Ann. Rev. Pharmacol. Toxicol. 1988. 28:247–68

ENDOGENOUS ANORECTIC AGENTS—SATIETINS

Joseph Knoll

Department of Pharmacology, Semmelweis University of Medicine, Budapest, Hungary, 1445

INTRODUCTION

Overweight is a serious predisposing factor to ill-health and mortality, and fat phobia exists due to the present dominance of the lean body image in the richer part of the world. These two recognitions have considerably increased interest in pharmacologic and other strategies for the treatment of obesity. Despite all efforts, however, none of the numerous anorectics marketed during the last two decades have lived up to expectations in 2–3-year follow-up studies. There is still a desperate need to provide a solution to the problem of obesity. It was unexpectedly observed that a number of endogenous substances, mainly peptides, with a well-known physiological spectrum, like glucagon, insulin, cholecystokinin, calcitonin, etc., also inhibit food intake, and that the satietins, a hitherto unknown family of α_1-glycoproteins in human and mammalian sera have potent and selective anorectic activity. These observations represent the most promising new line of research. This line intends to understand better the control of food intake, to clarify the etiology of feeding-related pathologies, and to elaborate a more efficient therapy for obesity.

MONOAMINERGIC TRANSMITTER MECHANISMS AND CURRENT PHARMACOTHERAPY FOR OBESITY

The classical demonstration by Hetherington & Ranson in 1940 (1) that lesion of the ventromedial hypothalamus (VMH) in rats leads to hyperphagia, and the brilliant experiments of Anand & Brobeck in 1951 (2) proving that the lesioning of the lateral hypothalamus (LH) in rats and cats results in hypophagia, led to the promulgation of the theory that feeding is regulated by a "feeding center" in the LH and a "satiety center" in the VMH (3).

Another influence on our present knowledge about the regulation of feeding

0362-1642/88/0415-0247$02.00

was Nathanson's 1937 observation (5) that the sympathomimetic drug, amphetamine, introduced as a central stimulant by Prinzmetal & Bloomberg in 1935 (4), reduces body weight in humans. Its anorectic effect was clearly proved in clinical trials by Harris et al in 1947 (6) and by Williams et al in 1948 (7).

Further studies revealed that the α-adrenergic receptor mechanism plays a role in the stimulation of natural hunger. Leibowitz proposed that the stimulatory mechanism is mediated by adrenergic pathways that originate in the midbrain and ascend through the periventricular region of the diencephalon (8).

On the other hand, catecholamines and amphetamine, injected into the hypothalamus of hungry animals, were found to suppress feeding behavior in animals (8, 9). Both α- and β-receptor stimulants, as well as dopamine, proved to be effective. Catecholaminergic mechanisms located in the LH, which inhibit food intake, help to explain the anorectic effect of the phenylisopropylamine relatives, which act primarily as releasers of noradrenaline. It now seems highly probable that ascending noradrenergic pathways that pass through the hypothalamus serve a satiety function in the intact animal, as the injection of 6-hydroxy-dopamine into the ventral noradrenergic bundle in the midbrain was found to induce hyperphagia (10). Small doses of amphetamine that release primarily noradrenaline strongly inhibit food intake. This speaks in favor of the major role of noradrenaline in the anorectic effect of small and medium doses of amphetamine, a view further supported by the fact that pretreatment of the animals with α-methylparatyrosine, which blocks the synthesis of noradrenaline, prevents the satiety effect of amphetamine (11).

Amphetamine differs only in one methyl group (attached to the α-carbon) from β-phenylethylamine, an endogenous releaser of noradrenaline in the brain. Noradrenaline was claimed to act as a physiological central nervous stimulant (for reference, see 12) with a putative relation also to regulation of feeding, as an inhibitor of food intake (13).

Whereas amphetamine and its many variants act as releasers of noradrenaline, mazindol—an imidazol derivative and much weaker central nervous stimulant than the amphetamines—exerts its anorectic effect by enhancing mainly dopaminergic tone in the brain, inhibiting the reuptake of this amine.

There is, however, a second, serotonin-mediated satiety system in the brain. This system is facilitated by those amphetamine derivatives halogenated in the para- and/or meta-position; these are highly selective and potent releasers of serotonin and inhibitors of the reuptake of this amine. Out of the anorectics in medicinal practice, fenfluramine is the drug which typically acts through this mechanism (14). The anorectic effect of fenfluramine remains unchanged in animals pretreated with α-methylparatyrosine (11).

The role of monoaminergic mechanism in the control of feeding was critically reviewed by Hoebel in 1977 (15) and by Leibowitz in 1980 (16).

ANORECTIC EFFECT OF ENDOGENOUS PEPTIDES AND THE HYPOTHESIS OF A SATIETY CASCADE

The restricted therapeutic value of the different types of anorectic drugs in clinical practice focused the attention on the hitherto unexplored details of the biogrammar of feeding. The recognition of the anorectic effect of known endogenous substances and above all the discovery of hitherto unknown endogenous anorectic compounds represent the most promising new strategy to approach feeding-related pathologies.

This new line of research started with the paper by Gibbs, Young & Smith (17) who reported that cholecystokinin (CCK) decreased food intake in rats. Also the synthetic octapeptide (CCK-OP) and decapeptide, caerulein, have a food intake suppressing effect (18). CCK, released by ingesta entering the intestine, might play the role of an intestinal satiety signal under normal feeding conditions and may act as a short-term satiety agent. This view is substantially supported by a number of observations in different animals (17, 19–22). Even the possibility that cholecystokinin octapeptide, known to be released in the brain (23), may act as a physiological suppressor of feeding was proposed by Della-Ferra & Baile (24) on the basis of the long-lasting anorectic effect of continuous lateral cerebroventricular injections of CCK-OP in sheep. Knoll (11) was unable to detect the suppression of food intake in rats deprived of food for 96 hr in response to intracerebroventricular administration of a single huge dose of CCK-OP; thus, the possibility that the continuous administration of the octapeptide might exert an effect through the release of calcitonin in the brain deserves attention. CCK and bombesin stimulate the release of calcitonin from the thyroid gland after meals, and calcitonin is a highly potent, long-lasting suppressor of food intake. This was discovered by Freed et al in 1979 (25) and corroborated by others (26–28). Highly specific calcitonin receptors exist in the brain (29) and this peptide is also released in brain tissue, so an involvement of calcitonin in the physiological regulation of feeding, by acting locally on the hypothalamic control of food intake, cannot be excluded. It has to be considered, however, that in comparison to the physiological concentrations of calcitonin, relatively high amounts are needed for inhibiting food intake. It was shown by Levine and Morley (28) that the anorectic effect of calcitonin is secondary to its inhibition of calcium uptake into hypothalamic neurons.

Gibbs et al (30) also reported on bombesin, a tetradecapeptide, originally isolated from amphibian skin (31), a potent releaser of gastrin and CCK, and a highly potent stimulant of intestinal, gallbladder, urinary tract, and uterine smooth muscle. Bombesin suppressed food intake in rats in a way similar to CCK. As bombesin-like immunoactivity is widely distributed in mammalian organs, including the brain, and is present in large quantities in the gastric mucosa, Gibbs et al (30) assumed that it may act as a gastric satiety signal.

Besides CCK, bombesin, and calcitonin, a number of known peptides were reported to suppress feeding behavior, among them thyrotropin releasing hormone (TRH) (32) and its metabolite, histidyl prolin diketopiperazine (33), corticotropin releasing hormone (34), neurotensin (35), somatostatin (36), enterogastrone (37), pancreatic polypeptide (38) and vasoactive intestinal peptide (39).

A TRH analogue, pGlu-His-GlyOH, claimed to have potent anorectic effect, was extracted from the urine of anorexia nervosa patients by Reichelt et al (40) and seemed at first to open an interesting new line of research. Intravenous and intracerebroventricular administration of high doses of this tripeptide did not, however, influence food intake in rats deprived of food for 96 hr (11). Nance and co-workers using the experimental conditions described by Reichelt et al were also unable to establish the anorexogenic potency of pGlu-His-GlyOH (41).

Among the endogenous peptides related to feeding, glucagon and insulin are of special importance. Both were reported to suppress feeding and to be released from the pancreas following food stimulation in the small intestine.

That glucagon inhibits food intake in humans (42, 43) and rats (44) was observed in early studies and explained as the consequence of hepatic glycogenolysis. Also hepatic-portal infusion of glucagon was reported to decrease feeding in rats (45).

Insulin is present in the cerebrospinal fluid (CSF) in dogs (46) and humans (47). Obese humans have higher insulin levels, and it was proposed that CSF insulin serves as an adiposity-signal (48). Insulin is also claimed to play the role of a satiety hormone (49). Chronic insulin infusion was found to suppress food intake and body weight gain in rats (50). In baboons chronic intracerebroventricular infusion of insulin also reduced food intake and body weight (51).

Free fatty acid metabolites, 2-deoxytetronate (2-DTA), 3-deoxytetronate (3-DTA), and 3-deoxypentonate (3-DPA) were found to be present in the blood of rats during various fasting stages and were proposed to contribute to satiation (52, 53). After one short application of 2-DTA, into the third cerebral ventricule in normal rats at 1 μmol concentration, food intake was continuously suppressed for 24 hr; 3-DPA clicited food intake; and their effects are claimed to be mediated through the glucose-sensitive neurons in the lateral hypothalamic area (54).

The intracerebroventricular administration of PGE_2 and PGF_2 also suppressed food intake in several feeding models; this was shown first in cats by Horton in 1964 (55) and was corroborated by many others in different species (28, 56–58).

The number of known endogenous substances which have been proved (at least under appropriate experimental circumstances) to inhibit food intake is rapidly increasing. However, neuropeptide-Y is the only substance described

in the literature which, when given intracerebroventricularly, stimulates food and water intake (59–61).

Out of numerous known endogenous peptides with anorectic activity, CCK and calcitonin seem to be the most important ones from the point of view of the regulation of feeding.

Gibbs et al proposed the hypothesis that CCK is primarily responsible for postprandial satiety. Since the blood-brain barrier is almost impermeable for CCK and abdominal vagotomy abolished the satiety effect of the peptide, the anorectic effect of CCK is clearly vagally mediated. The same is true for glucagon and somatostatin, whereas bombesin acts via a nonvagal mechanism. While CCK in very small doses (8 μg/kg) inhibits food intake without interfering with water intake, the latter is inhibited only with higher doses of CCK. In different species (mouse, rat, monkey, and human), CCK disrupted the normal behavioral sequence that characterizes satiety. It also terminated sham feeding and elicited the behavioral sequence of satiety in food-deprived rats, which do not satiate spontaneously (for review see 62). All these data speak in favor of the assumption that CCK might be the most important gastrointestinal satiety signal. Of the endogenous substances possessing anorectic activity, calcitonin is the most potent and has the longest duration of action. CCK evidently cannot play the role of a blood-borne satiety signal, but is such a role also unlikely for calcitonin? This question will be treated in the next section.

The multiplicity of endogenous substances and systems related to the regulation of feeding and the extremely high complexity of the factors influencing hunger and appetite led to the widespread view that a single satiety signal playing a rate-limiting role in the termination of feeding is unlikely. As Morley & Levine expressed it, "the modern day Sir Galahads seeking the single Holy Grail to cure obesity may well search in vain" (63).

Morley et al tried to integrate almost all substances and systems involved in feeding regulation in a mechanism called the satiety cascade (64). In contrast to their firm dismissal of the Holy Grail of Satiety, they suggest an unknown "major inhibitory substance" as the centrally acting main satiety signal in the medial hypothalamus. Their "proposals" that either protaglandins or calcitonin gene related peptide may play this role are evidently unfounded, and the authors themselves are not entirely serious about them. An unknown substance of extremely high importance is unquestionably missing in the satiety cascade. This point is the subject of the next section.

A BLOOD-BORNE, RATE-LIMITING SATIETY SIGNAL WITH POTENT SELECTIVE ANORECTIC ACTIVITY

Deprivation of food leads to deficiencies. Essential nutrients run low, energy stores diminish, etc. The longer the duration of food deprivation, the more

marked are these changes. The intensity of that specific kind of central excitatory state, which we call the hunger drive, is within reasonable time limits proportional to the duration of food deprivation. This is the physiological mechanism which determines that the animal will be ready to surmount every obstacle, even if life is in the balance, to seize its food (for review, see 65).

The relation between the duration of food deprivation and the intensity of the specific central excitatory state, i.e. the hunger drive, is shown in Figure 1. This figure shows the phenomenon of essential importance for further research. In the rats deprived of food for 132 hr, feeding for 30 min decreased the intensity of the hunger drive from 6.2 to 2.8 units. During this short period, clearly, neither the energy stores nor the amount of the essential nutrients in the organism changed. A time-consuming complex process of digestion and resorption is the sine qua non for restoring the deficits caused by the long-lasting food deprivation. This means that the brain is sated before the process to replenish the deficits in the organism starts to operate. Thus, the ingestion of food appropriate in quality and quantity may lead to an immediate activation of a rate-limiting satiety signal in the blood, which terminates the hunger drive (for review see 66).

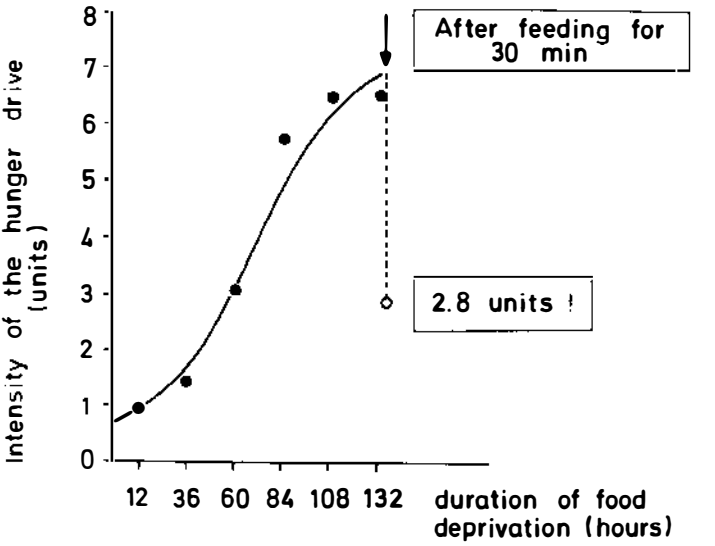

Figure 1 The gradual increase of the intensity of the hunger drive in rats during 132-hr-long starvation and its extinction within a short period of feeding. The intensity of the hunger drive was expressed in units according to a method based on the assessment of the spontaneous motility of starving rats in an open field (see Ref. 65, Chap. 11). The experiment was performed with a group of 20 rats weighing 200–250 gr before starvation and supplied with water ad libitum. Chow pellets were offered for 30 min after 132-hr-long starvation.

This hypothesis initiated the search for a substance which (*a*) easily penetrates across the blood-brain barrier, (*b*) is active by intracerebroventricular administration, (*c*) is present in detectable amounts in the blood of different species, and (*d*) can block the hunger drive at its peak intensity, i.e. inhibits food intake in rats deprived of food for 96 hr.

Such a substance in human serum was detected in 1977 and named *satietin* (11, 67). Satietin was isolated from human plasma and identified as a previously unknown α_1-glycoprotein. As the substance envisioned was thought to be a smaller molecule, the procedure started with the ultrafiltration of human plasma through an Amicon UM 10 flat membrane, to eliminate the serum constituents with molecules larger than 10 kd. Satietin activity was found in the filtrate, but it was soon realized that the molecular size of the substance responsible for the potent and long-lasting anorectic activity was over 50 kd, i.e. satietin passed through the Amicon UM 10 membrane in a paradoxical manner. This first step of the isolation procedure separated satietin from the serum constituents larger than 10 kd.

A second step helpful for separating satietin from a great number of contaminating constituents depended on its resistance to trichloroacetic acid (TCA). With the aid of TCA most of the higher molecular weight proteins and peptides that accompany satietin in the ultrafiltrate could be percipitated. The isolation procedure was then followed by two steps of gelchromatography (using Sephadex G 15 and Bio-Gel P-2 columns), continued with affinity chromatography on Con-A-Sepharose column, and finished by a last desalting step on Bio-Gel P-2 column (68–74).

Satietin was found to be an α_1-glycoprotein with a molecular size of 64 kd. An unusually high carbohydrate content (70–75%) and a protein content of 14-15 wt % was described for satietin (75–77).

Two anomalic carbohydrate constitutents, rhamnose and glucose, were detected in satietin. Human serum contains a high number of glycoproteins, i.e. proteins containing covalently linked carbohydrates. The pentose in all the glycoproteins in human serum described in the literature is fucose. In striking contrast to the known glycoproteins in human serum, the pentose constituent in satietin is rhamnose. The hexoses found in the glycoproteins in human serum are mannose and galactose, exclusively. Satietin contains three hexoses: mannose, galactose, and glucose. The apparently highly individual carbohydrate composition in satietin hints at the specific nature of this glycoprotein (75, 76).

Further studies revealed the presence in human serum of a close relative to satietin, named satietin-D. This serum constituent was detected unexpectedly when human serum was filtered, not through the Amicon UM 10 flat membrane, but through an Amicon hollow fiber concentration system operated with an Amicon high-performance hollow fiber cartridge with a nominal

cutoff of 10 kd. Satietin unexpectedly did not pass through this membrane. From the filtrate, however, another α_1-glycoprotein, with an action similar to satietin, was isolated. This substance, named satietin-D, proved to be a glycoprotein with a molecular size of 43-45 kd. The carbohydrate content in satietin-D was described as 55–60%, and the protein content was 20.5 wt %. Satietin-D, like satietin, showed resistance towards TCA. The carbohydrate constituents—rhamnose, mannose, galactose, and glucose—found in satietin-D were the same as those in satietin (78–82).

When the isolation procedures described for satietin and satietin-D, respectively, were used, about 10 mg/liter was the total amount of the two glycoproteins extracted from human serum. Considering that the amount of protein containing covalently bound carbohydrate is over 20 g/liter in human serum, the satietins represent a trace fraction of glycoproteins only. This explains why the satietins remained undiscovered until specific biological activity revealed their presence in the serum (for review see 75).

Hypothesis of the relation of the anorectic effect of satietin-D to the α_1-glycoprotein was substantially supported by a series of experiments using immunological methods. Anti-satietin-D lgG from rabbit serum was isolated for these experiments by ion-exchange chromatography, and the anti-satietin-D lgG was coupled to an activated AH-sepharose 4B gel column. Then semi-purified satietin-D was loaded onto this column. Anti-satietin-D absorbed satietin-D during this procedure, and the bound satietin-D was desorbed with ammonium rhodanide. The desorbed substance gave a typical one-band reaction with satietin-D antiserum. The substance was found to be a 41-kd glycoprotein containing rhamnose, glucose, mannose, and galactose that retained its characteristic long-lasting anorectic effect in rats deprived of food for 96 hr (75, 83). Whether the large molecule serves only as a vehicle for a smaller active molecule, which is then released in the brain by a hitherto unknown mechanism is a question that remains for the time being unanswered. In this case, however, the linkage between such a biologically active small molecule and the glycoprotein must be extremely stable and specific as it survives different steps of purification. Anyway, further research is needed to learn whether the whole glycoprotein molecule, a part of it, or a small entity coupled with the glycoprotein is responsible for the biological activity.

Satietin activity was demonstrated in the serum of mammals belonging to different orders. Of the rodents, mouse, different strains of rats, guinea pig, and rabbit were taken as examples. Cattle and horse represented the ungulates; cat and dog, the order of carnivora. Satietin was also detected in avian blood. Goose serum was used as an example of the order of anseriformes. According to preliminary observations, satietins found in the sera of different animal species are closely related glycoproteins, but they are not identical with each other or with human satietin (for review see 75).

The first paper on satietin (11) showed that the anorectic effect of the almost unpurified material reached its peak within 5 hr when given intracerebroventricularly. In contrast, the inhibition of food intake developed with short latency at the intravenous route of administration of this preparation, and the peak effect was reached within 1 hr. As soon as it was realized that TCA leaves satietin activity unchanged, and TCA precipitation of proteins before gel chromatographic purification of satietin was introduced, samples were produced which inhibited feeding with a rapid onset when given intracerebroventricularly. This suggested that in its native form satietin in human serum is coupled with a carrier protein precipitated by TCA (for review see 75).

Satietin proved to be a highly potent anorectic substance which inhibited food intake in a dose dependent way in rats deprived of food for 96 hr. A biological assay for measuring satietin activity in units was developed. One unit of activity was defined for satietin as the amount which, when given intracerebroventricularly, decreased the chow pellet consumption of rats deprived of food for 96 hr during the first day of feeding from 24.04 ± 0.76 g to 10 g. With higher intracerebroventricular doses of satietin (2–3 units/rat), the 24-hr consumption of the fasting rats could be reduced to 3–5 g, and the animals began to eat more on the second day of feeding only. With the highest grade of purification, satietin preparations reached 100 units/mg activity (68, 70, 71).

Satietin and satietin-D were found on a weight basis to be approximately equally potent anorectic substances in the rat at intracerebroventricular, intravenous, or subcutaneous administration. Independent of the route of administration, the peak effect was reached within 1 hr, and the inhibition of food intake lasted over 24 hr (74, 84, 85).

Because the supply of material is scant, most of the biological information on satietin and satietin-D stems from the intracerebroventricular administration of these substances. For the same reason the anorectic effect of satietins during long-term administration remains to be studied.

Table 1 shows the unchanged efficiency of two consecutive doses of highly purified satictin sample in a group of normally fed rats. The effect of the first dose of satietin lasted about 48 hr, and the food consumption returned to normal on the third day after intracerebroventricular injection of satietin. There was no sign of rebound. The intensity and the time course of effect of the second dose of satietin were identical with that of the first. Food consumption was severely depressed during the 48 hr after the injection of satietin, but no significant difference in the third day consumption was to be observed. Food consumption again remained normal on the consecutive days.

A recent review on the satietins (75) emphasized that though it was fortunate to detect the anorectic effect of human satietins in the rat, they are alien substances in this species. Thus, no conclusion can be drawn from the

Table 1 Unchanged efficiency of two consecutive doses of satietin and the lack of rebound in normally fed rats[a,b]

Days	1	2	3	4	5	6	7	8	9	10	11	12
Daily food consumption (g)	26.2	6.89[c]	14.8[c]	27.4	25.2	24.2	4.5[c]	13.2[c]	21.8	26.6	29.2	27.4
	↑ satietin					↑ satietin						

[a] Statistics: Student's *t* test for two means.
[b] n = 12; 40 μg satietin was injected intracerebroventricularly into the lateral ventricle (right and left side)
[c] Significant; $p < 0.001$
(A highly purified satietin sample containing 100 units/mg was used in the experiment.)

unexpectedly long offset of their effect with regard to the nature and mechanism of action of satietin and satietin-D in humans. As different animal species seem to possess their own characteristic satietins, only the analysis of the effects of satietin isolated from rat serum may reveal the role of this glycoprotein family in the regulation of food intake in the rat.

Physiological, pharmacological, and behavioral studies unequivocally support the view that what is primarily regulated in the brain is satiety (86–88). According to the satietin hypothesis of the regulation of feeding (for review see 75), the ingestion of the appropriate amount of proper food liberates an amount of satietin in the blood that fully activates the satiety center. That in turn, like a brake, keeps the feeding center inhibited. This state in the brain was thought to be experienced subjectively as the feeling of fullness. The hypothesis further postulates that, following the last meal, as time passes the satietin concentration in the appropriate brain area continuously decreases. In proportion to this natural and unavoidable process of elimination, the satiety center gradually tends toward the resting state. That is, the brake is released, the feeding center is disinhibited, and this state in the brain is thought to be experienced subjectively as the feeling of hunger, the intensity of which is proportional to the duration of food deprivation.

Physiological considerations speak in favor of the assumption that the satietins act through a satiety mechanism. As the essential mechanism of action of an anorectic compound, i.e. whether it acts via feeding or satiety, can be resolved by analyzing the substance-induced changes in the microstructure of eating, the effect of satietin in comparison to amphetamine and fenfluramine was studied in this respect (74, 89).

Blundell et al (90) demonstrated that amphetamine, which inhibits a feeding mechanism, significantly prolonged in rats the time elapsed before taking the first tablet in an eatometer, whereas fenfluramine, which acts by stimulating a satiety mechanism, left this latency unchanged. In the satietin-treated rats, although this difference was not statistically significant, there was a tendency for taking the first tablet after a shorter latency, indicating that this endogenous substance activates a satiety mechanism (for review see 75).

In another series of experiments, the microstructure of eating was analyzed in three groups of rats deprived of food for 24 hr by measuring the number of tablets eaten in the eatometer during four consecutive 15 min periods, during which one group was under the influence of amphetamine, fenfluramine, and satietin, respectively. The doses of all three drugs were equianorectic. In the first 15-min period, food consumption after amphetamine injection was significantly lower than it was for groups treated with fenfluramine and satietin. As the doses given were equianorectic, amphetamine-treated animals ate significantly more in the third and fourth 15 min periods of the eating session than did the fenfluramine and satietin treated ones. Fenfluramine and satietin

treated animals ate similar amounts of food during the four 15 min periods of the eating session. These data further supported the conclusion that satietin acts via a satiety mechanism (89).

It is well known that fenfluramine-induced satiety is related to the serotonergic system. For the lesioning of serotonergic neurons through coagulation of their cells in the medial and dorsal raphe system leads to the loss of fenfluramine's effect on feeding. Satietin, however, remained fully active in the raphe-lesioned animals (91).

Because satietin acted on feeding, as did fenfluramine, via a satiety mechanism, the effect of these two substances on serotonergic transmission was compared by measuring the contraction of the m. tibialis following stimulation of the hind paw in acutely spinalized rats. Serotonin is known to be involved in this spinal reflex. Fenfluramine in doses even lower than the anorectic dose level, increases the contractions of the m. tibialis anterior to stimulation by enhancing the activity of the serotonergic link in the reflex. Satietin proved to have no effect on the contractions even when given intravenously in a much higher than anorectic dose.

That amphetamine and mazindol act through a catecholaminergic transmitter is clearly shown by the antagonism of the anorectic effect of these drugs in rats pretreated with α-methyl-paratyrosine, which blocks the synthesis of the catecholamines. The anorectic effect of the satietins remains unchanged in α-methylparatyrosine-treated rats. Thus, the mechanism of action of the anorectic effect of satietin is different from the amphetamine-type anorexia, as well as, from the fenfluramine-type satiety (for review see 75).

During the last decade a number of investigators have studied the relationship between opioid peptides and feeding. Several lines of investigation seem to support the idea that opioid peptides are also involved in the highly complex chain of events forming the biochemical basis of hunger drive. In 1977 Grandison & Guidotti demonstrated that injections of the GABA agonist muscimol and of the potent endogenous opioid peptide β-endorphin into the medial hypothalamus stimulate food intake (92). These effects are antagonized by naloxone (93). In 1978 Margules et al found that genetically obese mice and rats have higher concentrations of β-endorphin in the hypophysis and plasma than do their leaner littermates (94). Fasting was found to be associated with decrease in hypothalamic β-endorphin (95). In agreement with these findings, exogenous as well as endogenous opiate receptor agonists were observed to stimulate food and water intake (96–98), and opiate antagonists proved to possess anorectic potency in animal species as well as in humans (99–106). To check the possible relation between satietin and opiate receptors, the effect of satietin was investigated on isolated organs (longitudinal muscle strip of the guinea pig ileum, mouse vas deferens, and cat splenic strip) used for testing opiate agonists and antagonists. Satietin did not

exert any effect in these tests, and it failed to influence the effect of opiate agonists (70, 71).

The anorectic drugs used today in medicinal practice act via catecholaminergic or serotonergic transmission, and because these transmitter mechanisms are involved in a great number of different functions in the brain and in the periphery, none of the anorectics inhibit food intake selectively. The lack of selectivity leads to a number of side effects. Anorectic drugs are mainly considered to be short-term adjuvants in a more complex therapy of obesity including calorie-restriction, appropriate exercize, and psychological support.

As mentioned previously, a number of well-known endogenous substances, mainly peptides, also suppress food intake. Nine peptides, the most relevant ones, are listed in Table 2, together with their main effects in the physiological dose range. As much higher doses are usually needed for the suppression of food intake, the anorectic effect of these endogenous peptides is more of theoretical than of practical interest.

The satietin family of peptides represents, for the time being, the only group that seems to suppress food intake without having any noticeable central and peripheral effect in the anorectic dose range.

Table 3 summarizes the effects of equianorectic doses of satietin, calcitonin, amphetamine, and fenfluramine on the behavior of rats in a battery of tests. Fenfluramine, in the anorectic dose, was found to be a strong inhibitor in all the tests studied. It decreased locomotion in the open field, strongly interfered with unconditioned avoidance reactions, inhibited the development of conditioned reflexes, blocked learning and retention in one-way and two-way avoidance systems, inhibited the recall of previously firmly developed conditioned response, and completely inhibited copulatory behavior in male rats. In contrast to fenfluramine, amphetamine was stimulatory in the tests and facilitated performances. The anorectic dose of calcitonin inhibited the acquisition of a conditioned reflex in a one-way avoidance system, blocked the recall of a firmly established conditioned response, and left the rat's performances in three other tests unchanged. Satietin was ineffective in all tests.

Table 4 compares the effects of satietin, calcitonin, amphetamine, and fenfluramine on the metabolic rate, body temperature, and blood pressure. Again, none of the parameters were influenced by a dose of satietin, which blocked food intake in rat completely.

Quite recently B. Knoll and coworkers (115) demonstrated that satietin-D was also ineffective in the same battery of behavioral tests.

That satietin is highly selective to suppress food consumption is further supported by the finding that the intracerebroventricular administration of 1–2 units of satietin into the lateral ventricle, which exerts a strong anorectic

Table 2 Most important endogenous peptides which, besides their known physiological roles, also suppress food intake

Name of the peptide	Physiological significance	Described to have food intake suppressing effect in Year	Chemical nature	Does the substance inhibit food intake in rats deprived of food for 96 hours?
Glucagon	Hormone of fuel mobilization	1957	3,5 kd polypeptide	No
Cholecystokinin	Produces contraction of the gall bladder and relaxation of the sphincter of Oddi	1973	3.9 kd polypeptide	No
TRH	Thyrotropin-releasing hormone	1977	L-pyroglutamyl-L-histidyl-L-prolin amide	No
Insulin	Hormone of fuel storage	1979	6 kd polypeptide	No
Bombesin	Releaser of gastrin	1979	tetradecapeptide	No
Calcitonin	Hypocalcemic hormone inhibits bone resorption by altering osteoclastic and osteocytic activity	1979	3.6 kd polypeptide	Yes
Somatostatin	Inhibits secretion of growth hormone. Inhibits the release of insulin and glucagon	1979	tetradecapeptide	No
Vasoactive intestinal polypeptide	Vasodilator and pancreatic secretagogue	1981	3 kd polypeptide	No
Neurotensin	Increases secretion of ACTH, gondatropins and glucagon. Decreases secretion of insulin	1982	tridecapeptide	No

Table 3 Comparison of the effects of satietin, calcitonin, amphetamine, and fenfluramine on the behavior of rats in a battery of tests[a,b]

	Number of rats	Satietin, i.c.v.	Calcitonin, i.c.v.	Amphetamine, i.v.	Fenfluramine, i.c.v.	Method
Locomotor activity	10	none	none	strong facilitation	strong inhibition	open field
One-way avoidance	10	none	none	strong facilitation	strong inhibition	modified jumping test (107)
Two-way avoidance	12	none	none	facilitation	inhibition	shuttle-box
One-way conditioning	10	none	strong inhibition	strong facilitation	strong inhibition	screening test 1 (108)
Consolidated conditioned reflex	6	none	inhibition	none	strong inhibition	jumping test (109)
Male copulatory behavior	13	none	—	facilitation	inhibition	(110)

[a] All compounds were administered in the dose equianorectic with satietin (usually 1–2 units), either intracerebroventricularly (icv) or intravenously (iv).
[b] Satietin was isolated from human serum. For methodological and other details, see Refs. 111 and 112.

Table 4 Comparison of the effects of satietin, calcitonin, amphetamine, and fenfluramine on metabolic rate, body temperature, and blood pressure in the rat[a,b]

	Satietin	Calcitonin	Amphetamine	Fenfluramine	Method
Metabolic rate	none	significant increase (39%)	significant increase (97%)	none	Issekutz & Issekutz (113)
Body temperature	none	slight, statistically insignificant elevation	slight, statistically insignificant elevation	none	continuous measurement of rectal temperature
Blood pressure	none	none	significant increase	slight decrease	Via the carotid artery

[a] All compounds were administered intracerebroventricularly in doses equianorectic with the dose of satietin. Satietin prepared from human serum was used in the experiments.
[b] For methodological and other details, see Ref. 114.

effect, has no effect on the water intake of rats (for review see 75). In a series of experiments, the effects of satietin and calcitonin on the water intake of water-deprived rats were compared. Administration of satietin left water intake unchanged, whereas 1 MRC unit of calcitonin decreased it significantly (116). Satietin did not change the water intake in rats deprived of food for 96 hr but supplied with water ad libitum. In contrast to satietin, amphetamine doubled the water intake of rats under the same experimental circumstances (75).

In a series of experiments the influence of food deprivation on the blood levels of glucose, insulin, and glucagon in rats was studied. The rats deprived of food for 96 hr maintained normal glucose and glucagon levels. The blood concentration of insulin, however, dropped from 232.02 ± 23.93 pmol/l to 12.48 ± 0.7 in the fasting animals. The intracerebroventricular administration of satietin did not change the blood sugar, insulin, and glucagon levels in the blood either in normally fed rats or in rats deprived of food for 96 hr. Amphetamine treatment increased the insulin concentration in the blood of food deprived rats from 11.37 to 73.47 pmol/l without altering the blood levels of glucose and glucagon (117). Because an intracerebroventricular dose of satietin, which suppressed food intake significantly for more than 24 hr, did not induce any significant change in the blood levels of glucose, insulin, and glucagon in both normally fed and food deprived animals, it was concluded that the anorectic effect of satietin is unrelated to carbohydrate metabolism (for review see 75).

The findings show that satietins are highly potent and selectively acting endogenous anorectics widely distributed in the world of vertebrates; reasonable amounts of these glycoproteins are present in human blood; the site of effect of these substances is in the central nervous system; and they inhibit food intake via the activation of a satiety mechanism. The hypothesis was thus put forward that the satietin family may play the role of a rate-limiting, blood borne satiety-signal system in the negative feedback of food intake. That is, it may serve as the essential link in the regulation of feeding connecting the gastrointestinal tract and the brain via the blood stream (71).

Calcitonin, like the satietins, suppresses food intake in rats deprived of food for 96 hr, whereas all other endogenous substances reported to have an anorectic effect are ineffective in animals with such an intense hunger drive. Calcitonin has its specific hormonal effect; it inhibits bone resorption by altering osteoclastic and osteocytic activity. Considering the high potency of calcitonin in influencing calcium metabolism, in harmony with the very low physiological concentrations (70–120 picogram/ml) of this hormone, it inhibits food intake in a relatively high amount only. The reduction of feeding in rats by calcitonin is evidently secondary to its inhibition of calcium uptake into hypothalamic nerves; this was shown by Levine & Morley (28), as

mentioned previously. Even if calcitonin is lacking the selectivity of satietin, its potential role as a blood-borne satiety signal needs consideration. The satietins were calculated to be present in human blood in about 4500 times greater molecular concentration than calcitonin, whereas their anorectic potency, on a molecular basis, proved to be approximately equal (for review see 75). These facts preclude the possibility that calcitonin connects via the blood stream the gastrointestinal tract with the areas in the brain that regulate feeding. According to our present knowledge satietins are, no doubt, the best by far for this role. However, as calcitonin is also released in the brain, it might be involved in the physiological regulation of feeding as a locally acting hormone in the hypothalamus.

The hypothesis that satietins play the role of satiety signals of crucial importance in the negative feedback of food intake imperatively raises the question of the possible relationship between the satietins and feeding-related pathologies.

The first, and until now the only paper in this direction was an attempt by Harmath, Barna & Knoll to investigate satietin-D in the blood of obese versus normal weight humans (83). The presence of satietin-D with satietin-D antiserum in 72 obese persons with no detectable illness and in 19 normal weight healthy volunteers was tested. The electrophoretogram of the serum of the 19 healthy volunteers with normal weight mixed with satietin-D antiserum was highly characteristic. Satietin-D antibodies precipitated satietin-D in these sera in the form of a diffuse band. Remarkable changes in satietin-D were found in 14 out of 72 sera taken from the obese persons. The absence of satietin-D was found in five cases, a deficiency in the satietin-D content in three cases, a changed mobility of satietin-D in five cases, and a deficiency coupled with changed mobility in one case. All in all, in this first preliminary series of experiments, conspicuous changes in satietin-D in one fifth of the otherwise healthy obese persons were observed.

DIRECTIONS FOR FUTURE RESEARCH

An obvious interpretation of these data would be that in a part of the obese patients overeating is related to the hypofunction of the satiety system. The prerequisites, however, for the formulation of the attractive hypothesis that hypo- and hyperfunction of the satiety system may play an etiological role in feeding related pathologies are numerous: the exact chemical nature of the satietins has to be clarified; methods measuring their blood concentrations with high accuracy must be developed; the pathways of their synthesis and metabolism need to be discovered; and last, but not least, the specific binding sites for the satietins and their location in the brain have to be identified and characterized.

Literature Cited

1. Hetherington, A. W., Ranson, S. W. 1940. Hypothalamic lesions and adiposity in the rat. *Anat. Rec.* 78:149–72
2. Anand, B. K., Brobeck, J. R. 1951. Hypothalamic control of food intake in rats and cats. *Yale Biol. Med.* 24:123–40
3. Stellar, E. 1954. The psychology of motivation. *Psychol. Rev.* 61:5–22
4. Prizmetal, M., Bloomberg, W. 1935. The use of benzedrine for the treatment of narcolepsy. *JAMA* 105:2051–54
5. Nathanson, M. H. 1937. Central action of β-amino-propyl-benzene (benzedrine): Clinical observations. *JAMA 108:*528–31
6. Harris, S. C., Ivy, A. C., Searly, L. M. 1947. Mechanism of amphetamine-induced loss of weight. Consideration of hunger and appetite. *JAMA* 134:1468–75
7. Williams, R. H., Daughadey, W. H., Rogers, W. F., Aspar, S. P. Towery, D. T. 1948. Obesity and its treatment with particular reference to use of anorexogenic compounds. *Ann. Intern. Med.* 29:510–32
8. Leibowitz, S. F. 1976. Brain catecholaminergic mechanisms for control of hunger. In *Hunger: Basic Mechanisms and Clinical Implications,* ed. D. Novin, W. Wyrwicka, G. A. Bray, pp. 1–18. New York: Raven
9. Booth, D. A. 1968. Amphetamine anorexia by direct action in the adrenergic feeding system of rat hypothalamus. *Nature* 217:869–70
10. Ahlskog, E., Hoebel, B. G. 1973. Overeating and obesity from damage to a noradrenergic system in the brain. *Science* 182:166–68
11. Knoll, J. 1979. Satietin: a highly potent anorexogenic substance in human serum. *Physiol. Behav.* 23:497–502
12. Sabelli, H. C., Mosnaim, A. D. 1974. Phenylethylamine hypothesis of affective behavior. *Am. J. Psychiatry* 131:695–99
13. Paul, S. M., Hulihan-Giblin, B., Skolnick, P. 1982. (+) Amphetamine binding to rat hypothalamus: Relation to anorexic potency of phenylethylamines. *Science* 218:487–90
14. Blundell, J. E. 1977. Is there a role for serotonin (5-hydroxy-tryptamine) in feeding? *Int. J. Obesity,* 1:15–42
15. Hoebel, B. G. 1977. Pharmacologic control of feeding. *Ann. Rev. Pharmacol. Toxicol.* 17:605–21
16. Leibowitz, S. F. 1980. Nuerochemical systems of the hypothalamus in control of feeding and drinking behavior and water and electrolyte excretion. In *Handbook of the Hypothalamus,* ed. P. Morgane, J. Panksepp, 3:299–437. New York: Decker
17. Gibbs, J., Young, R. C., Smith, G. P. 1973. Cholecystokinin decreases food intake in rats. *J. Comp. Physiol. Psychol.* 84:488–95
18. Stern, J. J., Cudillo, C. A., Kruper, J. 1976. Ventromedial hypothalamus and short-term feeding suppression by caerulein in male rats. *J. Comp. Physiol. Psychol.* 90:484–90
19. Gibbs, J., Falasco, J. D., McHugh, P. R. 1976. Cholecystokinin elicits satiety in rats with open gastric fistulas. *Nature* 245:323–25
20. Gibbs, J., Smith, G. P. 1977. Cholecystokinin and satiety in rats and rhesus monkeys. *Am. J. Clin. Nutr.* 30:758–61
21. Antin, J., Gibbs, J., Holt, J., Young, R. C., Smith, G. B. 1975. Cholecystokinin elicits the complete behavioral sequence of satiety in rats. *J. Comp. Physiol. Psychol.* 89:784–90
22. Mueller, K., Hsiao, S. 1977. Specificity of cholecystokinin satiety effect: reduction of food but not water intake. *Pharm. Biochem. Behav.* 6:643–46
23. Innis, R. B., Correa, F.M.A., Ukl, G. R., Schneider, S. H. 1979. Cholecystokinin octapeptide-like immunoreactivity: histochemical localization in rat brain. *Proc. Natl. Acad. Sci. USA* 76:521–25
24. Della-Ferra, M. A., Baile, C. A. 1980. CCK-oktapeptide injected in CSF decreases meal size and daily food intake in sheep. *Peptides* 1:51–54
25. Freed, W. J., Perlow, M. J., Wyatt, R. J. 1979. Calcitonin: inhibitory effect on eating in rats. *Science* 206:850–52
26. Knoll, J. 1980. The anorexogenic effect of satietin in comparison to the effects of calcitonin, cholecystokinin and pGlu-His-GlyOH. *Neurosci. Letts. Suppl.* 5:317
27. Perlow, M. J., Freed, W. J., Carman, J. S., Wyatt, R. J. 1980. Calcitonin reduces feeding in man, monkey and rat. *Pharm. Biochem. Behav.* 12:609–12
28. Levine, A. S., Morley, J. E. 1981. The effect of prostaglandins (PGE_2 and $PGF_2\alpha$) on food intake in rats. *Pharmacol. Biochem. Behav.* 15:735–38
29. Rizzo, A. J., Goltzman, D. 1981. Calcitonin receptors in the central nervous system of the rat. *Endocrinology* 108:1672–77
30. Gibbs, J., Fauser, D. J., Rowe, E. A.,

Rolls, B. J., Rolls, E. T. Madison, S. P. 1979. Bombesin suppresses feeding in rats. *Nature* 282:208–10

31. Anastasi, A., Erspamer, V., Bucci, M. 1971. Isolation and structure of bombesin and alytesin, two analogous active peptides from the skin of the European amphibians Bombina and Alytes. *Experientia* 27:166–67
32. Vijayan, E., McCann, S. M. 1977. Suppression of feeding and drinking activity in rats following intraventricular injection of thyrotropin releasing hormone. *Endocrinology* 100:1727–30
33. Morley, J. E., Levine, A. S., Prasad, C. 1981. Histidylproline diketopiperazine decreases food intake in rats. *Brain. Res.* 210:475–78
34. Morley, J. E., Levine, A. S. 1982. Corticotropin releasing factor, grooming and ingestive behavior. *Life Sci.* 31:1459–60
35. Hoebel, B. G., Hernandez, L., McLean, S. 1982. Catecholamines, enkephalin and neurotensin in feeding and reward. In *The Neuronal Basis of Feeding and Reward*, ed. B. G. Hoebel, D. Novin, pp. 465–78. Brunswick, Mass: Haer Inst.
36. Lotter, E. C., Krinsky, R., McKay, J. M., Trenner, C. M., Porte, D. Jr., Woods, C. S. 1981. Somatostatin decreases food intake of rats and baboons. *J. Comp. Physiol. Psychol.* 95:278–87
37. Schally, A. V., Redding, R. W., Lucien, H. W., Meyer, J. 1967. Enterogastrone inhibits eating by fasted rats. *Science* 157:210–11
38. Malaisse-Lagae, F., Carpentier, J. L., Patel, Y. C., Malaisse, W. J., Orci, L. 1977. Pancreatic polypeptide: a possible role in the regulation of food intake in the mouse. Hypothesis. *Experientia* 33:915–18
39. Woods, S. C., West, D. B., Stein, L. J., McKay, L. D., Lotter, E. C., Porte, S. G., Kenney, N. J., Porte, D. Jr. 1981. Peptides and the control of meal size. *Diabetologia* 20:305–13
40. Reichelt, K. L., Foss, I., Trygsted, O., Edmison, P. D., Johansen, J. H., Boler, J. B. 1978. Humoral control of appetite. II. Purification and characterization of an anorexogenic peptide from human urine. *Neuroscience* 3:1207–11
41. Nance, D. M., Coy, D. H., Kastin, A. J. 1979. Experiments with a reported anorexogenic tripeptide: Pyro-Glu-His-Gly-OH. *Pharmacol. Biochem. Behav.* 11:733–35
42. Schulman, J. L., Carleton, J. L., Whitney, G., Whitehorn, J. C. 1957. Effect of glucagon on food intake and body weight in man. *J. Appl. Physiol.* 11:419–21
43. Pennick, S. D., Hinkle, L. E. Jr., 1961. Depression of food intake induced in healthy subjects by glucagon. *New Engl. J. Med.* 264:893–97
44. Salter, J. M. 1960. Metabolic effects of glucagon in the Wistar rat. *Am. J. Clin. Nutr.* 8:535–39
45. Martin, J. R., Novin, D. 1977. Decreased feeding in rats following hepatic-portal infusion of glucagon. *Physiol. Behav.* 19:461–66
46. Margolis, R. U., Altszuler, N. 1967. Insulin in the cerebrospinal fluid. *Nature* 215:1375–76
47. Owen, O. E., Reichard, G. A. Jr., Boden, G., Shuman, C. R. 1974. Comparative measurements of glucose, betahydroxybutyrate, acetoacetate, and insulin in blood and cerebrospinal fluid during starvation. *Metabolism* 23:7–14
48. Woods, S. C., Porte, D. Jr. 1978. The central nervous system, pancreatic hormone, feeding and obesity. *Adv. Metab. Dis.* 9:282–312
49. Anika, S. M., Houpt, T. R., Houpt, K. A. 1980. Insulin as a satiety hormone. *Physiol. Behav.* 25:21–23
50. Wanderweele, D. A., Pi-Sunyer, F. X., Novin, D., Bush, M. J. 1980. Chronic insulin infusion suppresses food ingestion and body weight gain in rats. *Brain Res. Bull. 5, Suppl 4*:7–11
51. Woods, S. C., Lotter, E. C., McKay, L. D., Porte, D. Jr. 1979. Chronic intracerebroventricular infusion of insulin reduces food intake and body weight in baboons. *Nature* 282:503–5
52. Sakata, T., Oomura, Y., Fukushima, M., Tsutsui, K., Hashimoto, K., Kuhara, T. T., Matsumoto, I. 1980. Circadian and long term variation of certain metabolites in fasted rats, implications. *Brain. Res. Bull.* 5 (Suppl. 4):23–28
53. Oomura, Y. 1981. Chemosensitive neuron in the hypothalamus related to food intake behavior. *Japan. J. Pharmacol.* 31 (Suppl. 1):1P–12P
54. Oomura, Y., Shimizu, N., Inokuchi, A., Sakata, T., Arase, K., Fujimoto, M., Fukushima, M., Tsutsui, K. 1982. Regulation of feeding by blood borne hunger and satiety substances through glucose-sensitive neurons. *12th Annual Meeting of Society for Neuroscience*, Vol. 8, Part 1. Minneapolis, Minn. Abstr. 75.10
55. Horton, E. W. 1964. Actions of prostaglandins E_1, E_2 and E_3 on the central

nervous system. *Br. J. Pharmacol. Chemother.* 22:189–92
56. Scaramuzi, O. E., Baile, C. A., Mayer, J. 1971. Prostaglandins and food intake of rats. *Experientia* 27:256–57
57. Baile, C. A., Simpson, C. W., Bean, S. M., McLaughlin, C. L., Jacobs, H. L. 1973. Prostaglandins and food intake of rats: A component of energy balance regulation? *Physiol. Behav.* 10:1077–85
58. Doggett, N. S., Jawaharlal, K. 1977. Anorectic activity of prostaglandin precursors. *Br. J. Pharmacol.* 60:417–23
59. Tatemoto, K. 1982. Neuropeptide Y: Complete amino acid sequence of the brain peptide. *Proc. Natl. Acad. Sci. USA* 79:5485–89
60. Emson, P. C., Dequidt, M. E. 1984. NPY—A new member of the pancreatic polypeptide family. *Trends Neurosci.* 7:31–35
61. Morley, J. E., Levine, A. S. 1985. The pharmacology of eating behavior. *Ann. Rev. Pharmacol. Toxicol.* 25:127–46
62. Smith, G. P. 1984. Gut hormone hypothesis of postprandial satiety. In *Eating and Its Disorders,* ed. A. J. Stunkard, E. Stellar, pp. 67–75. New York: Raven
63. Morley, J. E., Levine, A. S. 1983. The central control of appetite. *Lancet* 1:398–401
64. Morley, J. E., Gosnell, B. A., Levine, A. S. 1984. The role of peptides in feeding. *Trends Pharmacol. Sci.* 5:468–71
65. Knoll, J. 1969. *Theory of Active Reflexes.* Budapest: Akademiai Kiado
66. Knoll, J. 1986. Satietins: a family of highly potent and selective anorectic glycoproteins in human and mammalian blood. In *Endogenous Anorectics,* ed. B. Knoll, J. Nagy, J. Timar, pp. 233–69. Budapest: Akademiai Kiado
67. Knoll, J. 1978. New aspects in the chemoregulation of food intake. *Neurosci. Letts. Suppl.* 1:S55
68. Knoll, J. 1980. Highly selective peptidechalones in human serum. A concept on the control of physiological functions by blood-borne selective inhibitors of neurochemical transmission. In *Modulation of Neurochemical Transmission,* ed. E. S. Vizi, pp. 97–125, Budapest: Akademiai Kiado
69. Knoll, J. 1982. Satietin: endogenous regulation of food intake. In *Regulatory Peptides: From Molecular Biology to Function,* eds. E. Costa, M. Trabucchi, pp. 501–9, New York: Raven
70. Knoll, J. 1982. Anorectic agents and satietin, an endogenous inhibitor of food intake. In *Advances in Pharmacology and Therapeutics II,* Vol. 1. Ed. H. Yoshida, Y. Hagihara, S. Ebashi, pp. 147–62. Oxford/New York: Pergamon
71. Knoll, J. 1982. Satietin: a centrally acting potent anorectic substance with a long-lasting effect in human and mammalian blood. *Pol. J. Pharmacol. Pharm.* 34:3–16
72. Nagy, J., Kalasz, H., Knoll, J. 1982. An improved method for the preparation of highly purified satietin samples from human serum. *Pol. J. Pharmacol. Pharm.* 34:47–52
73. Nagy, J., Kalasz, H., Knoll, J. 1983. Isolation and characterization of a highly selective anorexogenic substance by chromatography and electrophoresis. In *Chromatography and Mass Spectrometry in Biological Sciences 2,* ed. A. Frigerio, pp. 421–32. Amsterdam: Elsevier
74. Knoll, J. 1984. Satietin: a 50,000 dalton glycoprotein in human serum with potent, long-lasting and selective anorectic activity. *J. Neural. Transm.* 59:163–94
75. Knoll, J. 1987. Satietins, α_1-glycoproteins in human plasma with potent, long-lasting and selective anorectic activity. *Medicinal Res. Rev.* 7:107–44
76. Nagy, J., Meszaros, M., Csizer, E., Knoll, J. 1986. Isolation and chemical analysis of human satietins. See Ref. 66, pp. 271–85
77. Varady, L., Nagy, J., Knoll, J. 1986. Electrophoretic studies on human satietins. See Ref. 66, pp. 287–92
78. Knoll, J. 1985. Satietin, a potent and selective endogenous anorectic glycoprotein. In *Psychopharmacology and Food,* ed. M. Sandler, T. Silverstone, pp. 110–29. Oxford/New York/Tokyo: Oxford Univ. Press
79. Knoll, J. 1985. Satietin, a blood-borne glycoprotein in the regulation of food intake. In *Endocoids,* ed. H. Lal, F. Labella, J. Lane, pp. 515–22. New York: Liss
80. Nagy, J., Mazsaroff, I., Varady, L., Knoll, J. 1985. Studies on the purification and properties of satietin, an anorexoginic glycoprotein of biological origin. In *Chromatography, The State of the Art,* ed. H. Kalasz, L. S. Ettre, pp. 337–58. Budapest: Akademiai Kiado
81. Nagy, J., Varady, L., Kalasz, H., Knoll, J. 1984. Molecular weight measurement of human satietin. *J. Chromatog.* 317:165–72
82. Mazsaroff, I., Varady, L., Nagy, J., Knoll, J. 1986. Subsequent analysis to proteolytic digestion of satietin-D. See Ref. 80, pp. 527–32.
83. Harmath, S., Barna, I., Knoll, J. 1986. Isolation of satietin-D by immu-

noadsorbent chromatography and the immunochemical detection of satietins in human plasma. In *Endogenous Anorectics*, eds. B. Knoll, J. Nagy, J. Timar, pp. 293–300. Budapest: Akademiai Kiado

84. Knoll, J. 1984. Satietin: a blood-borne, highly selective and potent anorectic glycoprotein. In *Biomedical Significance of Peptide Research*, ed. F. A. Laszlo, F. Antoni, pp. 103–14. Budapest: Akademiai Kiado
85. Knoll, J. 1984. Satietin, a blood-borne anorectic glycoprotein, as the putative rate-limiting satiety signal in the negative feedback of food intake. *Zeitschr. Ernährungswissenschaft* 23:85–103
86. Novin, D., Wyrwicka, W., Bray, G. A. 1976. *Hunger: Basic Mechanisms and Clinical Implications*. New York: Raven
87. Silverstone, T. 1976. *Appetite and Food Intake*. Berlin: Dahlem Konf.
88. Wayner, M. J., Oomura, Y., eds. 1975. *Central Neural Control of Eating and Obesity. Pharm. Biochem. Behav. Suppl. 1*.
89. Sandor, G., Knoll, J. 1985. The time structure of the anorectic effect of satietin. *Physiol. Behav.* 34:851–53
90. Blundell, J. E., Latham, C. J., Leshem, M. B. 1976. Differences between the anorexic actions of amphetamine and fenfluramine—possible effects on hunger and satiety. *J. Pharm. Pharmacol.* 25:471–77
91. Knoll, B., Timar, J., Knoll, J. 1982. Comparison of the effect of satietin, amphetamine and fenfluramine in raphe lesioned rats. *Neurosc. Letts. Suppl. 10* S. 269
92. Grandison, S., Guidotti, A., 1977. Stimulation of food intake by muscimol and beta-endorphin. *Neuropharmacology* 16:533–36
93. Morley, J. E., Levine, A. S., Kneip, J. 1981. Muscimol-induced feeding: a model to study the hypothalamic regulation of appetite. *Life Sci.* 29:1213–18
94. Margules, D. L., Moisset, B., Lewis, M. J., Shibuya, J., Pert, C. B. 1978. Beta-endorphin is associated with overeating in genetically obese mice (ob/ob) and fa/fa). *Science* 202:988–91
95. Gambert, S. R., Garthwaite, T. L., Pontzer, C. H., Hagen, T. C. 1980. Fasting associated with decrease in hypothalamic beta-endorphin. *Science* 210:1271–72
96. Marks-Kaufman, R., Kanarek, R. B. 1980. Morphine selectively influences macronutrient uptake in the rat. *Pharm. Biochem. Behav.* 12:427–30
97. Sanger, D. J. 1981. Endorphinergic mechanism in the control of food and water intake. *Appetite* 2:193–208
98. Morley, J. E., Levine, A. S. 1982. The role of the endogenous opiates as regulators of appetite. *Am. J. Clin. Nutr.* 35:757–61
99. Holtzman, S. G. 1974. Behavioral effects of separate and combined administration of naloxone and d-amphetamine, *J. Pharm. Exp. Ther.* 189:51–60
100. Atkinson, R. L. 1982. Naloxone decreases food intake in obese humans. *J. Clin. Endocrinol. Metab.* 55:196–98
101. Kyriakides, M., Silverstone, T., Jeffcoate, W., Laurence, B. 1980. Effect of naloxone on hyperphagia in Prader-Willi syndrome. *Lancet* i:876–77
102. Thompson, D. A., Welle, S. L., Lilavivat, U., Pinecand, L., Campbell, R. G. 1982. Opiate receptors blockade in man reduces 2-deoxy-d-glucose-induced food intake but not hunger, thirst and hypothermia. *Life Sci.*31:849–52
103. Sternbach, H. A., Annitto, W., Pottash, A.L.C., Gold, M. S. 1982. Anorexic effect of naltrexone in man. *Lancet* i:388–89
104. Atkinson, R. L. 1983. Naltrexone for weight loss in obesity. *Abstr. 4th Int. Congr. Obesity* 30A:81
105. Morley, J. E. 1983. Neuroendocrine effects of endogenous opioid peptides in human subjects: A review. *Psychoneuroendocrinology*. 8:361–79
106. Knoll, J. 1984. Anorectic agents and endogenous substances. In *Proc. Second World Conf. Clinical Pharmacol. Therapeutics*, ed. L. Lemberger, M. M. Reidenberg, pp. 103–127. Bethesda: Am. Soc. Pharmacol. Exp. Ther.
107. Knoll, J., Knoll, B. 1964. The cumulative nature of the reserpine effect and the possibilities of inhibiting cumulation pharmacologically. *Arch. int. Pharmacodyn.* 148:200–16
108. Knoll, B., Held, K., Knoll, J. 1974. Rapid screening of drug action on learning and memory. In *Symposium on Pharmacology of Learning and Retention*, ed. B. Knoll, pp. 43–47. Budapest: Akademiai Kiado
109. Knoll, J., Knoll, B. 1958. Methode zur Untersuchung der spezifisch depressiven Wirkung von "Tranquilizern" auf das Zentralnervensystem. *Arzneimittel-Forsch.* 8:330–33
110. Beach, F. A. 1944. Relative effect of androgens upon the mating behavior of male rats subjected to forebrain injury or castration. *Exp. Zool.* 97:249–59
111. Knoll, B., Knoll, J. 1982. The selectivity of the anorectic effect of satietin. I.

The ineffectiveness of satietin in behavioral tests. *Pol. J. Pharmacol. Pharm.* 34:17–23
112. Yen, T. T., Dallo, J., Knoll, J. 1982. The selectivity of the anorectic effect of satietin. IV. The ineffectiveness of satietin on the sexual performance of male CFY rats. *Pol. J. Pharmacol. Pharm.* 34:41–45
113. Issekutz, B., Issekutz, B. Jr. 1942. Einfache Apparate zur Messung des Sauerstoff-Verbrauches von Versuchstieren. *Arch. Exp. Pathol. Pharmacol.* 199: 306–11
114. Timar, J., Knoll, J. 1982. The selectivity of the anorectic effect of satietin. III. The ineffectiveness of satietin on metabolic rate, body temperature and blood pressure. *Pol. J. Pharmacol. Pharm.* 34:33–39
115. Knoll, B., Timar, J., Knoll, J. 1986. The selectivity of the anorectic effect of satietin-D. In *Endogenous Anorectics*, eds. B. Knoll, J. Nagy, J. Timar, pp. 309–15. Budapest: Akademiai Kiado
116. Sandor, G., Knoll, J. 1982. The selectivity of the anorectic effect of satietin. II. Ineffectiveness of satietin on the water intake of food deprived rats. Effect of satietin in "conditioned aversion" paradigm. *Pol. J. Pharmacol. Pharm.* 34:25–32
117. Gyarmati, S., Foldes, J., Koranyi, L., Knoll, B., Knoll, J. 1985. The anorectic effect of satietin is unrelated to carbohydrate metabolism. *Physiol. Behav.* 34:167–70