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Metergoline Potentiates Natural Feeding and Antagonizes the Anorectic Action of Medial Hypothalamic 5-Hydroxytryptamine

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CURRIE, P. J. AND D. V. COSCINA. *Metergoline potentiates natural feeding and antagonizes the anorectic action of medial hypothalamic 5-hydroxytryptamine.* PHARMACOL BIOCHEM BEHAV **53**(4) 1023–1028, 1996. – Medial hypothalamic injections of 5-hydroxytryptamine (5-HT) or its agonists have been reported to inhibit feeding elicited by norepinephrine (NE), suggesting that these two transmitter systems interact antagonistically in the control of ingestive behavior. The present study was designed to directly test the hypothesis that 5-HT inhibits adrenergic feeding, specifically at the onset of the rat's nocturnal eating cycle. Free-feeding animals were injected with 5-HT (5–20 nmol) immediately before NE (20 nmol) and food intake was measured 1.5 h postinjection. In separate groups of rats, the serotonergic antagonist metergoline (MET) (2.5–20 nmol) was injected into the paraventricular nucleus (PVN) immediately before 5-HT, or before combined injections of 5-HT and NE. The feeding-stimulant action of MET alone, injected IP (0.25–2 mg/kg) or centrally (2.5–40 nmol), was also examined. Results indicated that administration of 5-HT into the PVN suppressed dark onset feeding and dose-dependently blocked NE-stimulated eating. Pretreatment with MET attenuated the inhibitory action of 5-HT on feeding, and reversed the serotonergic blockade of the adrenergic eating response. Further, systemically injected MET significantly increased dark onset feeding, whereas PVN injections failed to alter food intake reliably. These findings provide the first direct evidence that serotonergic and adrenergic systems within the PVN interact in a competitive manner to modulate the natural high rates of feeding displayed by rats during the early dark period. Although MET effectively blocked the anorectic effect of 5-HT, the feeding-stimulant action of this compound alone does not appear to be mediated within the PVN.

Metergoline 5-Hydroxytryptamine Norepinephrine Paraventricular nucleus Food intake

RECENT indirect evidence suggests that brain 5-hydroxytryptamine (5-HT) may exert its effects on natural feeding patterns, specifically by opposing the action of norepinephrine (NE) to potentiate food intake at the onset of the nocturnal cycle. Endogenous levels of both neurotransmitters in the medial hypothalamic paraventricular nucleus (PVN) exhibit a circadian rhythm with peak concentrations evident during the early dark period (25,27,36,37). The peak in extracellular levels of NE is associated with an increase in α_2-adrenergic receptor density and an increase in circulating corticosterone, a hormone which is critical for the expression of adrenergic feeding (16). Moreover, exogenous administration of NE is most effective in stimulating eating and carbohydrate appetite at the beginning of the dark phase (8,38). Similarly, 5-HT elicits its most potent anorectic effects when injected into the PVN at this particular time (22,23).

Indirect support for the hypothesis that medial hypothalamic 5-HT suppresses ingestion at dark onset through an inhibitory interaction with NE is also derived from previous work showing that PVN administration of 5-HT or its agonists during the mid-light cycle suppresses feeding, and specifically, eating stimulated by NE (13,14,41). This effect is reversed by pretreatment with the serotonergic antagonist metergoline (MET) (41) which, when injected systemically, has been found to increase food intake (9,11) and to inhibit the anorectic action of *d*-fenfluramine (15,32). Intraperitoneal MET injec-

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tions have further been shown to potentiate carbohydrate ingestion selectively (35) and to exaggerate the natural tendency of the rat to consume this macronutrient at dark onset (17).

The following experiments were designed to provide direct tests of the hypothesis that serotonergic and adrenergic receptor mechanisms within the paraventricular nucleus interact antagonistically to control ingestive behavior during the early dark period. This was accomplished by examining the dose-response effectiveness of 5-HT to inhibit eating elicited by NE at this time. Additional studies were conducted to characterize the impact of MET on spontaneously motivated feeding occurring at the start of the nocturnal cycle and to determine its ability to reverse the anorectic action of 5-HT on NE-stimulated eating.

METHODS

Animals

Adult male Sprague-Dawley rats (Charles River, St.-Constant, Quebec, Canada), weighing 275-300 g at the time of surgery, were individually housed in hanging wire-mesh cages. Rats had free access to Purina lab chow pellets (Bio-Serv, Frenchtown, NJ) and water. The animal colony room was maintained on a 12 L : 12 D cycle (lights on at 0700 h) and at a temperature of 22 ± 2°C.

Surgery

Rats were anesthetized with pentobarbital sodium (50 mg/kg, IP; Somnotol) and placed in a stereotaxic frame with the incisor bar set 3.6 mm below the interaural line. Coordinates for the guide cannula relative to bregma were AP −1.5 mm, L −0.3 mm, and V −4.5 mm (28). Guide cannulae (22 ga; Plastics One, Roanoke, VA) were implanted 4 mm dorsal to the PVN. Implants were secured with acrylic cement and three stainless-steel screws which penetrated the skull. A 28-ga stainless-steel stylet was used to maintain cannula patency. Behavioral testing began after a 1-week postoperative recovery period. During this time, each rat was handled frequently and received several mock injections.

Drugs

Norepinephrine bitartrate and 5-HT maleate were purchased from Sigma Chemical Co. (St. Louis, MO). Metergoline was generously supplied by Farmitalia (Milan, Italy). NE and 5-HT were dissolved in sterile physiologic (0.9%) saline, and MET was dissolved in 1% tartaric acid. Drugs were injected into the PVN using a 28-ga microinjection assembly (Plastics One) attached to a 5-μl Hamilton syringe (Bonaduz, Switzerland). The injector cannula extended 4 mm beyond the indwelling guide cannula. A volume of 0.4 μl was injected over a period of 1 min with the injector cannula left in place for an additional 30 s to permit drug diffusion.

Procedure

Experiment 1. Separate groups of rats were used to examine the feeding-suppressive effects of 5-HT (n = 12), the feeding-stimulatory action of NE (n = 15), and the combined impact of NE and 5-HT (n = 8) on dark onset feeding. Food pellets were removed from home cages 10 min before the start of testing and weighed. 5-HT (1.3-20 nmol) or saline, and NE (5-40 nmol) or saline were injected into the PVN immediately before the start of the dark period. Under conditions requiring combined NE and 5-HT treatment, PVN infusion of 5-HT (5-20 nmol) was followed by NE (20 nmol) injection, and food intake measurements were taken 1.5 h after drug injection. Any spillage, although minimal during this brief test period, was collected from beneath the cage and added to the unconsumed total. Drug or vehicle injections were administered in randomized order, and a minimum of 3 drug-free days separated successive tests. Fresh food was provided daily approximately 5 h before lights out to minimize disturbance to animals at the onset of the dark period. Similar test procedures were carried out in each of the experiments which followed.

Experiment 2. This study examined the impact of MET on the serotonergic inhibition of dark onset feeding and assessed the ability of MET to reverse the suppression of NE-stimulated eating elicited by 5-HT. All compounds were injected directly into the PVN. To examine the potential blockade of 5-HT-induced anorexia following MET administration (n = 12), the serotonergic antagonist (2.5-20 nmol) or its vehicle was injected 10 min before 5-HT (10 nmol) or saline. In an additional group of rats (n = 10) induced to eat with NE, MET (5-20 nmol) or vehicle was infused 10 min before 5-HT (10 nmol) or saline followed by NE (20 nmol) or saline. Injections of 5-HT and NE were administered immediately before the onset of the nocturnal cycle and food intake, corrected for spillage, was measured 1.5 h later.

Experiment 3. The effects of systemic and PVN injections of MET on dark onset feeding were examined. MET was administered IP (n = 16) immediately before the onset of the nocturnal cycle, at doses ranging from 0.25-2 mg/kg or vehicle. In a separate group of rats (n = 10), the serotonergic antagonist was infused into the PVN at doses of 2.5-40 nmol. After systemic or central drug treatment, rats were returned to their cages with preweighed food and intakes were measured 1.5 h postinjection.

Histology and Statistics

Following the completion of these experiments, histologic verification of cannula placements was performed. Rats were anesthetized with Somnotol and perfused transcardially with saline followed by 10% buffered formalin. The brains were extracted and stored in formalin for at least 7 days before 40-μm frozen coronal sections were cut, mounted on glass slides, and stained with Cresyl violet. Sections were viewed

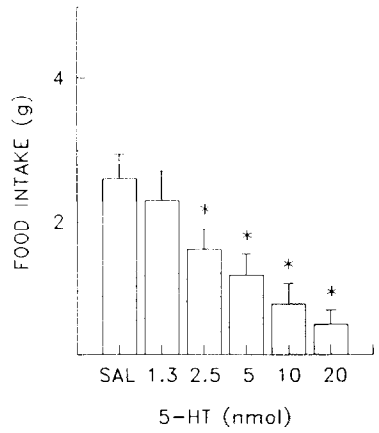

FIG. 1. Feeding-suppressive action of 5-HT administered into the PVN at the onset of the nocturnal cycle (n = 12). Data were analyzed by ANOVA for repeated-measures and Tukey tests (*$p < 0.05$ compared to intake after saline injection).

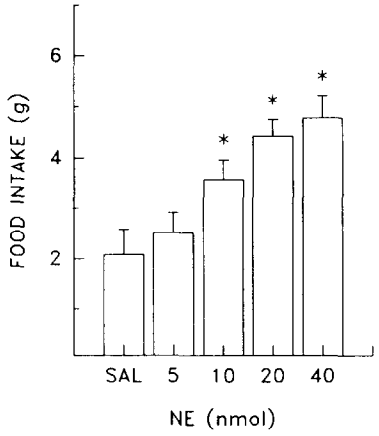

FIG. 2. Eating-stimulatory action of NE injected into the PVN (n = 15). Statistical significance was determined using repeated-measures ANOVA and posthoc Tukey tests (*p < 0.05 compared to saline).

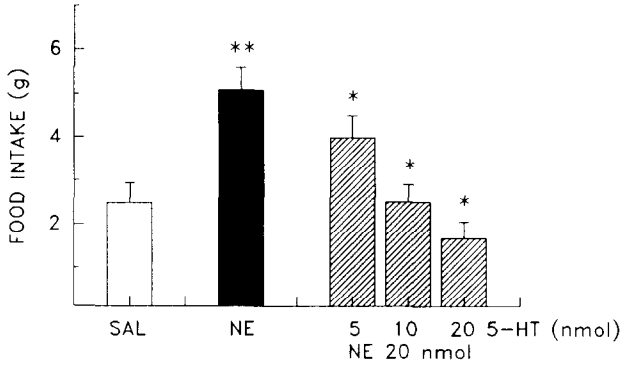

FIG. 3. Effects of 5-HT on eating stimulated by injection of NE into the PVN at the start of the active feeding cycle (n = 8). Statistical differences between treatments were determined by repeated-measures ANOVA and posthoc Tukey tests of individual mean comparisons (*p < 0.05 compared to food intake following injection of NE; **p < 0.05 for comparison between NE and saline).

relative to the stereotaxic atlas of Paxinos and Watson (28). All rats reported in these studies were found to have injector needle tracts extending into the dorsomedial region of the PVN.

Statistical evaluations were based on one-way analyses of variance (ANOVA) for repeated measures followed by individual mean comparisons using posthoc Tukey tests. For all statistical tests, the α level was set at $p < 0.05$.

RESULTS

The effects of PVN injections of 5-HT and NE on food ingestion occurring at the onset of the nocturnal cycle are shown in Figs. 1 and 2. ANOVA confirmed that treatment with 5-HT evoked a dose-dependent reduction in dark onset feeding [$F(5, 66) = 6.4, p < 0.001$], yielding a maximal suppressive effect in excess of 80%. Even a low dose of 5 nmol suppressed intake by 64%. In contrast, NE administration significantly increased feeding behavior [$F(4, 70) = 10.9, p < 0.001$], resulting in an over twofold increase in intake compared to saline. When 5-HT was infused into the PVN immediately before NE, a dose-dependent reduction in NE-stimulated eating ranging from 22–66% was evident [$F(4, 35) = 21.2, p < 0.001$] (Fig. 3).

As shown in Fig. 4, the serotonergic-induced suppression of dark onset feeding was significantly attenuated by MET pretreatment [$F(5, 66) = 18.8, p < 0.001$]. Similarly, MET injected into the PVN also antagonized the inhibitory impact of 5-HT on NE-stimulated eating [$F(5, 54) = 11.8, p < 0.001$] (Fig. 5). In each case, the effect of MET was dose dependent, achieving at the highest dose a near-total blockade of 5-HT-induced anorexia. As shown below, these same doses of MET failed reliably to alter baseline food intake when injected alone.

Figure 6 compares, as a function of dose, the feeding responses to IP and PVN injections of MET over the 1.5 h following dark onset injection. Although systemic administration of MET produced a significant increase in food intake [$F(4, 75) = 10.6, p < 0.001$], PVN injection of the serotonergic antagonist did not reliably alter food intake at any of the doses tested. With respect to IP administration, a maximal effect on food intake was found at the 1 mg/kg dose, whereas treatment with the highest dose of 2 mg/kg failed reliably to alter food intake compared to baseline. Because enhanced feeding was only evident following IP injection of 0.5–1 mg/kg, this suggests that the eating-stimulatory effects of MET are found within a very narrow dose range.

DISCUSSION

Previous work has provided compelling evidence that an increase in serotonergic function decreases food intake, suggesting an inhibitory role of this monoamine in the control of ingestive behavior (3,6,21,31,34). It has also been proposed that endogenous 5-HT acts within the medial hypothalamus to suppress ingestion through its inhibitory interaction with NE (20,41) and, more recently, that this antagonistic interaction occurs during the early dark period (20). The results of the present study support the hypothesis that 5-HT acts on satiety mechanisms to inhibit NE-stimulated eating by demonstrating a particularly strong antagonism between these two aminergic systems within the PVN on natural feeding behavior. Specifically, at the beginning of the dark period, PVN injections of NE increased eating, whereas 5-HT at low doses

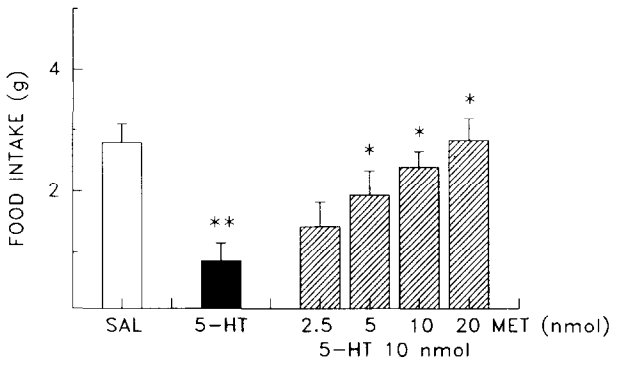

FIG. 4. Effects of PVN injection of MET on the serotonergic inhibition of dark-onset feeding (n = 12). Statistical significance between treatments was determined by repeated-measures ANOVA and Tukey tests (*p < 0.05 compared to intake after injection of 5-HT; **p < 0.05 for comparison between 5-HT and saline injections).

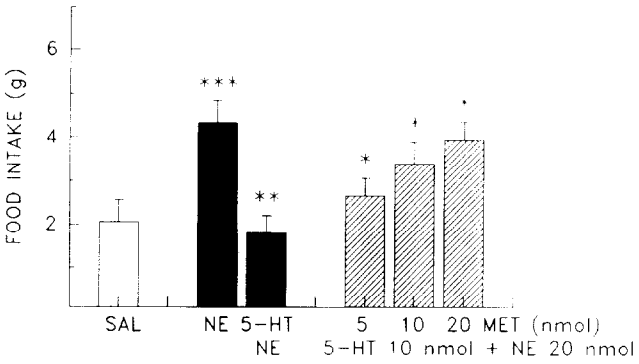

FIG. 5. Blockade of the serotonergic inhibition of NE-stimulated eating following PVN injection of MET (n = 10). Data were analyzed by ANOVA for repeated measures followed by posthoc Tukey tests (*$p < 0.05$ compared to intake after coinjection of 5-HT and NE; **$p < 0.05$ compared to NE; ***$p < 0.05$ compared to saline).

inhibited food intake. When both monoamines were coinjected, 5-HT antagonized NE-induced feeding. The inhibitory effect on the adrenergic ingestive response was dose-dependent.

Although 5-HT effectively suppressed spontaneous feeding behavior occurring at the start of the nocturnal cycle as well as eating stimulated by NE, pretreatment with the 5-$HT_{1/2}$-receptor antagonist MET, injected into the PVN, attenuated the anorectic action of 5-HT under both conditions. Autoradiographic studies have demonstrated the existence of serotonergic receptors including 5-HT_{1B} receptor binding sites in the medial hypothalamus (19,29,30), which may provide the neural substrate for 5-HT's impact on feeding behavior. The medial hypothalamus is densely innervated by both adrenergic and serotonergic neurons, particularly within the parvocellular region of the PVN, which is believed to be important in the control of feeding (18,33). In addition, biochemical evidence indicates that an antagonistic interaction exists between adrenergic and serotonergic systems in the hypothalamus with the activity of adrenergic neurons and release of NE inhibited by 5-HT (2,40). These findings may provide further support for the proposed antagonistic interaction between adrenergic and serotonergic systems in the control of food intake through their action on medial hypothalamic satiety mechanisms.

Circadian variations in medial hypothalamic neurotransmitter release and activity, receptor density, and/or affinity, as well as in metabolic processes, are all believed to be closely related to expressions of natural feeding behavior and to variations in feeding responses resulting from exogenous neurotransmitter administration (20,23,38). Feeding which occurs at the onset of the dark phase, for example, is associated with low levels of plasma glucose, increased carbohydrate utilization, and increased activity of adrenergic and serotonergic systems (1,16,24,36,37). In the present study, injection of NE at the start of the dark cycle evoked a robust and dose-dependent increase in food intake, whereas low doses of 5-HT suppressed dark onset feeding and eating elicited by adrenergic stimulation. The anorectic action of 5-HT was impeded by treatment with the serotonergic antagonist MET which, when injected alone into the PVN, had no impact on feeding behavior. As such, these data provide the first direct evidence in support of an antagonistic interaction between medial hypothalamic adrenergic and serotonergic systems controlling food intake during the early dark period. The data are also in agreement with one previous report in which MET injected into the PVN, during the mid-light cycle, antagonized the serotonergic suppression of NE-induced feeding (41), although in the present study considerably lower doses of MET were effective in evoking a near-total blockade of this effect.

Natural feeding behavior in the rat follows a circadian rhythm with a peak in eating occurring at the beginning of the nocturnal cycle (1,7,39). In the current study, systemic injections of the serotonergic antagonist MET reliably enhanced feeding during the early dark period. This finding, while providing additional support for the hypothesis that 5-HT is involved in controlling food intake, specifically during the early portion of the dark cycle, is in line with previous results showing that peripheral administration of MET stimulates food intake in satiated rats when administered in the mid-light cycle (9,11), and with one additional report indicating that IP MET selectively potentiates carbohydrate appetite at dark onset (17).

The question of whether serotonergic antagonists stimulate ingestion through peripheral or central mechanisms has been previously discussed (5,11). When 5-HT is injected systemically, it does not cross the blood–brain barrier but elicits a potent suppression in food intake (10,12). This anorectic effect can be blocked by serotonergic antagonists (4,26), sug-

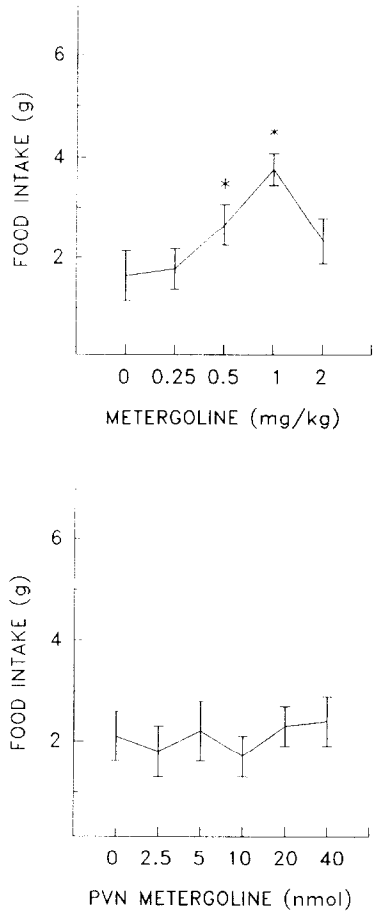

FIG. 6. Effects of IP (n = 16) and PVN (n = 10) injections of MET on dark-onset feeding. Significant treatment effects were determined by repeated-measures ANOVA followed by posthoc Tukey tests of mean comparisons (*$p < 0.05$ compared to saline).

gesting that the antagonists may be active at peripheral 5-HT receptors where changes in eating behavior are evoked.

With respect to central mechanisms of action, the PVN is considered to be an important brain site mediating the feeding-inhibitory effects of 5-HT and its agonists (20,21,41). However, in the current study, microinjection of MET into this medial hypothalamic nucleus did not stimulate feeding. Other evidence has shown that peripheral injections of serotonergic antagonists do not block the anorectic effects of 5-HT injected into the PVN (26). These findings suggest that the PVN is not critically involved in mediating the feeding stimulant-action of serotonergic antagonists. However, it remains possible that MET may act on other central sites to elicit its feeding effects, as recent work has demonstrated that intraventricular injections of this compound significantly increase eating behavior (5). Consequently, 5-HT antagonists may stimulate feeding by affecting serotonergic mechanisms in other brain regions, including other hypothalamic areas, or through an action on peripheral 5-HT systems.

In summary, the attenuation of adrenergic feeding by 5-HT and the reversal of this effect by MET provide direct evidence that these two transmitter systems may interact in an inhibitory or competitive manner to control ingestive behavior during the early portion of the rat's normally active feeding cycle. Although the results of the current study also extend previous research findings to indicate that systemically administered MET potentiates dark onset feeding, the inability of MET to elicit food intake following direct injection into the PVN suggests that its eating-stimulatory effects are not mediated within this nucleus. The lack of effect of MET on food intake following PVN microinjection is consistent with additional evidence demonstrating the underlying importance of multiple hypothalamic or extrahypothalamic 5-HT systems critically involved in such 5-HT-related changes in food intake (5, 13,14).

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REFERENCES

1. Armstrong, S. A. A chronometric approach to the study of feeding behavior. Neurosci. Biobehav. Rev. 4:27–53; 1980.
2. Blandina, P.; Goldfarb, J.; Walcott, J.; Green, J. P. Serotonergic modulation of the release of endogenous norepinephrine from rat hypothalamic slices. J. Pharmacol. Exp. Ther. 256:341–347; 1991.
3. Blundell, J. E. Serotonin and the biology of feeding. Am. J. Clin. Nutr. 55:155S–159S; 1992.
4. Carruba, M. O.; Mantegazza, P.; Memo, M.; Missale, C.; Pizzi, M.; Spano, P. F. Peripheral and central mechanisms of action of serotonergic anorectic drugs. Appetite 7(Suppl):105–113; 1986.
5. Coscina, D. V.; Feifel, D.; Nobrega, J. N.; Currie, P. J. Intraventricular but not intraparaventricular metergoline elicits feeding in satiated rats. Am. J. Physiol. 266:R1562–R1567; 1994.
6. Currie, P. J. Differential effects of NE, CLON, and 5-HT on feeding and macronutrient selection in genetically obese (ob/ob) and lean mice. Brain Res. Bull. 32:133–142; 1993.
7. Currie, P. J.; Coscina, D. V. Diurnal variations in the feeding response to 8-OH-DPAT injected into the dorsal or median raphe. NeuroReport 4:1105–1107; 1993.
8. Currie, P. J.; Wilson, L. M. Potentiation of dark onset feeding in obese mice (genotype ob/ob) following central injection of norepinephrine and clonidine. Eur. J. Pharmacol. 232:227–234; 1993.
9. Dourish, C. T.; Clark, M. L.; Fletcher, A.; Iversen, S. D. Evidence that blockade of postsynaptic 5-HT_1 receptors elicits feeding in satiated rats. Psychopharmacology 97:54–58; 1989.
10. Fletcher, P. J. The anorectic action of peripheral 5-HT examined in the runway: Evidence for an action on satiation. Psychopharmacology 93:498–501; 1987.
11. Fletcher, P. J. Increased food intake in satiated rats induced by the 5-HT antagonists methysergide, metergoline and ritanserin. Psychopharmacology 96:237–242; 1988.
12. Fletcher, P. J.; Burton, M. J. Microstructural analysis of the anorectic action of peripherally administered 5-HT. Pharmacol. Biochem. Behav. 24:1133–1136; 1986.
13. Fletcher, P. J.; Coscina, D. V. Injecting 5-HT into the PVN does not prevent feeding induced by injecting 8-OH-DPAT into the raphe. Pharmacol. Biochem. Behav. 46:487–491; 1993.
14. Fletcher, P. J.; Ming, Z. H.; Zack, M. H.; Coscina, D. V. A comparison of the effects of 5-HT_1 agonists TFMPP and RU 24969 on feeding following peripheral or medial hypothalamic injection. Brain Res. 580:265–272; 1992.
15. Goodall, E.; Silverstone, T. Differential effect of *d*-fenfluramine and metergoline on food intake in human subjects. Appetite 11: 215–228; 1988.
16. Jhanwar-Uniyal, M.; Roland C. R.; Leibowitz, S. F. Diurnal rhythm of α_2-noradrenergic receptors in the paraventricular nucleus and other brain areas: Relation to circulating corticosterone and feeding behavior. Life Sci. 38:473–482; 1986.
17. Leibowitz, S. F.; Alexander, J. T.; Cheung, W. K.; Weiss, G. F. Effects of serotonin and the serotonin blocker metergoline on meal patterns and macronutrient selection. Pharmacol. Biochem. Behav. 45:185–194; 1993.
18. Leibowitz, S. F.; Brown, L. L. Histochemical and pharmacological analysis of noradrenergic projections to the paraventricular hypothalamus in relation to feeding stimulation. Brain Res. 201: 289–314; 1980.
19. Leibowitz, S. F.; Jhanwar-Uniyal, M. 5-HT_{1A} and 5-HT_{1B} receptor binding sites in discrete hypothalamic nuclei: Relation to feeding. Soc. Neurosci. Abstr. 15:655; 1989.
20. Leibowitz, S. F.; Shor-Posner, G.; Weiss, G. F. Serotonin in medial hypothalamic nuclei control circadian patterns of macronutrient intake. In: Paoletti, R.; Vanhoutte, P. M., eds. Serotonin: From cell to pharmacology and therapeutics. Amsterdam: Kluwer Academic; 1990:203–211.
21. Leibowitz, S. F.; Weiss, G. F.; Shor-Posner, G. Hypothalamic serotonin: Pharmacological, biochemical and behavioral analyses of its feeding-suppressive action. Clin. Neuropharmacol. 11:S51–S71; 1988.
22. Leibowitz, S. F.; Weiss, G. F.; Suh, J. S. Medial hypothalamic nuclei mediate serotonin's inhibitory effect on feeding behavior. Pharmacol. Biochem. Behav. 37:735–742; 1990.
23. Leibowitz, S. F.; Weiss, G. F.; Walsh, U. A.; Viswanath, D. Medial hypothalamic serotonin: Role in circadian patterns of feeding and macronutrient selection. Brain Res. 503:132–140; 1989.
24. Le Magnen, J. The metabolic basis of dual periodicity of feeding in rats. Behav. Brain Sci. 4:561–607; 1981.
25. Martin, K. F; Marsden, C. A. In vivo diurnal variations of 5-HT release in hypothalamic nuclei. In: Redfern, P. H.; Campbell, I. C.; Davies, J. A.; Martin, K. F., eds. Circadian rhythms in the central nervous system. London: Macmillan; 1985:81–92.
26. Massi, M.; Marini, S. Effect of the 5-HT_2 antagonist ritanserin on food intake and on 5-HT induced anorexia in the rat. Pharmacol. Biochem. Behav. 26:333–340; 1987.
27. Paez, X.; Stanley, B. G.; Leibowitz, S. F. Microdialysis analysis of norepinephrine levels in the paraventricular nucleus in association with food intake at dark onset. Brain Res. 606:167–170; 1993.
28. Paxinos, G.; Watson, C. The rat brain in stereotaxic coordinates. 2nd ed. New York: Academic Press; 1986.

29. Pazos, A.; Cortes, R.; Palacios, J. M. Quantitative autoradiographic mapping of serotonin receptors in the rat brain. II. Serotonin-2 receptors. Brain Res. 346:231–249; 1985.
30. Pazos, A.; Palacios, J. M. Quantitative autoradiographic mapping of serotonin receptors in the rat brain. I. Serotonin-1 receptors. Brain Res. 346:205–230; 1985.
31. Samanin, R.; Garattini, S. Serotonin and the pharmacology of eating disorders. Ann. NY Acad. Sci. 575:194–208; 1989.
32. Samanin, R.; Mennini, T.; Bendotti, C.; Barone, D.; Caccia, S.; Garattini, S. Evidence that central 5-HT_2 receptors do not play an important role in the anorectic activity of *d*-fenfluramine in the rat. Neuropharmacology 28:465–469; 1989.
33. Sawchenko, P. E.; Swanson, L. W.; Steinbusch, H. W. M.; Verhofsted, A. A. J. The distribution of cells of origin of serotonergic input to the paraventricular and supraoptic nuclei of the rat. Brain Res. 277:355–360; 1983.
34. Shor-Posner, G.; Grinker, J. A.; Marinescu, C.; Brown O.; Leibowitz, S. F. Hypothalamic serotonin in the control of meal patterns and macronutrient selection. Brain Res. Bull. 17:663–671; 1986.
35. Stallone, D.; Nicolaidis, S. Increased food intake and carbohydrate preference in the rat following treatment with the serotonin antagonist metergoline. Neurosci. Lett. 102:319–324; 1989.
36. Stanley, B. G.; Schwartz, D. H.; Hernandez, L.; Hoebel, B. G.; Leibowitz, S. F. Patterns of extracellular norepinephrine in the paraventricular hypothalamus: Relationship to circadian rhythm and deprivation-induced eating behavior. Life Sci. 45:275–282; 1989.
37. Stanley, B. G.; Schwartz, D. H.; Hernandez, L.; Leibowitz, S. F.; Hoebel, B. G. Patterns of extracellular 5-hydroxyindoleacetic acid (5-HIAA) in the paraventricular hypothalamus (PVN): Relation to circadian rhythm and deprivation-induced eating behavior. Pharmacol. Biochem. Behav. 33:257–260; 1989.
38. Tempel, D. L.; Leibowitz, S. F. Diurnal variations in the feeding responses to norepinephrine, neuropeptide Y and galanin in the PVN. Brain Res. Bull. 25:821–825; 1990.
39. Tempel, D. L.; Shor-Posner, G.; Dwyer, D.; Leibowitz, S. F. Nocturnal patterns of macronutrient intake in freely feeding and food-deprived rats. Am. J. Physiol. 256: R541–R548; 1989.
40. Tian, Y.; Eaton, M. J.; Goudreau, J. L.; Lookingland, K. L.; Moore, K. E. Neurochemical evidence that 5-hydroxytryptaminergic neurons tonically inhibit noradrenergic neurons terminating in the hypothalamus. Brain Res. 607:215–221; 1993.
41. Weiss, G. F.; Papadakos, P.; Knudson, K.; Leibowitz, S. F. Medial hypothalamic serotonin: Effects on deprivation and norepineprhine-induced eating. Pharmacol. Biochem. Behav. 25:1223–1230; 1986.