failure to thrive in old age: Follow-up on a workshop. J Gerontol **52A:**M333–M336, 1997.
192. Klein DC, Moore RY, Reppert SM. Suprachiasmatic Nucleus: The Mind's Clock. New York: Oxford University Press, pp191–196; 233–245, 1991.
193. Kahler A, Geary N, Eckel LA, Campfield LA, Smith FJ, Langhans W. Chronic administration of OB protein decreases food intake by selectively reducing meal size in male rats. Am J Physiol **275:**R180–R185, 1998.