Pharmacology Biochemistry & Behavior, Vol. 16, pp. 101–111, 1982. Printed in the U.S.A.

Effects of Hypothalamic Knife Cuts on Feeding Induced by Paraventricular Norepinephrine Injections[1]

PAUL F. ARAVICH AND ANTHONY SCLAFANI[2]

Department of Psychology, Brooklyn College of the City University of New York, Brooklyn, NY 11210

AND

SARAH F. LEIBOWITZ

The Rockefeller University, New York, NY 10021

Received 20 August 1981

ARAVICH, P. F., A. SCLAFANI AND S. F. LEIBOWITZ. *Effects of hypothalamic knife cuts on feeding induced by paraventricular norepinephrine injections.* PHARMAC. BIOCHEM. BEHAV. **16**(1) 101–111, 1982.—The relationship between the fiber systems involved in the hypothalamic noradrenergic feeding response and the medial hypothalamic (MH) hyperphagia syndrome was appraised in male rats using knife cuts. Parasagittal knife cuts in the perifornical hypothalamus produced hyperphagia and excessive weight gain but failed to disrupt feeding in response to paraventricular hypothalamic injections of norepinephrine (NE). Coronal knife cuts in the posterior hypothalamus which extended from the midline to the lateral perifornical region also failed to disrupt NE feeding. These findings indicate that the output of the noradrenergic feeding system does not follow the feeding pathway implicated in the MH hyperphagia syndrome. They also suggest that the output of the noradrenergic feeding system is not directed laterally beyond the level of the fornix nor caudally into the lower brainstem over the medial forebrain bundle.

Noradrenergic feeding Hypothalamic hyperphagia Obesity Feeding behavior Knife cuts
Paraventricular hypothalamic nucleus Norepinephrine

IT is well known that microinjection of norepinephrine (NE) into the hypothalamus produces feeding in sated rats [8, 24, 73]. This effect is due to the activation of alpha-adrenergic receptors [9, 25, 38, 39, 67] and is most readily obtained from noradrenergic stimulation of the paraventricular hypothalamic nuclei (PVN) [40,46]. It is also well established that medial hypothalamic (MH) damage produced by lesions [2, 11, 33] or knife cuts [1, 21, 26, 59, 63] results in hyperphagia and obesity in rats and other animals.

Though there are exceptions (see Discussion), several lines of evidence suggest that NE-induced eating and the hyperphagia produced by MH damage may be mediated by a common feeding system. For example, both NE feeding [10,65] and hypothalamic hyperphagia [20,64] are blocked by quinine adulteration of the diet and are associated with decreased performance on appetitively reinforced operant tasks [15,72]. Furthermore, NE microinjections and MH damage, unlike food deprivation, do not produce food hoarding [7,31]. Finally, both NE-feeding and MH hyperphagia are accompanied by enhanced insulin secretion [45,68], and are blocked by subdiaphragmatic vagotomy [55,58]. Because of these functional similarities, several authors have hypothesized that hypothalamic NE injections and MH damage induce feeding by way of a common feeding system [7, 15, 38, 48].

According to this hypothesis, the NE feeding response results from inhibition of the ventromedial hypothalamus (VMH). It is predicted, therefore, that destruction of the VMH should remove the substrate upon which NE acts and, hence, block the NE feeding response. In a test of this prediction [32], it was found that hyperphagia-inducing lesions of the posterior VMH, did indeed block the NE feeding response. However, other hyperphagia-producing lesions located in the more anterior VMH did not block NE eating. Furthermore, lesions which produced the greatest hyperphagia, and therefore most effectively damaged the MH feeding system, "were not necessarily the most effective in blocking the action of NE" ([32] p. 1032).

The relationship between the NE feeding response and the MH syndrome thus remains uncertain. Moreover, the emphasis on the VMH as the critical site involved in hypothalamic hyperphagia is no longer warranted [22] since recent data implicate more anterior hypothalamic areas [62], particularly the PVN [23], as being involved in the hyper-

[1]Portions of this report were presented at the Society for Neuroscience Meeting, St. Louis, MO, 1978.
[2]To whom reprint requests should be sent.

phagia syndrome. It is possible, therefore, that NE injections and MH damage increase food intake by disrupting a common feeding inhibitory system originating in the area of the PVN. Consistent with this view are the findings that microiontophoretic application of NE inhibits the activity of some PVN neurons [49] and that PVN lesions produce hyperphagia [3, 19, 43] as well as block the feeding response induced by intraventricular injections of NE [44].

The present study further assessed the relationship between NE-induced feeding and MH hyperphagia. If NE injections elicit food intake by inhibiting the neural system whose destruction is responsible for MH hyperphagia, then hypothalamic knife cuts of this system should block NE feeding.

EXPERIMENT 1

The results of a number of knife cut studies indicate that MH hyperphagia results from damage to a longitudinally-oriented fiber pathway which interconnects the anteriomedial hypothalamus with the more caudal brainstem [23, 26, 27, 28, 50, 62]. At the level of the PVN this pathway courses laterally into the perifornical region; it then turns caudally in the perifornical area or the most medial portion of the medial forebrain bundle (MFB) to project to the lower brainstem. While it is not clear if this is an ascending or descending fiber system, the position of this pathway overlaps with many of the known efferents of the PVN [5, 14, 57, 69, 71]. In particular, hyperphagia is produced by parasagittal knife cuts in the anterior perifornical hypothalamus which presumably sever a portion of the lateral output of the PVN [1, 21, 63] and by coronal knife cuts in the posterior hypothalamus which are in a position to sever many of the descending efferents of the PVN [28,62]. If the output of the NE feeding system is identical to that of the MH feeding system, then these parasagittal and coronal knife cuts should disrupt the feeding response to PVN injections of NE. The following experiment tested this prediction.

METHOD

Subjects

The subjects in this and all subsequent experiments were adult male Sprague-Dawley rats (Charles River Breeding Labs., CD strain) weighing between 400 and 600 g. The animals were individually housed in wire mesh cages and kept in a temperature controlled colony with a 12:12 hr light-dark cycle. Six rats with parasagittal knife cuts and five rats with sham parasagittal cuts successfully completed all testing and were included in Part 1; eight rats with posterior hypothalamic coronal knife cuts and five rats with sham posterior cuts survived all testing and were included in Part 2. A chow diet (Purina Rodent Laboratory Chow, #5001) and water were freely available throughout all phases of the experiment.

Surgery

For the surgeries in this and subsequent experiments the animals were anesthetized with Equi-Thesin (3.0 cc/kg, IP) and mounted in a Kopf stereotaxic instrument with the incisor bar at 3.0 mm above the ear bars. All rats were implanted with a unilateral drug cannula (23 gauge stainless steel; 1.6 cm long) aimed at the PVN (0.5 mm posterior to bregma; 0.4 mm lateral to the superior sagittal sinus; and 7.8 mm beneath the surface of the skull). Four screws (0-80$\times$1/16) and dental cement secured the implant to the skull. When not in use, the cannulae were kept patent with flush-fitting inner stainless steel pins. In Experiment 1, the animals were also implanted with bilateral 18 gauge cannulae to be used for later knife cut surgery. The cannulae for the parasagittal cuts were positioned 1.0 mm anterior to bregma, 1.0 mm lateral to the sagittal sinus, and 1.8 mm beneath the surface of the skull; for the coronal cuts they were positioned 2.5 mm posterior to bregma, 2.0 mm lateral to the midline, and 1.8 mm beneath the surface of the skull. A postoperative recovery period of at least one week preceded drug testing.

In a second surgery, the animals were given either bilateral knife cuts or sham cuts according to the procedure of Sclafani [59]. In Part 1, parasagittal knife cuts lateral to the PVN were made by inserting the knife shaft through the chronic guide cannulae (1.0 mm anterior to bregma; $\pm$1.0 mm lateral to the midline). At a depth of 0.0 mm (ear bar reference), the cutting wire was extended 3.0 mm posteriorly and the knife was lowered to the base of the brain. Each cut was designed to extend from the anterior hypothalamus (rostral to the PVN) to the area just caudal to the ventromedial hypothamic nucleus. In Part 2, bilateral coronal knife cuts were made in the posterior hypothalamus using a spring-loaded knife carrier [29]. The cuts were designed to extend from the base of the hypothalamus dorsally 3.0 mm and from the lateral perifornical area (L$\pm$2.0 mm) to the midline. Sham lateral or posterior knife cuts were performed in a fashion identical to actual knife cuts except that the cutting wire was not extended into the brain and the knife shaft was not lowered to the floor of the cranium.

Behavioral Test Procedure

Hypothalamic microinjections were made through the chronically implanted drug cannula using a 30 gauge stainless steel injector cannula (1.6 cm long) connected to a microliter syringe (Hamilton Co., Reno, NV) via polyethylene tubing. The NE (l-norepinephrine bitartrate, Sigma Chemical Co., St. Louis, MO) or its vehicle (isotonic saline) was injected in 0.5 μl volumes. The microsyringe was fitted with a repeating dispenser (Hamilton Co.) to insure accurate volumetric delivery.

Feeding responsiveness to NE was appraised in the following fashion. During a one hour preinjection period, the animals were given fresh food and were frequently aroused in order to promote feeding and insure satiety at the time of testing. The subjects were then injected with 0.5 μl of saline and their food intake was measured after 1 hr. In Part 1, immediately following the saline injection period, the animals were injected with NE (35 nmol) and food intake during the next hour was determined. In Part 2, saline and NE tests were conducted on alternate days. A NE feeding score (corrected for spillage) was computed for each animal by subtracting the amount of food consumed after the saline injection from the amount of food consumed after the NE injection. In this and subsequent experiments a reliable NE feeding response was defined as a mean difference score of 1.0 g or more. In addition to these measures, daily 24-hr food intake and body weight were also determined.

The subjects were injected with NE and saline for two to three days to adapt them to the injection procedure. They were then given three NE-saline feeding tests on successive days. Because of the different designs, Part 1 required three days to complete these tests while Part 2 required six days. Animals which displayed a reliable NE feeding response

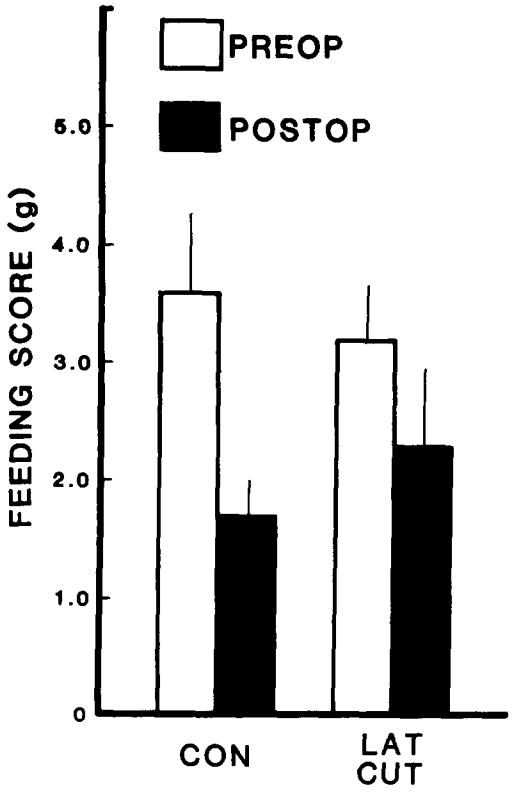

FIG. 1. Mean (±SEM) norepinephrine feeding score (g) before and after knife cut surgery for the control (CON) and lateral cut (LAT CUT) groups.

were retained for further study. In both parts of the experiment, the animals were then divided into two groups equated for feeding responsiveness to NE: the experimental group was given bilateral hypothalamic knife cuts while the control group was given sham knife cuts. Following a two day recovery period, postoperative NE drug testing was conducted in the same manner as preoperative testing. Thus, postoperative testing for Part 1 was conducted on days 3–5 after knife cut surgery while postoperative testing for Part 2 was conducted on days 3–8 following knife cut surgery.

RESULTS

Part 1

The preknife cut 24-hr food intake of the lateral cut group was slightly, but not significantly, below that of the sham cut group (21.8 vs 25.5 g, $p>0.05$). Following knife cut surgery, the cut group increased its daily food intake relative to preoperative levels whereas the control group decreased its intake somewhat (+72.9% for the cut group vs −8.4% for the control group; $t(9)=4.44$, $p<0.01$). The lateral cut animals were thus hyperphagic compared to the control animals during postoperative drug testing (37.8 vs 23.1 g/day; $t(9)=2.98$, $p<0.02$) and by postoperative day 18 had gained significantly more body weight than controls (69.0 vs 13.6 g; $t(9)=4.44$, $p<0.01$).

As illustrated in Fig. 1, prior to knife cut surgery, the NE feeding score of the lateral cut group was slightly below that of the control group, whereas, postoperatively, it was slightly above control levels. Analysis of variance indicated that the group effect and the group by surgery interaction were not significant. The feeding response of both groups, however, significantly declined after surgery, $F(1,9)=7.12$, $p<0.03$. Nevertheless, during postoperative drug testing, all of the knife cut rats and 4 out of the 5 control animals were reliable NE feeders according to the established criterion.

Figure 2A is a photomicrograph illustrating the knife cut and drug cannula placements of a representative animal from the lateral cut group. The cannulae for all animals were in or near the PVN. The knife cuts were positioned 0.1 to 0.3 mm lateral to the lateral edge of the drug cannula. They extended 0.5 to 1.0 mm anterior to the cannula and from 2.0 to 2.5 mm posterior to it, terminating in the mammillary region. There was some variability in the height of the cuts at the level of the PVN: in two animals, the cuts extended dorsally 2.5 to 3.0 mm from the base of the brain, whereas in the remaining animals they extended from the base of the brain 1.5 to 2.1 mm dorsally. In the two animals with the tallest cuts, one animal showed no change in its NE feeding response following surgery while the other animal demonstrated a slight increase.

Part 2

The daily food intakes of the posterior coronal cut and control groups were similar during the preoperative drug tests (30.5 vs 33.4 g). During postoperative drug testing, the daily food intake of the posterior cut animals was significantly less than that of the controls (23.6 vs 30.1 g; $t(11)=2.31$, $p<0.05$). By postoperative day 18 the posterior cut group had gained less weight than did the control group although this difference failed to reach significance (−4.0 vs 10.0 g).

Figure 3 illustrates the results of the NE tests in the posterior cut animals and their controls. The pre- and postoperative NE feeding scores of the posterior cut group were below those of the control group, although analysis of variance revealed that the group effect was not significant. Also, the posterior cut group was not differentially affected by the surgery (group × surgery interaction not significant) although both groups reliably decreased their feeding response postoperatively, $F(1,11)=19.75$, $p<0.002$. All control rats and 5 of the 8 posterior cut rats continued to be reliable NE feeders during the postoperative drug tests. The 3 posterior cut animals which did not display a reliable NE feeding response during the initial postoperative drug tests were given three additional tests. In these subsequent tests, 2 of the 3 rats recovered their NE feeding response. Since the knife cuts in these rats did not differ from those in the other posterior cut subjects, their initial response decrement was probably due to generalized surgical trauma. (Figure 3 does not reflect these additional data.)

Figure 2B is a photomicrograph of a representative animal from the posterior cut group. The knife cuts were in the caudal hypothalamus near the mammillary bodies and from 1.0 to 2.5 mm posterior to the cannula tip. They extended from the fornix to the midline and from the base of the brain dorsally 1.5 to 4.5 mm. The drug cannulae in all subjects were in or near the PVN.

EXPERIMENT 2

The results of Experiment 1 demonstrate that parasagittal knife cuts in the perifornical hypothalamus and coronal knife

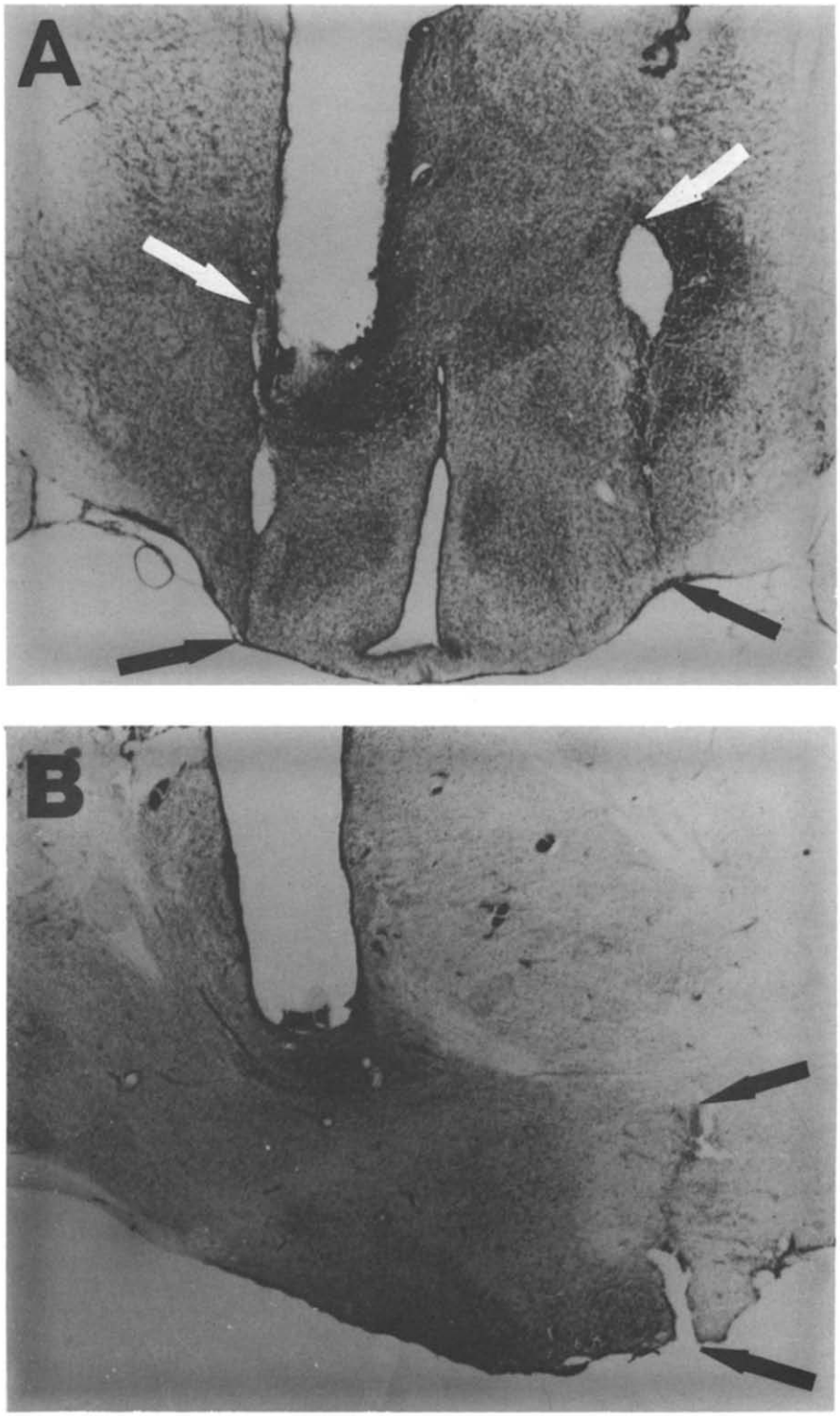

FIG. 2. A: Photomicrograph illustrating the cannula placement and knife cuts of an animal in the lateral cut group. B: Photomicrograph illustrating the cannula placement and knife cuts of an animal in the posterior cut group. (Arrows indicate the knife cuts.)

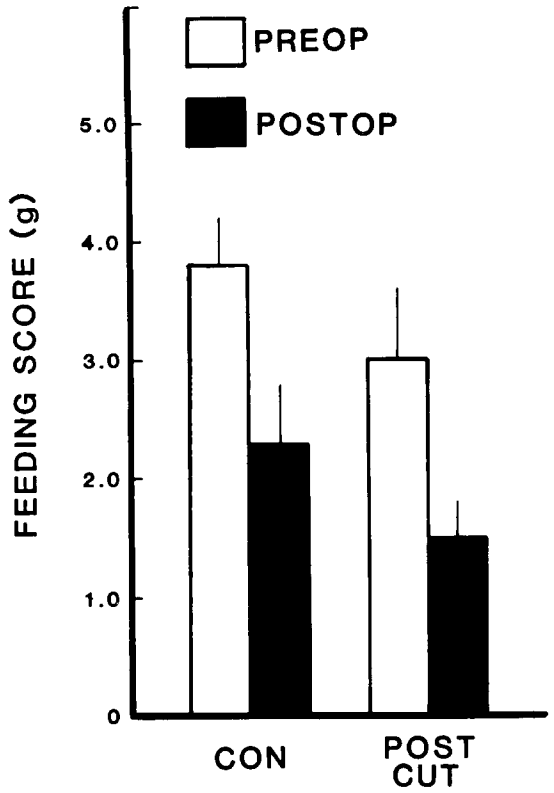

FIG. 3. Mean (±SEM) norepinephrine feeding score (g) before and after knife cut surgery for the control (CON) and posterior coronal cut (POST CUT) groups.

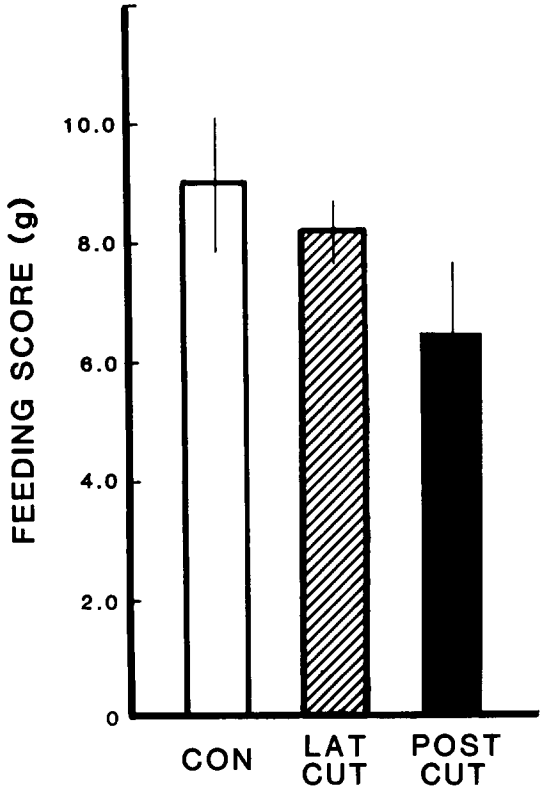

FIG. 4. Mean (±SEM) norepinephrine feeding score for the control (CON), lateral cut (LAT CUT) and posterior cut (POST CUT) groups of Experiment 2.

cuts in the posterior hypothalamus did not block the NE feeding response. These findings suggest that the NE feeding effect does not depend upon the fiber system whose destruction is responsible for MH hyperphagia. (The failure of the posterior cuts to produce hyperphagia is addressed in the Discussion.) These findings also suggest that transections of the lateral and ventroposterior efferents of the PVN are not essential to NE-induced feeding. In Experiment 1, however, the NE feeding response was attenuated following both knife cut surgery and sham-cut surgery, perhaps due to the impact of repeated NE testing and the general malaise produced by the second surgery. In the second experiment, therefore, the effects of hypothalamic knife cuts on the NE feeding response were examined using an experimental paradigm whereby knife cut surgery was performed prior to hypothalamic cannulation and drug testing. A palatable diet, known to enhance the NE feeding response [42], was also employed.

METHOD

Subjects

Five rats with lateral cuts, eight rats with posterior cuts, and six rats with sham cuts successfully completed the experiment and were included in the data analysis.

Surgery

In the first operation the rats were either given bilateral parasagittal cuts lateral to the PVN, coronal cuts in the posterior hypothalamus, or sham cuts. The lateral cuts were designed to be slightly larger than those employed in Experiment 1. The knife shaft was positioned 2.0 mm anterior to bregma and 0.8 mm lateral to the midline. At a depth of 0.0 mm above ear bar reference, a 3.5 mm length of cutting wire was extended in a caudal direction and the knife lowered to the base of the brain. The bilateral posterior cuts were performed using the procedure described in Experiment 1. Sham cuts were made as in the first experiment using the coordinates of either the lateral or the posterior cuts. Following a recovery period of 14 to 19 days, the animals were implanted with a unilateral drug cannula aimed at the PVN.

Behavioral Test Procedure

The testing procedure was similar to Part 1 of Experiment 1 except that a lower dose of NE was employed (20 nmol) and the animals were tested on a more palatable sweet milk-mash diet [300 ml of sweetened condensed milk (Magnolia brand, Borden Foods), 250 g of the powdered form of Purina laboratory chow, and 250 ml of water]. The subjects were maintained on a Purina chow diet for 12 to 13 days following knife cut surgery and were then given the milk-mash diet for the remainder of the experiment. The mash diet was presented in a feeding cup (LC-306, Wahmann Co.) attached to the front of each cage. Food intake measures during the drug tests were corrected for evaporation and spil-

lage. Testing began 23 to 25 days after knife cut surgery and 6 to 11 days after brain cannulation. Daily body weight, but not 24-hr food intake, was recorded throughout the experiment.

RESULTS

By day 21 after knife cut surgery (just prior to the onset of drug testing), the lateral cut animals had gained significantly more body weight than controls (85.8 vs 15.7 g; $t(10)=2.87$, $p<0.05$). The posterior cut animals also gained more weight than did controls during this time period, although this difference failed to reach significance (31.6 vs 15.7 g).

The results of the NE tests are shown in Fig. 4 and analysis of these data indicated that there were no reliable group differences. All animals displayed a reliable eating response to NE. The feeding scores ranged from 6.6 to 9.4 g in the lateral cut group, 1.4 to 11.9 g in the posterior cut group, and 4.5 to 13.0 g in the control group. The somewhat lower performance of the posterior cut group relative to the other groups was largely the result of the low score of one animal that was nearly two standard deviations below the group mean.

Figure 5 illustrates the placements of the knife cuts and cannulae in the experimental groups. The lateral cuts were positioned 0.8 to 1.0 mm lateral to the midline and extended from the posterior portion of the anterior commissure 3.5–4.0 mm caudally to the mammillary region of the hypothalamus. At the level of the PVN, the cuts extended 3.0–3.5 mm dorsally above the base of the brain. In the posterior mammillary region the lateral cuts severed all fibers within 2.0 mm of the base of the brain. The knife cuts in the posterior cut group were in the mammillary region; they extended from the midline 1.0 to 1.5 mm laterally into the perifornical area, and 2.5 to 3.0 mm dorsally from the base of the brain. In all animals, the drug cannula was positioned in the immediate vicinity of the PVN.

EXPERIMENT 3

The results of the preceding experiment replicate those of Experiment 1 and confirm that parasagittal cuts lateral to the PVN, as well as coronal knife cuts in the posterior hypothalamus, do not block NE feeding. The lateral knife cuts in these experiments did not extend to the most dorsal level of the PVN and, therefore, may have spared the more dorsolateral efferents of this nucleus [14]. Even though these dorsolateral efferents are not numerous, it is possible that they may mediate the NE feeding response. Experiment 3 thus investigated the effects of perifornical knife cuts extending well into the thalamus on the feeding response to NE.

METHOD

Subjects

Twelve animals with large lateral cuts and eight sham cut animals survived all testing and were included in the data analysis.

Surgery

The knife shaft was positioned 1.0 mm anterior to bregma, 0.8 to 1.0 mm lateral to the midline, and 2.5 mm above the ear bar reference point. The 3.0 mm cutting wire was then extended and lowered to the base of the brain. Sham cuts were performed as in Experiment 1. In a second operation, which was performed 9 to 13 days after knife cut surgery, the animals were implanted with a PVN drug cannula.

Procedure

The subjects were maintained and tested on a Purina chow diet. Drug testing began 18 days after knife cut surgery and from 6 to 11 days after implantation surgery. A NE dose of 40 nmol was employed using a testing procedure identical to that employed in Experiment 1, Part 1.

RESULTS

In the 18 day period between the knife cuts and the onset of drug testing, both groups lost body weight although the lateral cut group lost significantly more weight than controls (-46.2 vs -19.9 g; $t(18)=1.95$, $p<0.05$, see Discussion). The reason for the weight loss in the controls is unclear, but, at the start of testing, both groups were consuming a normal amount of food (30.8 g for the cut group; 29.8 g for the control group) and their body weights had stabilized.

The results of the NE tests can be seen in Fig. 6A. The mean feeding score of the knife cut animals was slightly but not significantly less than that of controls. Two cut animals and one control animal did not display a reliable NE feeding response. There was no correlation between NE feeding response and 18 day postknife cut body weight change in the experimental animals.

A representation of the knife cuts and cannula placements for the experimental subjects is shown in Fig. 6B. As in the previous experiments, the cannula placements were in or near the PVN. The medial-lateral and rostral-caudal placement of the knife cuts were quite similar to the lateral cuts employed in the preceding experiment. The cuts differed substantially, however, from those of the last experiment in terms of their dorsoventral extent since they extended from the base of the brain well into the thalamus or, in some instances, into the lateral ventricles.

DISCUSSION

The results of this investigation demonstrate that parasagittal knife cuts in the perifornical hypothalamus lateral to the PVN, which produced hyperphagia and excessive weight gain, did not disrupt the feeding response induced by PVN microinjections of NE. Coronal knife cuts in the posterior hypothalamus, which did not produce hyperphagia, but which were in a position to sever the pathway involved in the MH hyperphagia syndrome (see below), also did not block the NE feeding response. The failure of these knife cuts to impair noradrenergic feeding cannot be attributed to regeneration of the relevant fibers since behavioral testing in Experiment 1 began only two days after surgery. It is possible that the lateral cuts did not block the NE feeding response because of diffusion of the NE to receptor sites in the perifornical area. If this were the case, however, the lateral cuts should have significantly reduced the NE feeding response since previous work has established that the feeding stimulated by NE injections into the perifornical region is less than that obtained with PVN injections [40]. Since the posterior knife cuts were 1.0 to 2.5 mm away from the cannula, it is unlikely that the drug acted on receptors located caudal to these cuts. The findings of this experiment, therefore, indicate that the output of the hypothalamic noradrenergic feeding system does not follow the feeding pathway

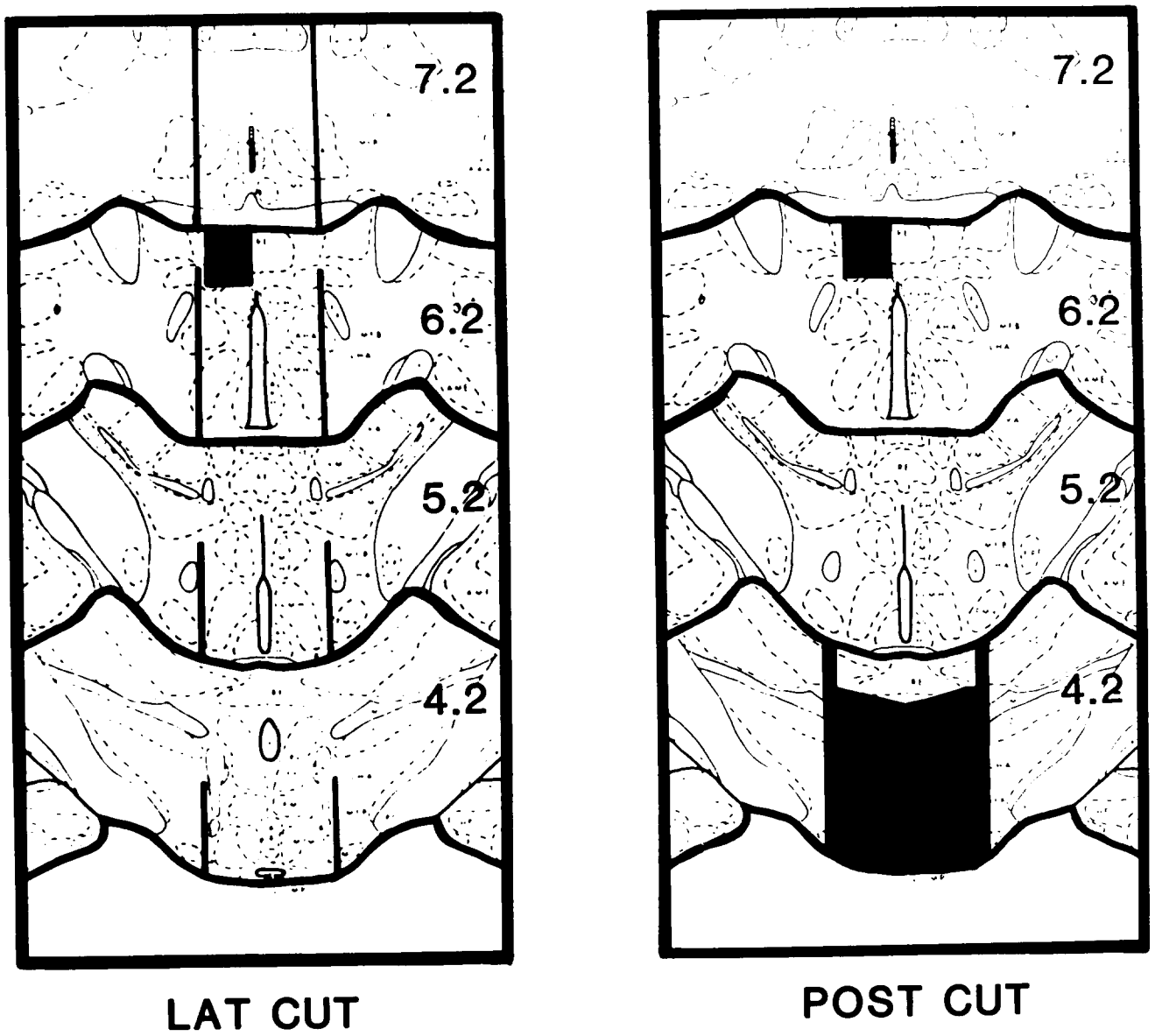

FIG. 5. Schematic representation of the knife cut and cannula placements in Experiment 2. The left panel represents the parasagittal cuts (LAT CUT) while the right panel represents the coronal cuts (POST CUT). (Based on Pellegrino and Cushman atlas [54] with AP level of each section indicated by number in extreme right.)

implicated in the medial hypothalamic hyperphagia syndrome.

As previously discussed, the pathway involved in the hypothalamic hyperphgia syndrome appears to be longitudinally situated in the perifornical region of the hypothalamus. Yet, the posterior coronal knife cuts in Experiments 1 and 2, which should have severed this pathway, did not produce overeating or obesity. Earlier studies have also observed that bilateral posterior coronal knife cuts result in little or no hyperphagia in male rats [28,61]. In female rats, on the other hand, such cuts produce hyperphagia although less than that produced by bilateral parasagittal knife cuts in the perifornical hypothalamus [62]. Furthermore, in both male and female rats, a unilateral coronal knife cut in the posterior hypothalamus combined with a contralateral parasagittal knife cut in the perifornical hypothalamus has been found to produce as much obesity as do bilateral parasagittal knife cuts [23, 61, 62]. Based on these results, Sclafani [61,62] proposed that posterior coronal knife cuts sever the same feeding pathway as do parasagittal knife cuts, but that the coronal cuts also sever additional fibers which, when bilaterally damaged, interfere with the expression of the hyperphagia syndrome. This proposed action of coronal knife cuts is of relevance to the present study because, while the bilateral posterior coronal cuts block the expression of hypothalamic hyperphagia, they do not abolish the feeding response to microinjections of NE.

A similar argument applies to the effects obtained with

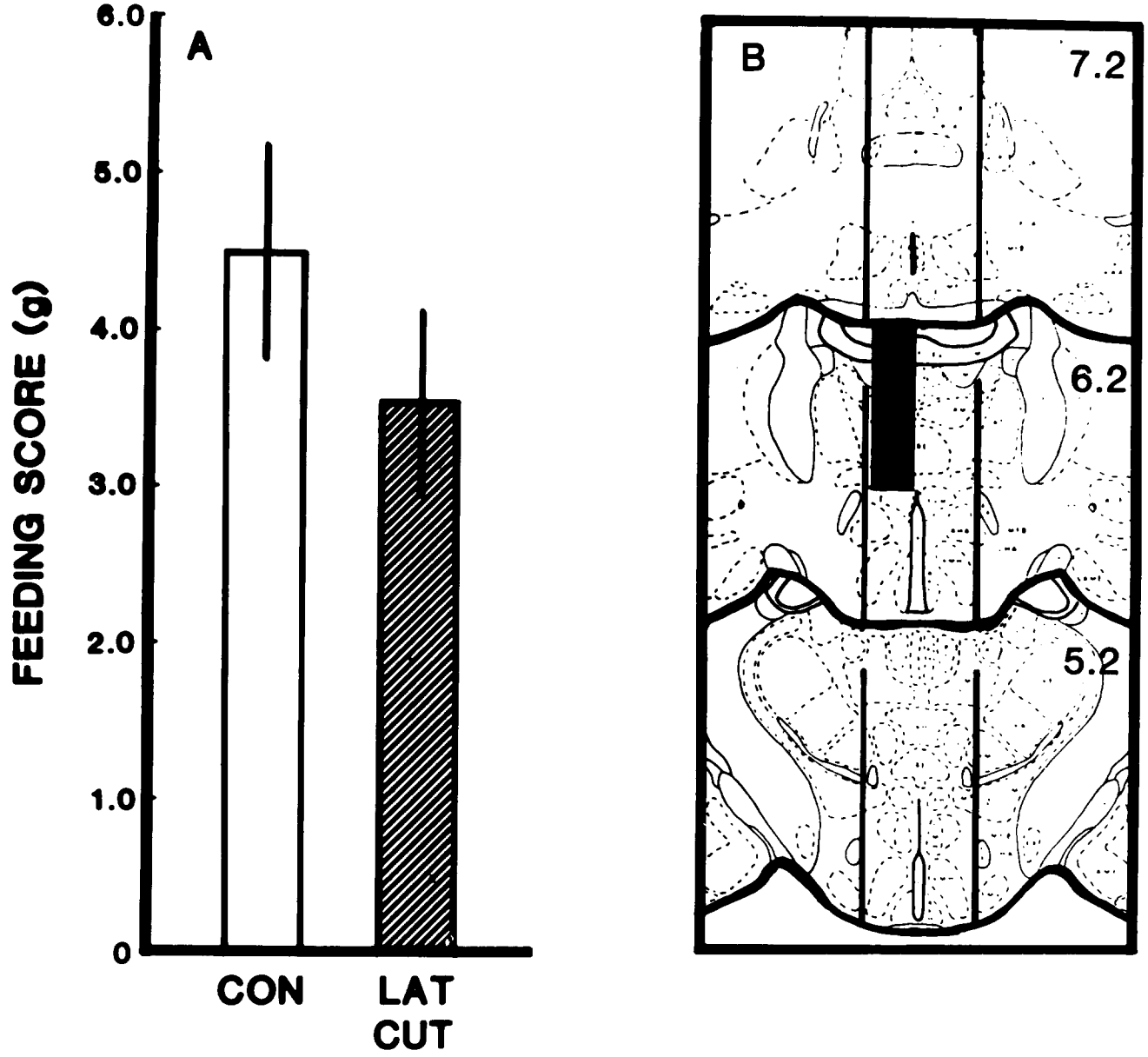

FIG. 6. Experiment 3. A: Mean (±SEM) norepinephrine feeding score for the control (CON) and large lateral cut (LAT CUT) groups. B: Schematic representation of the knife cut and cannula placements; based on Pellegrino and Cushman atlas [54].

the large parasagittal knife cuts used in Experiment 3. Since these cuts severed all the fibers damaged by the smaller, hyperphagia-inducing parasagittal cuts of Experiments 1 and 2, their failure to produce overeating and excessive weight gain can be attributed to the additional damage they produced to the more dorsal diencephalon. Yet, while these larger parasagittal cuts inhibited the hyperphagia syndrome, they did not interfere with NE feeding. Therefore, the results obtained with the nonhyperphagia-inducing knife cuts, as well as those obtained with the hyperphagia-inducing knife cuts, indicate that different systems are involved in the NE feeding response and the hypothalamic hyperphagia syndrome.

Further evidence indicative of a dissociation between the MH hyperphagia syndrome and the noradrenergic feeding response can be cited: (a) high fat diets greatly potentiate the hyperphagia induced by MH damage [12,16], but such diets do not exaggerate the NE feeding response [65]; (b) systemic injections of atropine block NE feeding [58] but not hypothalamic hyperphagia [13,66]; and (c) hypophysectomy inhibits the NE feeding response [41], but not MH hyperphagia [17,35].

Thus, while the noradrenergic feeding response and MH hyperphagia syndrome share some anatomical and behavioral features (see introductory remarks), the present results, and those cited above, suggest that they are mediated by

separate neural mechanisms. Although the nature of these mechanisms remains to be identified, they may involve different aspects of energy regulation. For example, the system involved in the MH hyperphagia syndrome may be related to the long-term regulation of body fat [34, 36, 60], while the noradrenergic feeding system may be involved in the short-term glucostatic control of feeding [41]. Consistent with this later hypothesis are the findings that glucoprivic eating induced by central or systemic injections of 2-deoxy-D-glucose (2DG) can be disrupted by intraventricular alpha-adrenergic blockade [6,51] and that 2DG glucoprivation enhances the release [47] and turnover [56] of hypothalamic NE.

In addition to providing data on the relationship between the MH hyperphagia syndrome and NE elicited feeding, the present study also provides information on the output fibers of the PVN which are *not* involved in the NE feeding response. Among the laterally projecting efferents of the PVN are fibers which form the supraopticohypophysial tract [5, 18, 30, 69, 71]. These PVN efferents pursue a circuitous route as they loop laterally through the perifornical area (or even the internal capsule) to the supraoptic nuclei. Together with fibers from the supraoptic nuclei, they then travel medially along the floor of the hypothalamus into both layers of the median eminence and the neurohypophysis. Since the perifornically positioned parasagittal knife cuts of the present study did not block the NE feeding response, but presumably redundantly damaged many of these fibers, these PVN efferents do not appear to be essential to the feeding effect of NE. However, the knife cuts spared more medially oriented PVN efferents to the median eminence and neurohypophysis [4, 5, 14, 71]. Since hypophysectomy blocks NE feeding [41], the hypothalamo-pituitary axis in general, and these medially directed PVN pituitary projections in particular, would seem to be implicated in the NE feeding effect. On the other hand, the finding [41] that corticosterone replacement therapy restores the noradrenergic feeding response in hypophysectomized rats suggests that the pituitary exerts only a permissive influence on NE eating.

According to Swanson [69], most of the neurophysin-containing autonomic projections of the PVN descend to the lower brainstem in the MFB, ventral and lateral to the fornix. The knife cuts employed in the current study were in a position to significantly impinge upon this projection system but failed to alter NE feeding. This suggests that PVN efferents in the MFB are also not essential to NE feeding. On the other hand, it is unlikely that the knife cuts severed all of the descending output of the PVN. In particular, while the various cuts severed PVN efferents in the medial and perifornical hypothalamus, they produced little or no damage to the more dorsally situated periventricular tract and dorsal longitudinal fasiculus [37] (see also [52]), which may also contain caudally directed PVN efferents to lower brainstem autonomic nuclei (compare [37] with [14]).

In conclusion, the present results indicate that the output of the PVN alpha-adrenergic feeding system does not follow the feeding pathway implicated in the MH hyperphagia syndrome. They further indicate that the output of the noradrenergic feeding system is not directed to the lower brainstem via PVN efferents in the medial or perifornical hypothalamus, nor to the pituitary by PVN efferents in the supraopticohypophysial tract. The results do not exclude the possibility that other caudally directed fiber pathways not transected in the present study (e.g., in the dorsal periventricular stratum) may mediate the NE feeding response. It is also possible that noradrenergic feeding involves a diffusely directed fiber system which cannot be sufficiently interrupted by a knife cut confined to a single plane. Finally, the possibility of a humoral rather than neural output for the noradrenergic feeding system cannot be discounted in light of the fact that the rich capillary network of the PVN is importantly innervated by adrenergic receptors [70] (but see [53]).

ACKNOWLEDGEMENTS

This research report is based on a dissertation submitted by the first author to the Graduate School of the City University of New York in partial fulfillment of the requirements for the Ph.D. degree. Support for this research was provided by NIMH grant MH-21563 and a grant from the Faculty Research Award Program of the City University of New York awarded to A. Sclafani, and by NIMH grant MH-22879 and a grant from the Whitehall Foundation awarded to S. Leibowitz. The assistance of Melvin Reichman is gratefully acknowledged.

REFERENCES

1. Albert, D. J. and L. H. Storlien. Hyperphagia in rats with cuts between the ventromedial and lateral hypothalamus. *Science* **165**: 599–600, 1969.
2. Anand, B. K. and J. R. Brobeck. Hypothalamic control of food intake in rats and cats. *Yale J. Biol. Med.* **24**: 123–140, 1951.
3. Aravich, P. F. and A. Sclafani. Hyperphagia syndromes produced by paraventricular hypothalamic lesions and hypothalamic knife cuts: effects of diet palatability. Paper presented at the Eastern Psychological Association Meeting, New York, NY, 1981.
4. Armstrong, W. E. and G. I. Hatton. The localization of pojection neurons in the rat hypothalamic paraventricular nucleus following vascular and neurohypophysial injections of HRP. *Brain Res. Bull.* **5**: 473–477, 1980.
5. Armstrong, W. E., S. Warach, G. I. Hatton and T. H. McNeill. Subnuclei in the rat hypothalamic paraventricular nucleus: a cytoarchitectural, horseradish peroxidase and immunocytochemical analysis. *Neuroscience* **5**: 1931–1958, 1980.
6. Berthoud, H. R. and G. J. Mogenson. Ingestive behavior after intracerebral and intracerebralventricular infusions of glucose and 2-deoxy-D-glucose. *Am. J. Physiol.* **233**: R127–R133, 1977.
7. Blundell, J. E. and L. J. Herberg. Adrenergic stimulation of the rat diencephalon and its effect on food intake and hoarding activity. *Q. Jl. exp. Psychol.* **22**: 125–132, 1970.
8. Booth, D. A. Localization of adrenergic feeding system in the rat diencephalon. *Science* **158**: 515–517, 1967.
9. Booth, D. A. Mechanism of action of norepinephrine in eliciting an eating response on injection into the rat hypothalamus. *J. Pharmac. exp. Ther.* **160**: 336–348, 1968.
10. Booth, D. A. and D. Quartermain. Taste sensitivity of eating elicited by chemical stimulation of the rat hypothalamus. *Psychon. Sci.* **3**: 525–526, 1965.
11. Brobeck, J. R., J. Tepperman and L. N. A. Long. Experimental hypothalamic hyperphagia in the albino rat. *Yale J. Biol. Med.* **15**: 831–853, 1943.
12. Carlisle, H. J. and E. Stellar. Caloric regulation and food preferences in normal, hyperphagic, and aphagic rats. *J. comp. physiol. Psychol.* **69**: 107–114, 1969.
13. Carpenter, R. G., B. A. Stamoutsos, L. D. Dalton, L. A. Frohman and S. P. Grossman. VMH obesity reduced but not reversed by scopolamine methyl nitrate. *Physiol. Behav.* **23**: 955–959, 1979.

14. Conrad, L. C. A. and D. W. Pfaff. Efferents from the medial basal forebrain and hypothalamus in the rat. II. An autoradiographic study of the anterior hypothalamus. *J. comp. Neurol.* **169:** 221–262, 1976.
15. Coons, E. E. and D. Quartermain. Motivational depression associated with norepinephrine-induced eating from the hypothalamus: resemblance to the ventromedial hypothalamic syndrome. *Physiol Behav.* **5:** 687–692, 1970.
16. Corbit, J. D. and E. Stellar. Palatability, food intake, and obesity in normal and hyperphagic rats. *J. comp. physiol. Psychol.* **58:** 63–67, 1964.
17. Cox, V. C., J. W. Kakolewski and E. S. Valenstein. Effects of ventromedial hypothalamic damage in hypophysectomized rats. *J. comp. physiol. Psychol.* **65:** 145–148, 1968.
18. Defendini, R. and E. A. Zimmerman. The magnocellular nerosecretory system of the mammalian hypothalamus. In: *The Hypothalamus,* edited by S. Reichlin, R. J. Baldessarini and J. B. Martin. New York: Raven Press, 1978, pp. 137–152.
19. Eng, R., R. M. Gold and A. Nunez. Elemental hypothalamic obesity after discrete lesions of the paraventricular nucleus. *Soc. Neurosci. Abstr.* **5:** Abs. #699, 1979.
20. Ferguson, N. B. L. and R. E. Keesey. Effect of a quinine adulterated diet upon the level of body weight maintained by male rats with ventromedial hypothalamic lesions. *J. comp. physiol. Psychol.* **89:** 478–488, 1975.
21. Gold, R. M. Hypothalamic hyperphagia produced by parasagittal knife cuts. *Physiol. Behav.* **5:** 23–25, 1970.
22. Gold, R. M. Hypothalamic obesity: the myth of the ventromedial nucleus. *Science* **182:** 488–490, 1973.
23. Gold, R. M., A. P. Jones, P. E. Sawchenko and G. Kapatos. Paraventricular area: Critical focus on a longitudinal neurocircuitry mediating food intake. *Physiol. Behav.* **18:** 1111–1119, 1977.
24. Grossman, S. P. Eating or drinking elicited by direct adrenergic or cholinergic stimulation of hypothalamus. *Science* **132:** 301–302, 1960.
25. Grossman, S. P. Effects of adrenergic and cholinergic blocking agents on hypothalamic mechanisms. *Am. J. Physiol.* **202:** 1230–1236, 1962.
26. Grossman, S. P. Changes in food and water intake associated with an interruption of the anterior or posterior fiber connections of the hypothalamus. *J. comp. physiol. Psychol.* **74:** 23–31, 1975.
27. Grossman, S. P. and L. Grossman. Food and water intake in rats after transections of fibers en passage in the tegmentum. *Physiol. Behav.* **18:** 647–658, 1977.
28. Grossman, S. P. and J. W. Hennessy. Differential effects of cuts through the posterior hypothalamus on food intake and body weight in male and female rats. *Physiol. Behav.* **17:** 89–102, 1976.
29. Hamilton, L. W., E. Worsham and S. Capobianco. A spring loaded carrier for transection of fornix and other large fiber bundles. *Physiol. Behav.* **10:** 157–159, 1973.
30. Haymaker, W. Hypothalamo-pituitary neural pathways and the circulatory system of the pituitary. In: *The Hypothalamus,* edited by W. Haymaker, E. Anderson and W. J. H. Nauta. Springfield, IL: C. C. Thomas, 1969, pp. 219–250.
31. Herberg, L. J. and J. E. Blundell. Non-interaction of ventromedial and lateral hypothalamic mechanisms in the regulation of feeding and hoarding behavior in the rat. *Q. Jl. exp. Psychol.* **22:** 133–141, 1970.
32. Herberg, L. J. and K. B. J. Franklin. Adrenergic feeding: Its blockade or reversal by posterior VMH lesions; and a new hypothesis. *Physiol. Behav.* **8:** 1029–1034, 1972.
33. Hetherington, A. W. and S. W. Ranson. The relation of various hypothalamic lesions to adiposity in the rat. *J. comp. Neurol.* **76:** 475–499, 1942.
34. Hoebel, B. G. and P. Teitelbaum. Weight regulation in normal and hypothalamic obese rats. *J. comp. physiol. Psychol.* **61:** 189–193, 1966.
35. Ieni, J. R. and R. M. Gold. Two satiety systems revealed by hypothalamic knife cuts in hypophysectomized rats. *Brain Res. Bull.* **2:** 367–374, 1977.
36. Kennedy, G. C. The role of depot fat in the hypothalamic control of food intake in the rat. *Proc. R. Soc.* B **140:** 578–592, 1953.
37. Krieg, W. S. S. The hypothalamus of the albino rat. *J. comp. Neurol.* **55:** 19–33, 1932.
38. Leibowitz, S. F. Reciprocal hunger regulating circuits involving alpha- and beta-adrenergic receptors located, respectively, in the ventromedial and lateral hypothalamus. *Proc. natn. Acad. Sci. U.S.A.* **67:** 1063–1070, 1970.
39. Leibowitz, S. F. Ingestion in the satiated rat: Role of alpha and beta receptors in mediating effects of hypothalamic adrenergic stimulation. *Physiol. Behav.* **14:** 743–754, 1975.
40. Leibowitz, S. F. Paraventricular nuclei: a primary site mediating adrenergic stimulation of feeding and drinking. *Pharmac. Biochem. Behav.* **8:** 163–175, 1978.
41. Leibowitz, S. F. Neurochemical systems of the hypothalamus in control of feeding and drinking behavior and water-electrolytic excretion. In: *Handbook of the Hypothalamus,* vol. 3A, edited by P. J. Morgane and J. Panksepp. New York: Marcel Dekker, 1980, pp. 299–437.
42. Leibowitz, S. F., A. Arcomano and N. J. Hammer. Tranylcypromine: Stimulation of eating through alpha-adrenergic neuronal system in the paraventricular nucleus. *Life Sci.* **23:** 749–758, 1978.
43. Leibowitz, S. F., N. J. Hammer and K. Chang. Hypothalamic paraventricular nucleus lesions produce overeating and obesity in the rat. *Physiol. Behav.* **27:** 1031–1040, 1981.
44. Leibowitz, S. F. Functional and anatomical studies of noradrenergic system of the paraventricular hypothalamus that controls feeding behavior. *Soc. Neurosci. Abstr.* **5:** Abs. #715, 1979.
45. Martin, J. M., W. Konijnendijk and P. R. Bouman. Insulin and growth hormone secretion in rats with ventromedial hypothalamic lesions maintained on restricted food intake. *Diabetes* **23:** 203–208, 1974.
46. Matthews, J. W., D. A. Booth and I. P. Stolerman. Factors influencing feeding elicited by intracranial noradrenaline in rats. *Brain Res.* **141:** 119–128, 1978.
47. McCaleb, M. L. and R. D. Myers. Hypothalamic norepinephrine activity in the rat is altered by peripheral 2-deoxy-D-glucose or insulin. *Fedn Proc.* **38:** 1132, 1979.
48. Morley, J. E. The neuroendocrine control of appetite: the role of the endogenous opiates, cholecystokinin, TRH, gamma-amino-butyric-acid and the diazepam receptor. *Life Sci.* **27:** 355–368, 1980.
49. Moss, R. L., I. Urban and B. A. Cross. Microelectrophoresis of cholinergic and aminergic drugs on paraventricular neurons. *Am. J. Physiol.* **223:** 310–318, 1972.
50. Mufson, E. J., A. Sclafani and P. F. Aravich. Fiber degeneration associated with hyperphagia-inducing knife cuts in the hypothalamus. *Expl. Neurol.* **67:** 633–645, 1980.
51. Muller, E. E., D. Cocchi and P. Mantegazza. Brain adrenergic system in the feeding response induced by 2-deoxy-D-glucose. *Am. J. Physiol.* **223:** 945–950, 1972.
52. Nauta, W. J. H. and W. Haymaker. Hypothalamic nuclei and fiber connections. In: *The Hypothalamus,* edited by W. Haymaker, E. Anderson and W. J. H. Nauta. Springfield, IL: C. C. Thomas, 1969, pp. 136–209.
53. Olschowka, J. A., M. E. Molliver, R. Grzanna, F. L. Rice and J. T. Coyle. Ultrastructural demonstration of noradrenergic synapses in the rat central nervous system by dopamine-β-hydroxylase immunocytochemistry. *J. Histochem. Cytochem.* **29:** 271–280, 1981.
54. Pellegrino, L. J. and A. J. A. Cushman. *A Stereotaxic Atlas of the Rat Brain.* New York: Appleton-Century-Crofts, 1967.
55. Powley, T. L. and C. A. Opsahl. Ventromedial hypothalamic obesity abolished by subdiaphragmatic vagotomy. *Am. J. Physiol.* **226:** 25–33, 1974.
56. Ritter, R. C. and M. Neville. Hypothalamic noradrenaline turnover is increased during glucoprivic feeding. *Fedn Proc.* **35:** 642, 1976.

57. Saper, C. B., A. D. Loewy, L. W. Swanson and W. M. Cowan. Direct hypothalamic-autonomic connections. *Brain Res.* **117:** 305–312, 1976.
58. Sawchenko, P. E., R. M. Gold and S. F. Leibowitz. Evidence for vagal involvement in the eating elicited by adrenergic stimulation of the paraventricular nucleus. *Brain Res.*, 1981, in press.
59. Sclafani, A. Neural pathways involved in the ventromedial hypothalamic lesion syndrome. *J. comp. physiol. Psychol.* **77:** 70–96, 1971.
60. Sclafani, A. Appetite and hunger in experimental obesity syndromes. In: *Hunger: Basic Mechanisms and Clinical Implications*, edited by D. Novin, W. Wywicka and G. Bray. New York: Raven Press, 1976, pp. 281–295.
61. Sclafani, A. Hypothalamic obesity in male rats: comparison of parasagittal, coronal, and combined knife cuts. *Behav. Neural Biol.*, 1981, in press.
62. Sclafani, A. and C. N. Berner. Hyperphagia and obesity produced by parasagittal and coronal hypothalamic knife cuts: further evidence for a longitudinal feeding inhibitory pathway. *J. comp. physiol. Psychol.* **91:** 1000–1118, 1977.
63. Sclafani, A. and S. P. Grossman. Hyperphagia produced by knife cuts between the medial and lateral hypothalamus in the rat. *Physiol. Behav.* **4:** 533–537, 1969.
64. Sclafani, A., D. Springer and L. Kluge. Effects of quinine adulterated diets on the food intake and body weight of obese and non-obese hypothalamic hyperphagic rats. *Physiol. Behav.* **16:** 631–640, 1976.
65. Sclafani, A. and J. Toris. Influence of diet palatability on the noradrenegic feeding response in the rat. *Pharmac. Biochem. Behav.* **15:** 15–19, 1981.
66. Sclafani, A. and S. Xenakis. Atropine fails to block the overconsumption of sugar solutions by hypothalamic hyperphagic rats. *J. comp. physiol. Psychol.*, 1981, in press.
67. Slangen, J. L. and N. E. Miller. Pharmacological tests for the function of hypothalamic norepinephrine in eating behavior. *Physiol. Behav.* **4:** 543–552, 1969.
68. Steffens, A. B. and J. H. Strubbe. Blood composition and feeding behavior in the rat. 5th Int. Conf. Physiol. Food Fluid Int., Jerusalem, 1974.
69. Swanson, L. W. Immunohistochemical evidence for a neurophysin-containing autonomic pathway arising in the paraventricular nucleus of the hypothalamus. *Brain Res. Bull.* **128:** 346–353, 1977.
70. Swanson, L. W., M. A. Connelly and B. K. Hartman. Ultrastructural evidence for the central monoaminergic innervation of blood vessels in the paraventricular nucleus of the hypothalamus. *Brain Res.* **136:** 166–173, 1977.
71. Swanson, L. W. and H. G. J. M. Kuypers. The paraventricular nucleus of the hypothalamus: cytoarchitectonic subdivisions and organization of projections to the pituitary, dorsal vagal complex, and spinal cord as demonstrated by retrograde fluorescence double-labeling methods. *J. comp. Neurol.* **194:** 550–570, 1980.
72. Teitelbaum, P. Random and food directed activity in hyperphagic and normal rats. *J. comp. physiol. Psychol.* **50:** 486–490, 1957.
73. Wagner, J. W. and J. de Groot. Changes in feeding behavior after intracerebral injections in the rat. *Am. J. Physiol.* **204:** 483–487, 1963.