

EFFECTS OF STRIA MEDULLARIS-HABENULAR LESIONS ON FEEDING,
DRINKING, AND GNAWING BEHAVIOR IN MALE RATS

by

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TABLE OF CONTENTS

	Page
INTRODUCTION.	1
EXPERIMENT I.	7
Method	7
Subjects.	7
Apparatus	7
Surgery and histology	7
Procedure	8
Results.	9
Histology	9
Eating.	10
Body Weight	10
Drinking.	10
Gnawing	10
EXPERIMENT II	16
Method	16
Subjects and apparatus.	16
Surgery and histology	16
Procedure	17
Results.	18
Thyroidectomy and histology	18
Eating.	19
Body Weight	20
Drinking.	21
Gnawing	22
DISCUSSION.	31
REFERENCES.	37

The habenular complex borders the third ventricle in the midline epithalamic region anterior and ventral to the pineal gland, and in the rat is divided into medial and lateral nuclei. Most of the afferents to both nuclei travel in the stria medullaris thalami and the efferents project via the habenulopeduncular tracts (fasciculus retroflexus of Meynert) to the interpeduncular nucleus and midbrain tegmentum. Rostral projections of the habenular nuclei in the stria medullaris go to the lateral preoptic area, posterior medial septal nucleus, paraventricular nucleus, and amygdala. Both amygdalo-habenular and habenulo-amygdalar fibers travel through an exchange of fibers between the stria medullaris and stria terminalis. Other habenular projections have been traced to several thalamic nuclei and the posterior hypothalamus (Akagi and Powell, 1968). Wolf and Sutin (1966) followed projections of the lateral hypothalamus through the stria medullaris to their endings in the lateral habenular nuclei.

Experimental manipulations of the habenula have been reported to affect several behaviors, including sexual, ingestive, ambulatory, and avoidant. Little is known, however, about the major habenular afferent and efferent tracts, either physiologically or psychologically.

Two lesion studies of sexual behavior in female hooded rats have yielded apparently contradictory results. Rats with lesions of the medial habenular nuclei mated frequently during diestrus and anestrus (Zouhar and de Groot, 1963). The authors noted that pituitary luteotropin content decreased subsequent to the lesions, and interpreted their data as suggesting involvement of the medial habenular nuclei in reproductive behavior and in a mechanism to increase pituitary production of prolactin. Rodgers and Law (1967) observed a decreased frequency of lordosis during estrus, and increased

avoidance of males during estrus and diestrus. No differential placement of lesions within the habenula was attempted by these authors. Although this may account for the differences in behavior observed, further work is obviously needed to determine which parts of the habenula affect sexual behaviors, and in what ways.

Habenular lesions result in increased open-field ambulation and exploration (as measured by the number of cells entered in a complex maze during two 15 minute periods), but do not affect open-field defecation (Nielson and McIver, 1966). In the same study it was reported that subjects with habenular lesions show greatly impaired active avoidance learning. The avoidance task used was bar pressing in a T-maze, the alley initially chosen by each subject being incorrect, and the opposite side being correct on subsequent trials.

Learning in a step-off passive avoidance situation was not impaired by habenular lesions (Davis, Stevenson, McIver, and Nielson, 1966), but Van Hoesen, MacDougall, and Mitchell (1969) found that such lesions impaired learning of a food-reinforced passive avoidance task in which rats on a 23-hour food deprivation schedule were initially trained to obtain food from a cup, and then during passive avoidance training were shocked for attempting this behavior. The latter authors suggest that the step-off task used by Davis et al. does not correlate highly with other passive avoidance measures. Van Hoesen et al. (1969) also found that subjects with stria medullaris-habenular lesions acquired a conditioned avoidance response in fewer trials than normal or lesioned control subjects. The contradictory results found for active avoidance by Nielson and McIver (1966) were attributed to the subjects having difficulty with the required position reversal.

Altered water consumption subsequent to habenular lesions was reported

by Donovanick, Burrig, Kaplan, and Rosenstreich (1969, 1969). Drinking increased transiently in the habenular group after lesions were made, but this effect was not statistically significant. During a period of adulteration of the water with quinine, however, the habenular group showed a significantly greater decrease in intake than did controls. After this decreased intake due to abstention, both groups consumed more than their pre-quinine levels for an extended period when again given unadulterated water, but the effect was greater in the habenular group than in the control group.

The above observations suggest habenular involvement in several behaviors, but possible effects on feeding and related behaviors have not been studied systematically. Since the extensive literature on neural regulation of food ingestion is largely concerned with contributions of the hypothalamus, this is not surprising. Very recently a trend toward a broader conception of nervous control of eating has arisen, and it is now apparent that extensive circuitry is involved, including several portions of the limbic system.

In an attempt to localize adrenergic fibers of the rat diencephalon involved in feeding, Booth (1967) employed unilateral norepinephrine injection. He found that many of the sites at which stimulation most effectively elicits eating are in or associated with the origins and recurrent collaterals of the stria medullaris, including fibers from the lateral septal area and olfactory tubercle and to the anterior hypothalamus. Related fibers may account for the effective thalamic sites caudal to the stria medullaris. While Booth's argument for an adrenergic circuit controlling eating lacks the strength it would have if an opposite effect could be demonstrated with an adrenergic blocker such as dibenzylamine, it is supported by recent neuro-histochemical data showing relatively high concentrations of monoamine oxidase

in the stria medullaris and habenula (Manocha, Shantha, and Bourne, 1967; Tyrer, Eadie, and Kukums, 1968).

In view of Booth's elicitation of feeding by adrenergic stimulation of the stria medullaris and associated structures, and since the habenula is connected by the stria medullaris with several other limbic and hypothalamic structures important in feeding, one would expect alterations in food-related behaviors subsequent to stria medullaris-habenular lesions.

Previous work in this laboratory suggested that rats with stria medullaris tractotomies and habenular lesions exhibit food intake greater than that of operated controls while on a 23-hour food deprivation schedule, both absolutely and relative to body weight (Van Hoesen, Cooper, MacDougall, and Mitchell, unpublished). Twelve of 25 stria medullaris-habenular subjects died during the experiment, but 6 of 24 operated controls also died, probably of starvation. Some unidentified factors related to hoarding, a major variable in this study, may have interfered with learning to regulate food intake on the deprivation schedule. That more stria medullaris-habenular lesioned subjects died than controls, that they died more quickly, and that the survivors showed greater percentage of body weight decrease in spite of eating more suggested the operation of a metabolic factor or factors in the stria medullaris-habenular groups.

Recently, several authors have reported modifications in various measures of thyroid activity subsequent to experimental manipulations of the habenula. Effects of habenular lesions on thyroid-related functions include inhibition of the goitrogenic effects of thiouracil, inhibition of goitrogenesis due to low iodine intake, inhibition of the inactivation of thyrotropin (TSH) activity by thyroxine administration, and inhibition of TSH release after

thyroidectomy. Serum thyrotropin increases after the lesions, and the thyroid/serum iodine ratio increases much more in lesioned subjects under slight cold stress than it does in normal animals (Mess, 1964). While thyroid grafts in many areas of the rat brain function normally, those placed in the area of the habenula are inhibited (Yasumura, 1962). Finally, hemithyroidectomized rats with habenular lesions show a compensatory thyroidal radioiodide uptake several times greater than that of unlesioned subjects. This effect may last several months (Knigge, 1964).

No clear explanation of the above results has been advanced, but it is obvious that thyroid function is modified by lesions of the habenula. That habenular influences on thyroid function could act through connections with the anterior hypothalamic and preoptic regions was suggested by Knigge (1964) and confirmed by Mess (1967), who found that the anterior hypothalamic area affecting thyrotropin secretion must be intact for habenular lesions to produce their usual effects on thyroidal I^{131} uptake. The possibility exists that some of the changes in dietary behavior observed after production of habenular lesions are at least partially dependent on altered levels of thyroid hormone. In hyperthyroid conditions food intake increases but body weight decreases, as in the hoarding study (Van Hoesen et al., unpublished). Similarities were also noted in the temporal characteristics of thyroid effects produced by habenular lesions and feeding effects noted in the above study. Our data indicated that those subjects surviving initial periods of severe weight loss later increased their food consumption more than did controls, probably to compensate for the weight losses incurred. Since the lesion-induced effects on thyroid function last no longer than three weeks in most circumstances (Bogdanove, 1962; Yamada, 1961), the increased feeding may have occurred after thyroid function returned to normal.

While studying effects of habenular lesions on eating and hoarding in rats, Van Hoesen et al. (unpublished) observed that several of the lesioned subjects gnawed most or all of the approximately 60 food pellets (Purina laboratory rat chow) to bits in a period of only one hour. Thus, quantitative measurement of this gnawing effect was attempted in the experiments to be described.

Experiment I

Method

Subjects. Fourteen male Long-Evans hooded rats weighing from 257 to 378 grams at the beginning of the experiment were used. Food and water were available ad libitum.

Apparatus. All subjects were housed individually in 24 X 13 X 8 in. wire cages with one wall made of tempered masonite. Wooden boxes were placed in each cage to allow collection of food scraps with minimal fouling by urine and feces. A 6 in. long metal tube 3 in. in diameter provided the only entrance to each box, within which was a hopper containing Purina laboratory rat chow pellets. Subjects were required to chew this food through $\frac{1}{2}$ in. wire mesh. Food which was removed from the hopper but not eaten fell through the $\frac{1}{2}$ in. hardware cloth floor to the bottom of the box. The entire food box was easily removed from the cage for food collection.

Surgery and histology. All subjects were anesthetized by intraperitoneal injection of approximately 3.5 ml./kg. Equi-thesin (Jensen-Salsbery Laboratories, Inc., Kansas City, Missouri). Seven subjects were given stria medullaris-habenular lesions while seven others served as their operated controls.

Bilateral stria medullaris-habenular lesions were made by passing direct current through the uninsulated tip of an anodal steel electrode. The cathode was an anally inserted steel rod. Stereotactic coordinates in millimeters from bregma, midline, and top of the dura mater, with the head level, were 2.3 P, 0.6 L, and 4.4 D. Lesion parameters were 1.0 ma. and 12 sec. 45,000 units Bicillin was injected intramuscularly at the conclusion of each operation. Surgical treatment of the control group was similar, but the electrode

was lowered only 4.0 mm., and no current was passed through it.

After the experimental period, subjects were anesthetized and perfused successively with saline and 10% formalin. The brains were removed, embedded in celloidin, and sectioned coronally at 20 micra. Every fourth section from the posterior end of the anterior commissure to the posterior end of the lesion was stained for nuclei with cresyl violet acetate, and every fourth of these was additionally stained for fibers with luxol fast blue MBS. Sections were examined to determine lesion locations and extents, and to detect possible infections.

Procedure. Feeding, chewing, body weight, and drinking measures were taken daily for two to four weeks prior to operations and for 12 days post-operatively. Operations were performed when food intake had been stable for at least eight consecutive days.

Drinking was measured to the nearest milliliter. No allowance for spillage or evaporation was made since these were thought to be minimal. Amount of food ingested was determined by subtracting the sum of food left in the hopper plus that collected from the floor of the food box plus that collected from the main cage floor from the amount placed in the hopper 24 hours earlier. The few pellet portions which had been carried into the main cage were weighed even if fouled. This resulted in a negligible decrease in amount eaten. Scraps on the bottom of the food box and the bottom of the main cage constituted the gnawing measure. Both feeding and gnawing were measured to the nearest 1/10 gram. Subjects were weighed to the nearest gram. Cages were cleaned and supplied with fresh food and water each day after the collection of data.

Results

Since one of the controls escaped, the final numbers of subjects were seven for the stria medullaris-habenular group and six for the control group. All data were analyzed in four-day blocks by unweighted means analysis of variance of a two factor experiment with repeated measures on one factor. Specific comparisons were made by Newman-Keuls tests, using a .95 as the rejection level (Winer, 1962).

Histology. Histological examinations were conducted by two investigators without prior knowledge of behavioral results. All lesions were found to be acceptable, and no signs of infection were detected in the controls. Complete bilateral stria medullaris tractotomy was accomplished in all but one case, in which less than 10% of the tract remained on one side, with total severance on the other. Damage to the lateral and medial habenular nuclei was extensive or complete anteriorly, but absent or restricted to the lateral nuclei posterior to the stereotaxic plane 0.2 mm. anterior to the posterior commissure. Dorsomedial thalamic destruction was variable, being absent at both anterior and posterior ends, and varying from very slight to about 50% in the middle portions. The extent of this thalamic damage was not related to amount of chewing, eating, or drinking. In all cases there was extensive to complete destruction of the paraventricular thalamic nuclei at the level of maximum dorsomedial thalamic nuclear damage. The remaining areas of destruction were associated with the electrode tracks. Five subjects showed slight fornix damage, four had slight encroachment on the anterodorsal hippocampus, two had a little corpus callosum damage, and three had slight unilateral cortical involvement. Figure 1 shows a typical stria medullaris-habenular lesion.

Eating. Food intake by the stria medullaris-habenular group did not differ from that of the control group during any of the four-day blocks. The amount of food eaten, however, varied over time ($F = 33.70$, $df. = 4/44$, $p < .01$), and was significantly less during the four days following operations than at any other time, and less on postoperative days five through eight than on the final four preoperative days.

Body weight. Stria medullaris-habenular lesions did not affect body weight in this experiment. In both groups weights increased until the time of operations, decreased immediately thereafter, and then increased until the end of the experimental period, showing a final mean increase of over 100 grams ($F = 27.84$, $df = 4/44$, $p < .01$). All possible comparisons between blocks were significant except for that between preoperative days five through eight and postoperative days five through eight.

Drinking. Stria medullaris-habenular lesions did not affect the amount of water consumed. The only difference in drinking noted was that consumption was greater in the final postoperative block than in the final preoperative block ($F = 3.68$, $df = 4/44$, $p < .05$).

Gnawing. Figure 2 shows that the stria medullaris-habenular group gnawed more than the control group throughout the experiment ($F = 5.18$, $df = 1/11$, $p < .05$). Differences between the groups were not significant preoperatively, but the stria medullaris-habenular group gnawed significantly more than the control group in each of the three postoperative blocks.

Gnawing decreased in both groups during the first four postoperative days ($F = 4.82$, $df = 4/44$, $p < .01$), and was significantly less at this time than during any other period. This postoperative decrease was significant in

the control group, but the stria medullaris-habenular group showed no significant postoperative changes in gnawing.

Qualitative signs of gnawing were also noted during this experiment. After operations, only one control subject gnawed detectibly on its wooden food box while five of the seven subjects with stria medullaris-habenular lesions gnawed on the boxes and/or the tempered masonite cage walls.

FIGURE CAPTION

Fig. 1. A typical stria medullaris-habenular lesion is shown. There was often habenular damage posterior to that illustrated.

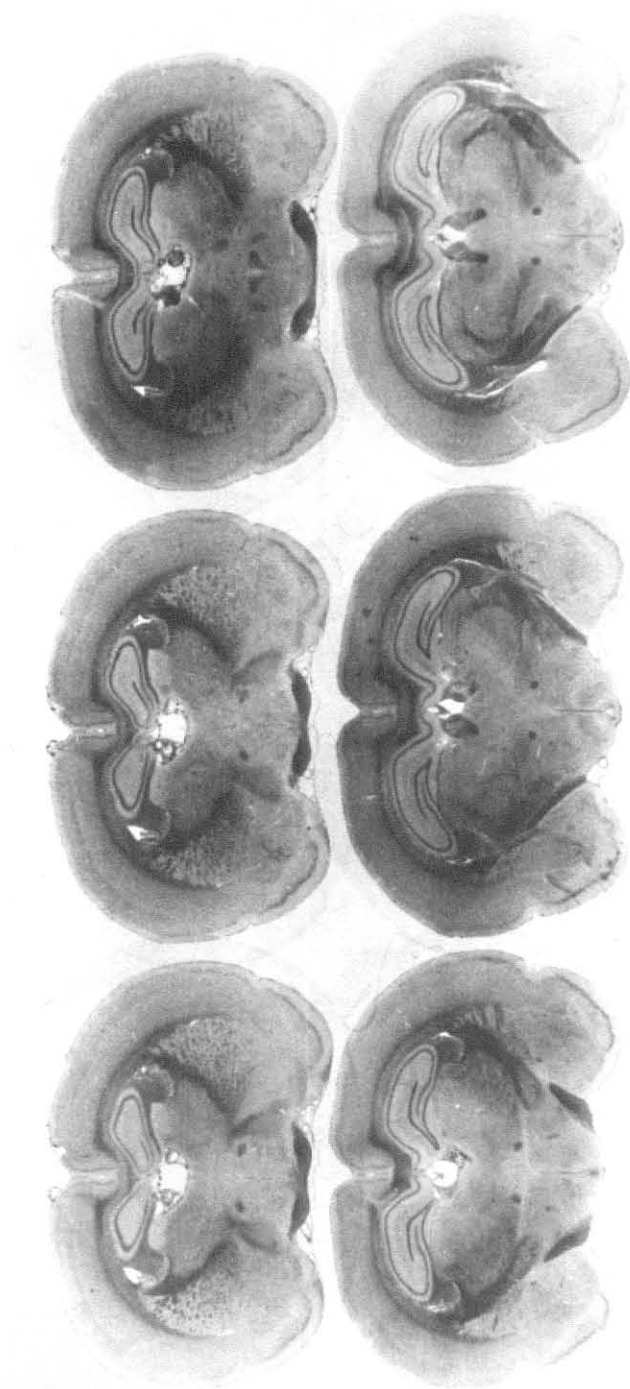
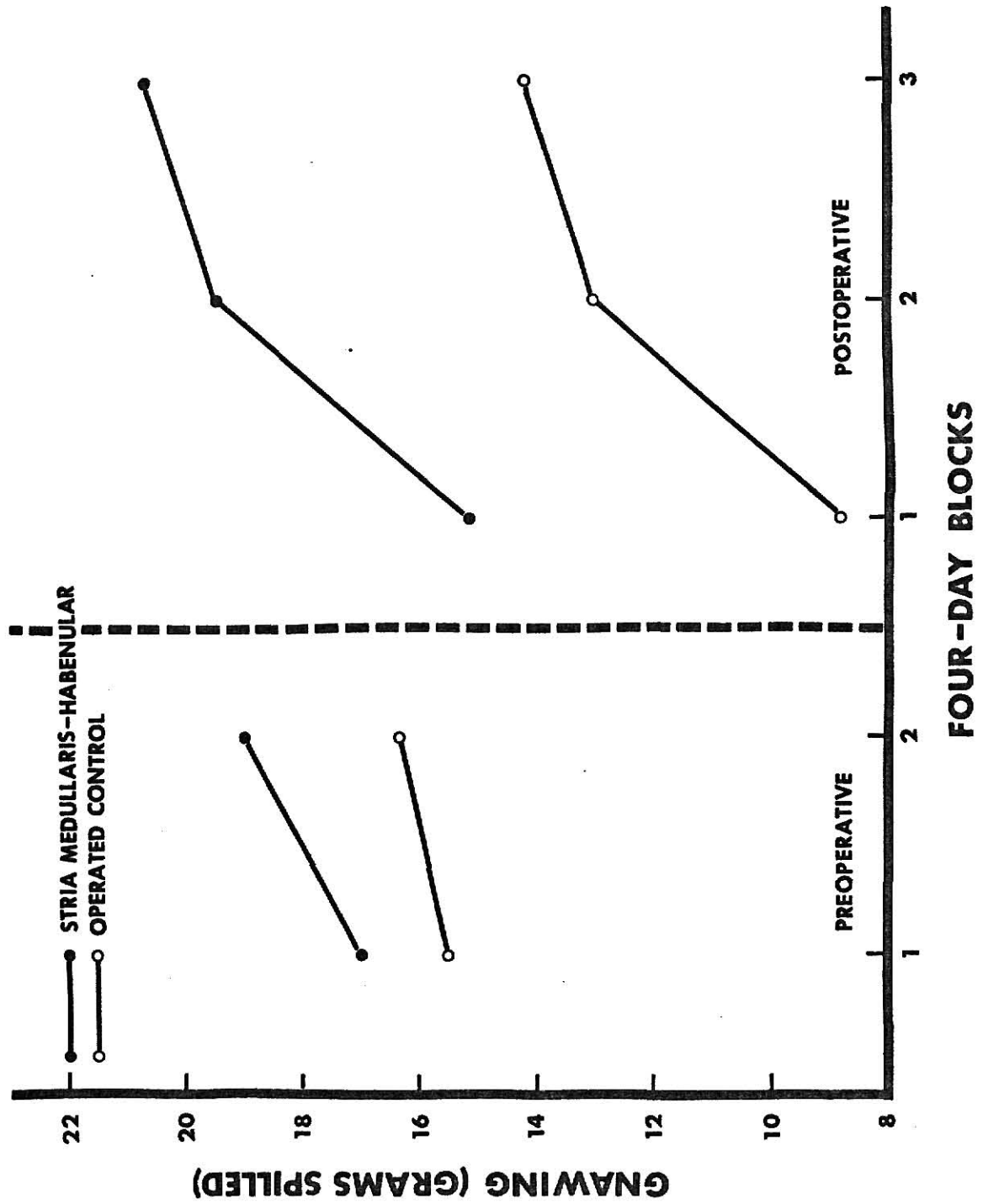


FIGURE CAPTION

Fig. 2. Mean of the total amount gnawed by stria medullaris-habenular and control subjects for each four-day block of experiment 1.



Experiment II

Since the expected effects on food intake were not observed on the ad libitum schedule and it was not possible to demonstrate a postoperative increase in gnawing above preoperative levels by the lesioned group, a 21-hour food deprivation schedule was used in the next experiment. The experiment outlined below was designed to determine whether rats with stria-medullaris-habenular lesions differ from normal rats on the measures used in experiment I when on a 21-hour deprivation schedule, and if so, whether the alterations in the measures are at least partially attributable to lesion produced effects on thyroid activity. To approach these questions, lesioned and control groups were divided into thyroidectomized, sham-thyroidectomized, and cervically intact subgroups. A euthyroid state was maintained in thyroidectomized subjects by replacement therapy to allow determination of the importance of altered thyroid function to lesion-induced changes in behavior.

Method

Subjects and apparatus. Forty-one male Long-Evans hooded rats weighing from 306 to 454 grams at the beginning of the experiment were used. Water was available ad libitum, but food intake was restricted to a three hour period each morning. The apparatus of experiment I was reused.

Surgery and histology. Sixteen subjects were bilaterally thyroidectomized, another 11 were sham-thyroidectomized, while the remaining 13 were left cervically intact. All groups were subjected to brain surgery while under the influence of anesthetic administered prior to neck operations. For the cervically intact group, there was merely a delay of approximately $\frac{1}{2}$

hour between anesthetization and operation. Brain surgery and histology were conducted as outlined in experiment I. Of the thyroidectomized subjects, nine were given stria medullaris-habenular lesions and seven served as their operated controls. The sham-thyroidectomized group consisted of five lesioned subjects and six controls, the cervically intact group of eight lesioned subjects and five controls.

In both thyroidectomy and sham-thyroidectomy, access to the anteroventral tracheal region was gained by blunt separation of the sternohyoideus muscles along the ventral midline. After this had been accomplished, both lobes of the thyroid and the thyroidal isthmus were removed in thyroidectomies. In sham-thyroidectomies, pressure was applied to the anteroventral portion of the trachea, but not to any part of the thyroid. Care was taken to avoid damage to the recurrent laryngeal nerve.

Procedure. Since the daily routine was much like that in experiment I, only differences need be noted. First, the experimental period was extended to include 20 postoperative days. Also, the ambient temperature varied between 19° and 23° C during the course of the experiment with the exception of one 12 hour interval during which the temperature rose to 26° C maximum. Food boxes were left in the cages only during the three hour feeding period. Beginning on the first postoperative day, thyroidectomized subjects were subcutaneously injected with 15 micrograms sodium levothyroxine (Levoid, Nutrition Control Products, 100 micrograms per ml.) per kilogram body weight per day (de Escobar and del Rey, 1967). The remaining animals were injected with .05 ml. physiological saline. Injections were made following measurement of body weight. No replacement therapy for parathyroid deficiency was attempted, but there were no signs of myoneural disturbance.

Results

Unweighted means analysis of variance for a three factor experiment with repeated measures on one factor was used to analyze data in four-day blocks. Specific comparisons were again made by Newman-Keuls tests. After subtraction of animals discarded as noted in the histology section, the number of subjects in each group was as follows: cervically intact group - 12 (seven cervically intact lesioned, five cervically intact unlesioned); sham-thyroidectomized group - 11 (five sham-thyroidectomized lesioned, six sham-thyroidectomized unlesioned); thyroidectomized group - 11 (seven thyroidectomized lesioned, and four thyroidectomized unlesioned). The lesioned group included all 19 subjects with stria medullaris-habenular lesions, while the unlesioned group was composed of their 15 controls.

Thyroidectomy and histology. Three of the thyroidectomized subjects (one lesioned and two controls) suffered recurrent laryngeal nerve damage. The two controls died, but the lesioned subject survived. Also, an undetermined number of subjects sustained irritation to or partial severance of one or both of the sternothyroideus muscles. Two thyroidectomized lesioned subjects died, eight and twelve days after operations, and a thyroidectomized control subject died after 11 days. No plausible explanation for the deaths of the lesioned subjects was found although one of them may have had a brain infection.

Inspection of the brain lesions indicated that they were similar to those described previously, with the following differences:

1. Complete bilateral stria medullaris transection was still the rule, but two subjects received only partial transection on both sides.

2. Habenular damage extended further posteriorly in several cases.
3. In three cases there was little damage to the paraventricular thalamic nuclei.
4. Destruction of the dorsomedial thalamic nuclei was similar to that described previously except that one subject showed nearly 80% obliteration of this area.
5. Slight damage to the cortex, corpus callosum, fornix, and anterodorsal hippocampus occurred in many subjects, and in a few cases was more extensive than in the subjects of experiment I.
6. In several subjects there was slight encroachment on the posterior ends of the anterodorsal, anteroventral, and/or paratenial thalamic nuclei.
7. One subject showed partial destruction of the hippocampal commissure and lateral septal nucleus.

One of the thyroidectomized subjects which died during the experiment received an essentially unilateral lesion and therefore was discarded. The thyroidectomized control which died after 11 days was found to have had a massive brain infection. A cervically intact subject which died on the tenth postoperative day was found to have had almost complete dorsomedial thalamic nuclear damage. The data from animals which died before the end of the experimental period were discarded. The brain of one sham-thyroidectomized lesioned subject was inadvertantly lost. Since this subject's behavior was typical for its group, and in view of the consistent success with which the lesions were made, its data were included in the statistical analyses.

Eating. Stria medullaris-habenular lesions did not produce alterations in food intake. The cervical treatment groups did not differ significantly

on this measure preoperatively, but during the first two postoperative blocks the thyroidectomized group ate less than the cervically intact and sham-thyroidectomized groups, and in the next two postoperative blocks the sham-thyroidectomized group consumed more than the other two groups ($F = 2.88$, $df = 12/168$, $p < .01$). By the final experimental block, differences due to cervical treatments were no longer significant.

In the cervically intact group, eating decreased immediately after operations and remained depressed until the fourth postoperative block. Sham-thyroidectomized subjects also showed a postoperative decrease in eating, but recovered more quickly, eating significantly more during the last eight days than during the first eight postoperative days. They ate more in the final two blocks than preoperatively, but these differences were not significant. Feeding by the thyroidectomized group followed a course similar to that of the cervically intact group, but the thyroidectomized group suffered a greater initial decrease in intake.

Body weight. There were no differences in body weight which could be attributed to stria medullaris-habenular lesions. Differences due to cervical treatments were not detected until the third postoperative block, at which time the sham-thyroidectomized group weighed more than the thyroidectomized group. Thereafter, the sham-thyroidectomized group was significantly heavier than either the thyroidectomized or cervically intact group. As might be expected from this description, the interaction between time and cervical treatments was significant ($F = 5.75$, $df = 12/168$, $p < .01$).

Cervically intact subjects lost weight postoperatively. They were significantly lighter during the second, third, and fourth postoperative blocks than preoperatively, and lighter during the third postoperative block than during the first. The thyroidectomized group showed a weight loss

similar to that in the cervically intact group, and in addition, was significantly lighter in the first postoperative block than in either preoperative block. The sham-thyroidectomized group showed a slight postoperative decrease in body weight, but this loss was not statistically significant. By the last postoperative block, this group weighed slightly more than preoperatively and significantly more than during the first two postoperative blocks.

Drinking. It was not possible to make the desired comparisons of water consumption between groups (Figure 3) because the three-way interaction (lesion X cervical treatment X time) was significant ($F = 1.94$, $df = 12/168$, $p < .05$). Newman-Keuls tests showed that the cervically intact lesioned group drank significantly more during the second postoperative block than on either preoperative block or on the fourth postoperative block, but the cervically intact unlesioned group drank significantly less during the first three postoperative blocks than preoperatively. The sham-thyroidectomized lesioned group drank significantly more on the last four days than preoperatively or during the first two postoperative blocks. In contrast, the amount drunk by the sham-thyroidectomized unlesioned group did not vary significantly over time. On the third, fourth, and fifth postoperative blocks, the thyroidectomized lesioned group drank significantly less than preoperatively or on the last four experimental days. Thyroidectomized unlesioned subjects drank significantly less during postoperative days one through four than at any other time. There were no significant differences between lesioned and unlesioned groups within a cervical treatment when compared on the same four-day blocks, but this was probably due to inflation or the error term by extreme postoperative variability in drinking within the thyroidectomized and sham-thyroidectomized groups.

Since the cervically intact lesioned group drank considerably more postoperatively than the cervically intact unlesioned group, and since these groups showed significant postoperative changes in opposite directions from their preoperative levels, an analysis was made of changes from the eight day preoperative mean for each subject in these groups (Figure 4). This analysis showed that the cervically intact lesioned subjects drank significantly more relative to preoperative means than did their controls in the first two postoperative blocks, but not thereafter.

Gnawing. As in experiment 1, lesion ($F = 13.58$, $df = 1/28$, $p < .01$) and time effects ($F = 5.37$, $df = 6/168$, $p < .01$) were significant. In the deprivation experiment, however, there was also a significant lesion X time interaction ($F = 6.75$, $df = 6/168$, $p < .01$). Lesion groups did not differ significantly in the preoperative blocks, but subjects with stria medullaris-habenular lesions chewed significantly more than their unlesioned controls in each of the postoperative periods (Figure 5). The unlesioned group did not show significant variation in amount of gnawing over time, but the stria medullaris-habenular group gnawed significantly more during the last four postoperative blocks than during the preoperative and first postoperative blocks. Cervical treatment effect, lesion X cervical treatment interaction, cervical treatment X time interaction, and lesion X cervical treatment X time interaction were nonsignificant.

Although the stria medullaris-habenular group as a whole gnawed significantly more postoperatively than preoperatively, and significantly more postoperatively than their controls, the cervically intact subgroups failed to show these relations. Fisher's exact probability test (Siegel, 1956) was used to show that the cervically intact lesioned group had a greater proportion

FIGURE CAPTION

Fig. 3. Amount of water consumed by the six groups of Experiment II expressed as the means of total consumption for subjects of each group during each four-day block. Group abbreviations are as follows: INT-HAB represents the cervically intact habenular group, STHY-HAB the sham-thyroidectomized habenular group, THY-HAB the thyroidectomized habenular group, INT-CONT the cervically intact operated control group, STHY-CONT the sham-thyroidectomized operated control group, and THY-CONT the thyroidectomized operated control group.

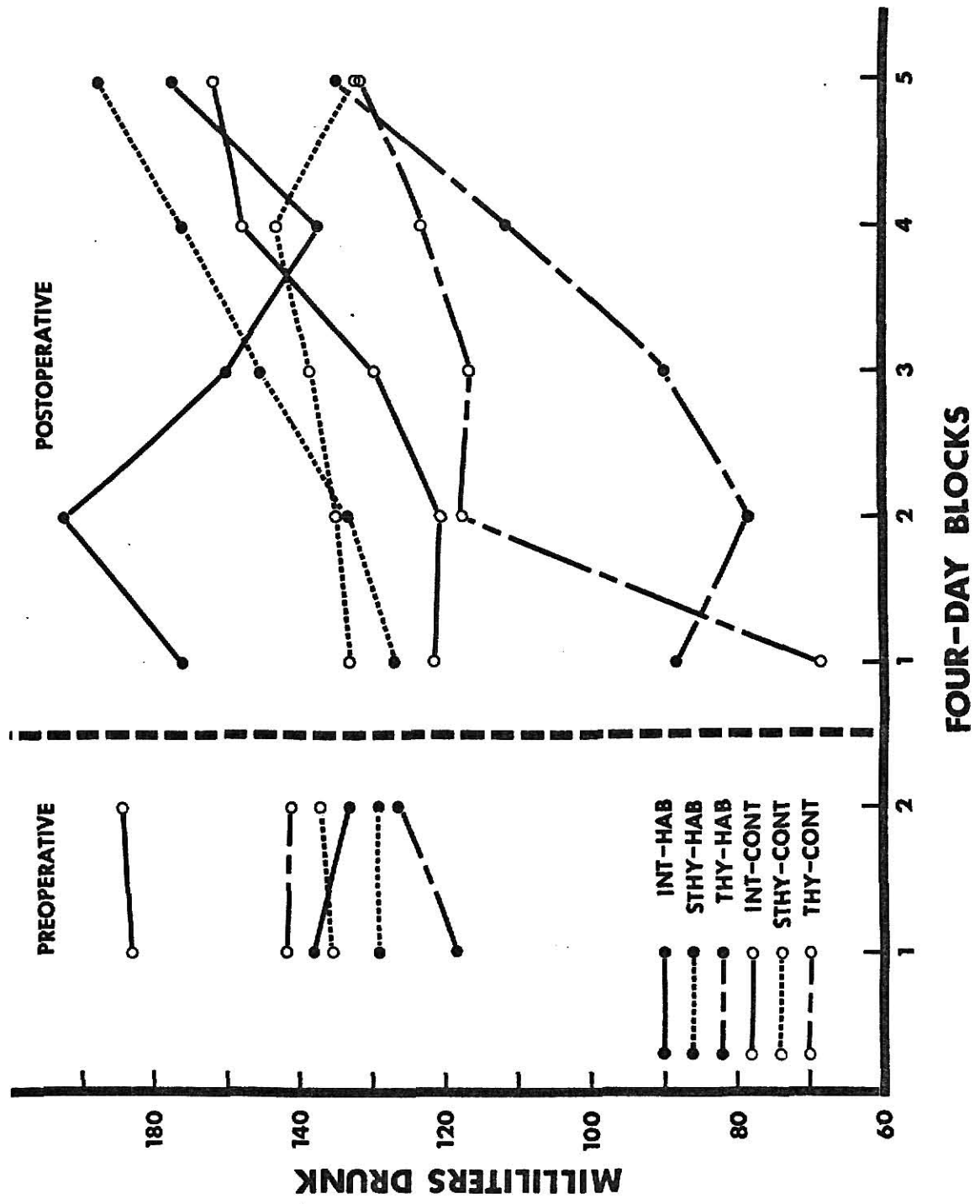


FIGURE CAPTION

Fig. 4. Postoperative changes in water consumption from eight day preoperative means by the cervically intact lesioned and cervically intact operated control groups in Experiment II.

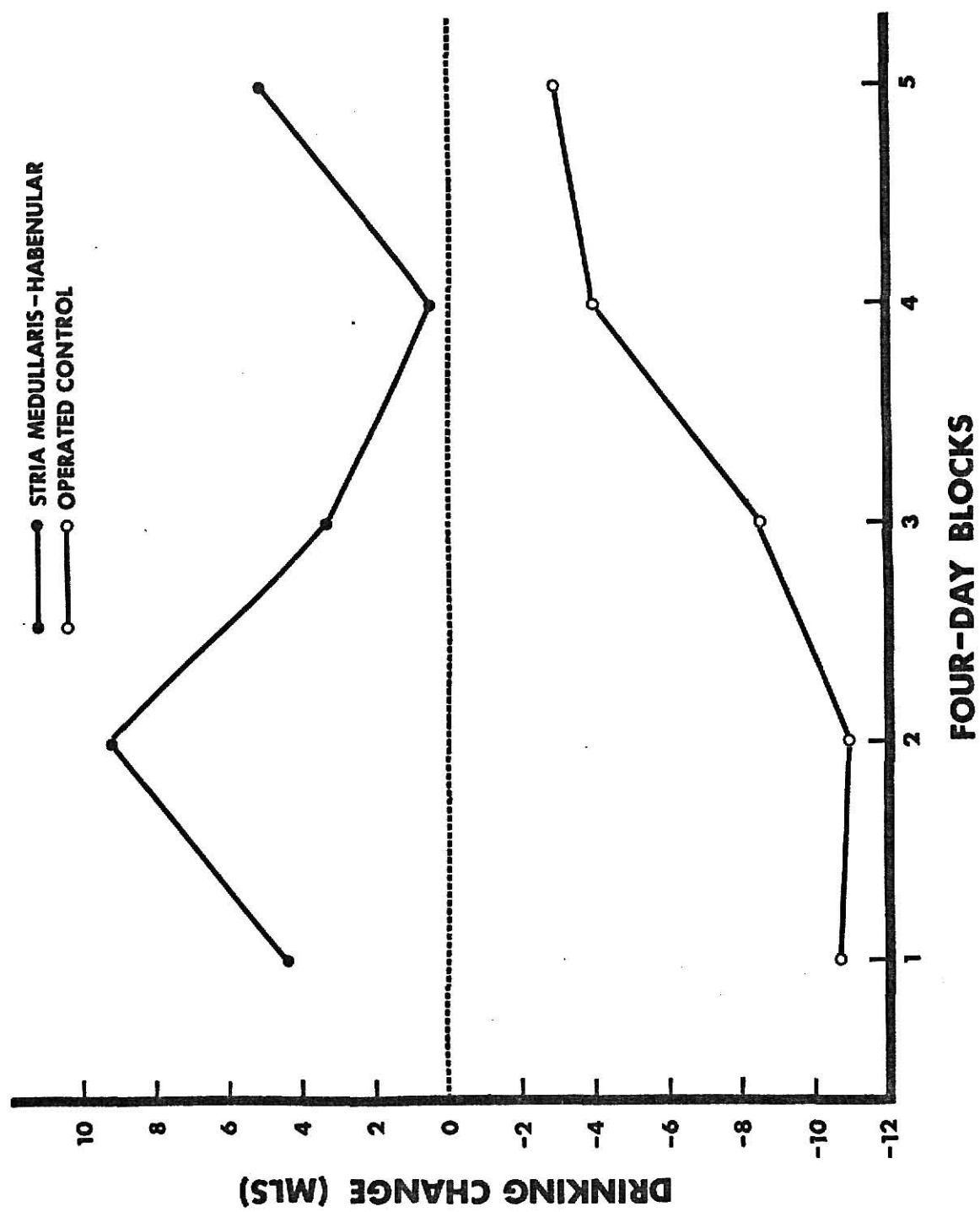
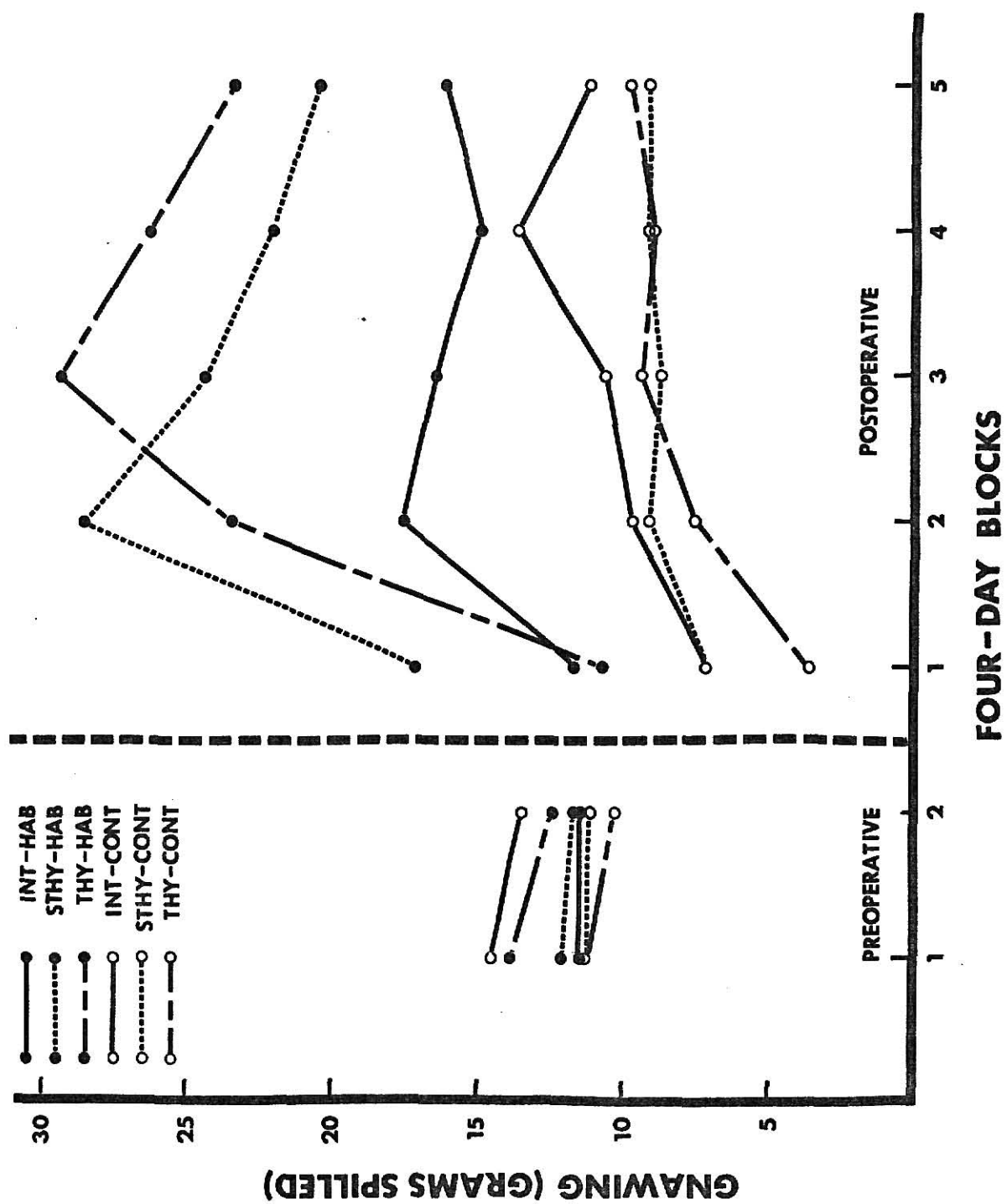


FIGURE CAPTION

Fig. 5. Gnawing expressed as the mean of the total for subjects of each group in each four-day block of Experiment II. Group abbreviations are the same as in Figure 3.



of subjects with more than the mean number of postoperative days on which gnawing was greater than on the highest preoperative day than the cervically intact unlesioned group ($p = .0265$). An unweighted means analysis of variance of postoperative gnawing change from preoperative means for each of the cervically intact subjects revealed no significant overall lesion effect, but this effect did approach significance at the .05 level. On postoperative days five through eight, the cervically intact lesioned group gnawed significantly more relative to preoperative means than did their controls. Similar differences were close to significance on the first, third, and fifth postoperative blocks.

Lesions were reexamined in an attempt to determine why these groups did not show greater differences, but there was no apparent variation among the lesions of the three cervical treatment groups or within the cervically intact group which could account for the differences observed. Finally, the data and laboratory notes for each subject in the cervically intact groups were examined. There were no suggestions of any unusual deviations among the controls. Several subjects in the cervically intact lesioned group, however, were peculiar in one or more respects. Five of them showed increased gnawing postoperatively and the other two showed little change. Of the two subjects which showed little change, one had decreased eating throughout the postoperative period and the other broke off its suture clips shortly after being operated upon, and lost blood due to scratching of the open wound. It was extremely active on postoperative days 10-13, and ate only 3.9 and 4.1 grams of food on the last two of these days. One of the remaining subjects showed increased gnawing during all but the third and fourth postoperative blocks, during which it ate and drank very little and decreased its gnawing greatly, on four days having less than one gram of

spillage. Two other subjects spent a great deal of time on several successive days gnawing on the one cage wall made of tempered masonite. During these times their gnawing measures decreased. It is likely that a combination of some or all of these factors decreased the statistical reliability of a true difference in amount of gnawing.

Discussion

No effects of stria medullaris-habenular lesions were found on body weight or food intake on either an ad libitum or a 21-hour food deprivation schedule. These findings were not expected. Rats with similar lesions in our previous study (Van Hoesen et al., unpublished) lost more weight than controls. Differences in procedure may account for this discrepancy since in the earlier study food was available for only one hour per day as opposed to three hours per day. More importantly, subjects in the unpublished study had no preoperative exposure to the food deprivation schedule and were thus forced to markedly adjust their ingestion patterns during the experimental period.

The earlier study suggested that there was increased food intake following stria medullaris-habenular lesions. Pizzi and Lorens (1967) likewise reported transient increases in intake in rats with lesions involving bilateral stria medullaris transection, as well as damage to the stria terminalis and descending columns of the fornix. As is the case for rats with ventromedial or lateral hypothalamic lesions, adrenergically stimulated rats were hyperresponsive to food flavor and would not work to obtain food (Booth, 1967). In view of this hyperresponsivity and failure to work, it is possible that the not especially palatable food used and/or the laborious feeding responses required by the apparatus in the present study were incompatible with any increase in food intake which might have been produced by the lesions. A further explanation could be that since the habenula has been implicated in the regulation of thyroid function, effects of habenular lesions on food ingestion may occur only at low environmental temperatures or in other situations severely affecting the thyroid.

No lesion-produced effect on drinking was found on the ad libitum schedule, confirming the report of Donovan et al. (1969). These investigators also stated that after periods of self-imposed water deprivation due to quinine aversion, rats with habenular lesions drank more than controls when again given unadulterated water. It is of interest that lesions of the habenula thus affect reactivity to taste, a property shared with septal, lateral hypothalamic, and ventromedial hypothalamic lesions (Mook and Blass, 1968). Lesions of these latter structures, however, produce altered consumption of water on ad libitum schedules.

Differences between habenular and control groups were found on a deprivation schedule in the present study, but the deprivation was of food, not water. In normal rats, food deprivation is accompanied by decreased water intake (e.g., Calvin and Behan, 1954), but cervically intact subjects with stria medullaris-habenular lesions drank more than either group in experiment I for several days at the time during which their feeding was less than it had been preoperatively. This disruption of the normal relation between feeding and drinking suggests that the habenular complex and/or stria medullaris may participate in the correlation of these behaviors. This possibility is supported by the observation that the habenula communicates via the stria medullaris with many of the structures known to affect feeding and drinking behaviors.

Thyroidectomy and, to a lesser extent, sham-thyroidectomy produced undesired modifications of body weight, eating, and drinking, affecting each of these regardless of effects produced by damage to the habenula. These were probably due to surgical trauma to the cervical region, particularly irritation of the musculature of this area, the trachea, and possibly the recurrent laryngeal nerve. Nevertheless, interpretation of statistically

significant effects of stria medullaris-habenular lesions was modified due to interaction with effects of cervical treatments only for drinking, in which case the data for the cervically intact groups were relied upon.

The present study gives the first report of increased gnawing produced by experimental brain lesions. Changes in the gnawing measure used could reflect altered eating patterns, but since many of the rats with stria medullaris-habenular lesions showed increased gnawing of inedible objects, such changes are interpreted as changes in gnawing rather than eating. Support for this notion is provided by the observation that while there were no lesion-produced effects on food ingestion, lesioned rats gnawed more food than their controls. Disturbances in eating ability may have required lesioned subjects to chew more food than controls to ingest comparable amounts, but there were no observations suggesting such a handicap in experiment I or in the cervically intact groups of experiment II. A handicap of this type could have occurred in the groups subjected to cervical operations. Sham-thyroidectomized lesioned subjects, however, showed increases in both eating and gnawing compared to preoperative levels in the latter portions of the experimental period. Increases in the gnawing measure also might represent greater "sloppiness" due to lack of coordination of the part of lesioned animals, but they were adept in handling wood and cardboard when gnawing. Since the increases in gnawing are not thought to represent a food-related behavior, a measure of gnawing of inedible objects would be more satisfactory in future studies.

The gnawing observed was directed to appropriate objects, and no gnashing or chattering of the teeth was seen. It is probable that food deprivation served to direct attention to food pellets as gnawing objects or that the rewarding effects of gnawing during eating were enhanced. Either of

these hypotheses could account for the greater effects observed on the deprivation schedule.

The results also show that the increased gnawing is not dependent on increased thyroxine levels produced as a result of the lesions. If thyroxine levels had any effect on gnawing in the lesioned groups, it would seem to have been inversely related to amount of gnawing. The thyroidectomized animals maintained in a euthyroid state reached a higher level of gnawing than any other group.

A recent series of studies at the University of Utrecht deals with gnawing produced by stereotactic implantation of DOPA or apomorphine in rat brains. Sites at which effective induction of gnawing occurred were the dorsal portion of the caudate nucleus and the globus pallidus. Ineffective sites included the ventral portion of the caudate nucleus, the lateral septal nucleus, subthalamic structures (unspecified), and the substantia nigra. At the effective sites "intense compulsive gnawing" began after $\frac{1}{2}$ to two hours and continued for two to three hours (Ernst and Smelik, 1966). After concluding that the site of action of the drugs is in the neostriatum, Smelik and Ernst (1966) studied the role of dopaminergic fibers from the substantia nigra to the neostriatum. Gnawing was produced by implantation of crystalline physostigmine, which causes local accumulation of acetylcholine by blocking cholinesterase, in the substantia nigra. Sites outside the substantia nigra, including the caudate nucleus and globus pallidus, were ineffective. Increased gnawing produced in this fashion was blocked in all cases by intraperitoneal injection of atropine (20 mg./kg.) 15 minutes prior to implantation. The authors concluded that the dopaminergic nigro-neostriatal fibers affect gnawing and are cholinergically activated. Roberts and Carey (1965) reported that electrical stimulation

of the middle and posterior portions of the lateral hypothalamus produced gnawing behavior in rats. They further demonstrated that gnawable objects were effective rewards for learning of position and black-white discrimination in a Y-maze by subjects stimulated in these areas, but not by subjects stimulated in other areas of the brain. Since onset of stimulation while a subject was eating powdered food was followed by cessation of eating in favor of gnawing wood, it appears unlikely that the subjects were gnawing due to hunger. Finally, gnawing occurred only in the presence of suitable objects, not being elicited reflexly.

This study was followed by the report that ventromedial hypothalamic lesions bring about a decrease in amount of wood gnawing by rats (Cox, Kakolewski, and Valenstein, 1967). Combining this observation and the data of Roberts and Carey (1965), Cox et al. argued that any direct effect of the ventromedial hypothalamic nuclei on lateral hypothalamic substrates of gnawing would probably be facilitatory.

In view of the direct connections between the lateral hypothalamus and habenula via the stria medullaris, it is not surprising that the latter structure(s) subserves gnawing, apparently having an inhibitory role in that behavior. Connections between the habenula, the lateral hypothalamic area and the ventromedial hypothalamic nuclei (Mitchell, 1963), between the lateral and ventromedial hypothalamus, and between the ventromedial hypothalamus and the globus pallidus (House and Pansky, 1960) as well as these nuclei, probably constitute part of a more extensive subcortical neural system affecting gnawing. Since there are habenular projections to tectal areas in both the superior and inferior colliculi, there is also a possibility at this level of interaction with nigral fibers, and thereby with the nigro-neostriatal system described by Smelik and Ernst (1966). No data exist on

the types of fibers in the stria medullaris and habenula which affect gnawing, but since reported concentrations of monoamine oxidase in the habenula are quite high (Tyrer, Eadie, and Kukums, 1968), it seems likely that they will prove to be aminergic.

Like other limbic and hypothalamic structures, the habenula has been implicated in several behavioral and physiological processes. Its relation to the anterior hypothalamic thyrotropic area makes it important in regulating thyroid function (Mess, 1967), and it apparently affects prolactin inhibiting factor as well, since lesions of the medial habenular nuclei result in decreased pituitary prolactin content (Zouhar and deGroot, 1963). Behaviors affected by experimental manipulations of the habenula are also affected by structures connected to the habenula by the stria medullaris, and it has been suggested that dorsal diencephalic septal projections to the habenula through the stria medullaris may be responsible for similarities between rats with septal and habenular lesions in quinine aversion (Donovick et al., 1969) and in performance of active and passive avoidance tasks (Van Hoesen et al., 1969). Likewise, habenular effects on gnawing are undoubtedly related to those of hypothalamic areas by the direct connections between these structures. From the scanty information available in this case, however, the lateral and ventromedial hypothalamic areas appear to have an influence opposed to that of the habenula, suggesting that the habenula may have an inhibitory influence on gnawing-related output from these centers.

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EFFECTS OF STRIA MEDULLARIS-HABENULAR LESIONS ON FEEDING,
DRINKING, AND GNAWING BEHAVIOR IN MALE RATS

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Effects of bilateral electrolytic lesions destroying the habenula and severing the stria medullaris were studied. Changes in gnawing, drinking, eating, and body weight were investigated. The lesions resulted in increased gnawing, and this increase was independent of alterations in thyroid function produced by the lesions. A possible neural circuit concerned with gnawing is discussed. No effect of the lesions on drinking was demonstrated on an ad libitum feeding schedule, but lesioned subjects drank more than controls on a 21-hour food deprivation schedule. Amount of food ingested and body weight were not modified by the lesions.