

EFFECTS OF SEPTAL OR VENTROMEDIAL, HYPOTHALAMIC
LESIONS ON THE DIURNAL FEEDING PATTERNS OF FEMALE RATS

by

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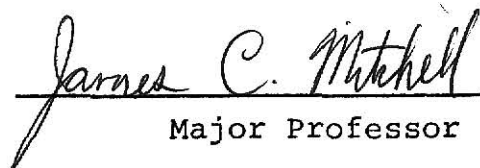
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TABLE OF CONTENTS

	Page
List of Figures and Tables	iii
Introduction	1
Method	6
Results	10
Daily Measures	10
Meal Patterns	15
Histological Analyses	27
Discussion	28
Water Intake	28
Food Intake	28
Correlations	36
References	40

LIST OF FIGURES AND TABLES

Figures	Page
1. Mean body weight and daily food and water intake for one preoperative and four post-operative weeks.	13
2. Mean measures of: percentage total daily intake, meal size, meal frequency and interval for the daytime and the nighttime for each lesion group.	18
3. Percentage frequency histogram for meal sizes. Percentages are plotted for day and nighttime data for each lesion group.	21
4. Percentage frequency histogram for post-meal intervals. Percentages are plotted for day and nighttime data for each lesion group.	23
Table	
1. Correlations of meal sizes and post-meal interval for day and night.	26

INTRODUCTION

Two important factors in the regulation of food intake of rats are: (1) caloric content and (2) orosensory cues of the food. Many investigations have shown that rats consume their daily food intake in discrete meals separated by predictable intervals (Richter, 1927; Brobeck, 1955; Teitelbaum and Campbell, 1958; Thomas and Mayer, 1968; LeMagnen, 1971). Caloric value of the food consumed determines the interval between meals (Adolph, 1947; Thomas and Mayer, 1968; Snowden, 1969, 1970; Snowden and Epstein, 1970; LeMagnen, 1971); and the palatability of the diet appears to be the major factor in determining the size of a given meal (Teitelbaum and Campbell, 1958; LeMagnen, 1971). This differentiation implies that postingestive or metabolic feedback is critical for determining when to initiate a meal and that orogastric cues such as taste or texture are involved in regulating meal size. When the diet is changed in caloric value by adding cellulose or water, rats maintain a stable caloric intake by eating more often; if orosensory cues are altered by adding quinine or saccharin, rats eat different sized meals but change the meal frequency in order to ingest an equal number of calories.

LeMagnen (1971) has shown that the size of a meal is positively correlated with the length of the post-meal interval, implying that a meal is initiated when the calories from the previous meal have been expended. In addition to the partitioning of the daily feeding into discrete meals and intervals, the feeding patterns differ during the day/night cycle. During the

night rats consume more than during the day by ingesting meals more frequently; the positive correlation between meal size and interval remains (Siegel, 1961; Balagura and Coscina, 1968; LeMagnen, 1971).

The relationship between meal size and interval has been shown to be disrupted by brain damage. The most widely studied of these disturbances is the hyperphagic syndrome produced by damage to the ventromedial hypothalamus. Early work by Brobeck, Tepperman and Long (1943) showed that rats with ventromedial hypothalamic (VMH) lesions greatly increased their daily intake and became obese. Brobeck et al (1943) also noted that most of the weight gain occurred during the first two postoperative months. During this initial "dynamic" phase, VMH rats ate two to three times their preoperative food intake level and showed corresponding rapid weight gain. With increases in weight however, the amount consumed began to decrease until a "static" phase was reached. Body weight stabilized, food intake nearly returned to the preoperative level, and further weight gains were comparable to controls. Although the static rat seems to regulate normally even at an exceedingly high body weight, more subtle deficiencies have been shown.

Teitelbaum and Campbell (1958) found that dynamic female rats increased their daily intake by eating larger meals and failing to compensate with less frequent meals. Statics, on the other hand, ingested slightly larger meals but also appropriately decreased the frequency of their meals. Thomas and Mayer (1968) confirmed these results in dynamics; in agreement with LeMagnen (1971) they also found meal size was correlated with post-meal

interval for normals ($r=.74$) as well as hyperphagic dynamics ($r=.78$). Although there was still a positive relationship between meal size and interval, VMH rats consumed larger meals and became obese.

The day/night feeding pattern is also disrupted in the female VMH hyperphagic rat. Balagura and Davenport (1970) report that the increase in intake of VMH rats was due to an overall increase in meal size and a 75% increase in meal frequency during the day over the preoperative level. This increase in the daytime meal frequency produced equal day and nighttime consumption.

In terms of controlling factors, there is evidence that the increase in meal size is due to an overreaction to oral cues (Teitelbaum, 1955). When their diet is adulterated with quinine or sucrose, VMH rats show an exaggeration of the tendency in the normal rat to drink less of the quinine and more of the sucrose adulterated food.

Therefore, the increase in food intake in the VMH rat is accounted for by two important deviations from the normal meal pattern. (1) They consume larger meals but do not compensate by increasing the time interval between meals, and (2) they eat more often during the daytime than normals.

Lesions of the septum also cause food and body weight changes. Although a number of studies have indicated that male rats with septal lesions maintain lower than normal body weights (Beatty and Schwartzbaum, 1967; Flarhety and Hamilton, 1971), little effort has been made to replicate this with female

rats. The only available study on sex differences indicated that female rats with large septal lesions maintained higher, while male rats maintained lower than normal body weights (Kondo and Lorens, 1970; Lorens and Kondo, 1971).

Rats with lesions of the septum have also shown changes in food intake. Beatty and Schwartzbaum (1967, 1968) found septal rats, like VMH rats to be hypersensitive to orosensory factors. When given water adulterated with quinine or sucrose, septal rats drank less quinine and more sucrose than normals. Septal rats may also have a postingestive feedback deficiency. They appeared to be insensitive to the nutrient effects of sucrose (Hamilton, 1971) as well as caloric differences in sucrose concentrations (Flarhety and Hamilton, 1971). In a more elaborate investigation, Hamilton, Capobianco and Worsham (1974) examined the septal response to the postingestive feedback of intragastric loads. The decrease in intake following glucose, sucrose or corn oil stomach loads was less for the septal than for sham rats. Thus, Hamilton concluded that lesions of the septum cause a reduced sensitivity to postingestive feedback, in addition to an increased sensitivity to orosensory cues.

Based on these conclusions, two predictions about the temporal meal patterns of septal rats can be made. (1) If the septal rat is hypersensitive to orosensory cues, then the meal sizes should be larger than those of control rats. (2) If the septal rat is hyposensitive to postingestive cues, then the correlation between meal size and post-meal interval should be lower than controls.

The present study will test these hypotheses by examining the meal patterns of rats with septal, VMH or sham lesions. Data from the VMH group will serve 3 purposes: (1) a replication of previous results concerning the meal patterns of VMH rats will validate the sensitivity of the procedures used, (2) previous results will be extended to examine the meal patterns as well as the diurnal cyclicity of static VMH rats, and (3) the relative changes in meal patterns following VMH or septal lesions will be compared.

METHOD

Subjects--Subjects were 20 Long-Evans hooded female rats. The weight range at the beginning of the experiment was 247-350 grams.

Feeding Apparatus and Housing--The diet¹ consisted of a variation of the enriched eggnog diet described by Epstein and Teitelbaum (1962) to be nutritionally adequate for long term maintenance. The diet for each animal was stored in an inverted 250 ml. bottle secured inside a small ice bucket with a container of ice. This food reservoir was situated above each animal's cage. A tube made of part plastic and part stainless steel led from the food bottle to a glass spout; the spout was inserted into the animal's cage and secured with a vinyl grommet. Licking was monitored by a drinkometer and recorded on an Esterline-Angus (Model No. AW) event recorder. A capacitor circuit caused the recorder pen to deflect upon drinkometer activation. Event records indicated the distribution of time spent eating over a 24 hour period. The measured amount of eating time accurately reflected food consumption ($r=.89$).

Water was available ad lib in an inverted graduated cylinder. Room lights were automatically turned on at 8:00 AM and off at 8:00 PM. White noise was provided by a Grason-Stadler (model 455c) noise generator at all times.

Surgery and Histology--The rats were divided into 3 groups; 10 sustained bilateral lesions of the septum, 5 sustained bilateral lesions of the ventromedial hypothalamus (VMH), while the remaining 5 sustained sham lesions. Under Equi-Thesin anesthesia,

rats were placed in a Kopf stereotaxic instrument. The skull was opened and the midline sinus exposed. With the incisor bar 5 deg above the interaural line, the coordinates for the septal lesions were 2.0mm anterior to bregma, .5mm lateral to the midline sinus, and 6.5mm ventral to the skull surface; a 1.5ma dc anodal current was passed for 20 sec, using an anal cathode. With the dorsal surface of the skull parallel to the table, the coordinates for the VMH lesions were 2.5 posterior to bregma, .5mm lateral to the midline sinus, and .75mm dorsal to the base of the skull; a 2 ma dc anodal current was passed for 15 sec, using an anal cathode. Sham operated rats were treated identically except that the electrode was not lowered into the brain. Following surgery, all animals received .2cc of bicillin.

Following testing, all lesioned rats were anesthetized and perfused with saline and 10% Formalin. Brains were removed, embedded in celloidon, sliced 30 micra thick and stained with cresylviolet.

Procedure--Upon arrival all animals were adapted to the light/dark cycle and the liquid diet for at least 2 weeks. Then, baseline measures of food and water intake and body weight were obtained for 1 week for all 20 animals.

In addition, for 15 animals, a baseline meal pattern of food intake (meal size and post-meal intervals) was obtained for 1 week. Five of these 15 animals were randomly assigned to the sham group and 10 were assigned to the septal group. The rats were then lesioned and returned immediately to their cages for further testing.

Of the rats sustaining VMH lesions, only those who subsequently became hyperphagic and obese were used. Five of 7 rats with VMH lesions showed an immediate postoperative hyperphagia; results for only these 5 rats will be further discussed. The beginning of the static stage was defined as a consecutive 5-day period during which the animals gained less than 3 grams per day. Determination of the static stage was made individually for each animal.

Daily measures of body weight, food intake and water intake were obtained for all animals for one preoperative week and at least 4 post-operative weeks. Meal patterns were recorded for sham and septal rats for the entire preoperative and post-operative period. Meal patterns for the VMH rats were recorded for only 1 week during the dynamic and 1 week during the static stages. Data Analyses--Data was collected for daily body weight, food intake and water intake. Daily intake was analyzed by meal size, meal frequency and post meal interval for each 24 hour period. Meal patterns were determined separately for day and night, and all analyses were made maintaining this distinction. For each animal the average meal size, meal frequency and post meal interval were computed for both day and night for each 24 hour period. Analyses of variance were performed on: (1) daily body weight, (2) daily food intake, (3) daily water intake, (4) average meal size for day and night, (5) average meal frequency for day and night, and (6) average post meal interval for day and night. T-tests were made to locate specific differences.

Comparisons made between sham and septal groups were based on 28 post-operative days. Data obtained for the 7 days during

the dynamic and 7 days during the static stages of VMH were compared to the data obtained for sham and septal groups for post-operative days 14-21. Days 14-21 were arbitrarily selected for comparisons since no differences were found across the 28 post-operative days for either the sham or septal groups.

Meals were defined as a period of licking greater than 10 sec which was separated from other periods of licking by at least 1 min. Since meals were measured in time spent licking, a transformation was made to obtain meal sizes in ml. Each meal (in min) was multiplied by the average amount consumed/minute for that 24 hour period per animal. This produced a measure of meal size in ml.

Correlation coefficients were computed to establish the relationship between meal size and the corresponding post-meal interval for each animal. Individual meals and intervals for every other post-operative day were used; again analyses for day and night were distinct. Correlation coefficients were determined for pre-operative and post-operative data for the sham and septal groups. Pre-operative correlations were determined based on data for 7 pre-operative days and data collapsed over 14 post-operative alternating days for both day and nighttime. Coefficients for the VMH groups were based on meal sizes and intervals for 7 days during the dynamic and 7 days during the static stages.

RESULTS

Daily Measures: Body Weight,
Food Intake and Water Intake.

Two types of comparisons were made to assess the effects of the lesions. (1) Measure of body weight, food intake and water intake were compared pre and post-operatively for each lesion group. (2) The same 3 measures were also compared post-operatively across lesion groups. Post-operative data were collapsed over 7 day periods and the means for each of the 4 post-operative weeks were computed. These means were compared to determine the effect of post-operative time. Figure 1 summarizes the grouped data for body weight, food intake and water intake. Unless otherwise noted all differences are significant at the .01 level.

Body Weight--As shown in Figure 1, body weight remained stable for the sham and septal groups but increased for the VMH group. The analysis of variance showed significant effects for: lesion ($F=16.8, 2/17$), pre-post ($F=112.4, 1/17$) and lesion X pre-post ($F=109.6, 2/17$). Individual comparisons indicated that these differences were due to the large weight gains by the VMH group ($t=10.4, 8$).

The analysis of variance comparing lesion groups post-operatively showed a significant lesion effect ($F=47.7, 2/17$). Individual comparisons indicated, however, that significant differences appear for the VMH group only: VMH rats are heavier than both sham ($t=6.45, 8$) and septal ($t=8.41, 13$) rats.

Significant effects were also found for post-operative time ($F=88.8$, 3/17) and lesion X time ($F=57.5$, 6/51). Inspection of the lower panel of Figure 1 indicates that changes over time occur only for the VMH group.

Food Intake--Pre to post-operative changes in food intake occur only in the VMH group as shown in Figure 1. The analysis of variance showed significant effects for: lesion ($F=29.1$, 2/17), pre-post ($F=15.6$, 1/17) and lesion X pre-post ($F=36.8$, 2/17). Individual comparisons showed that again the significant pre-post changes in food intake are only for the VMH group ($t=9.34$, 8).

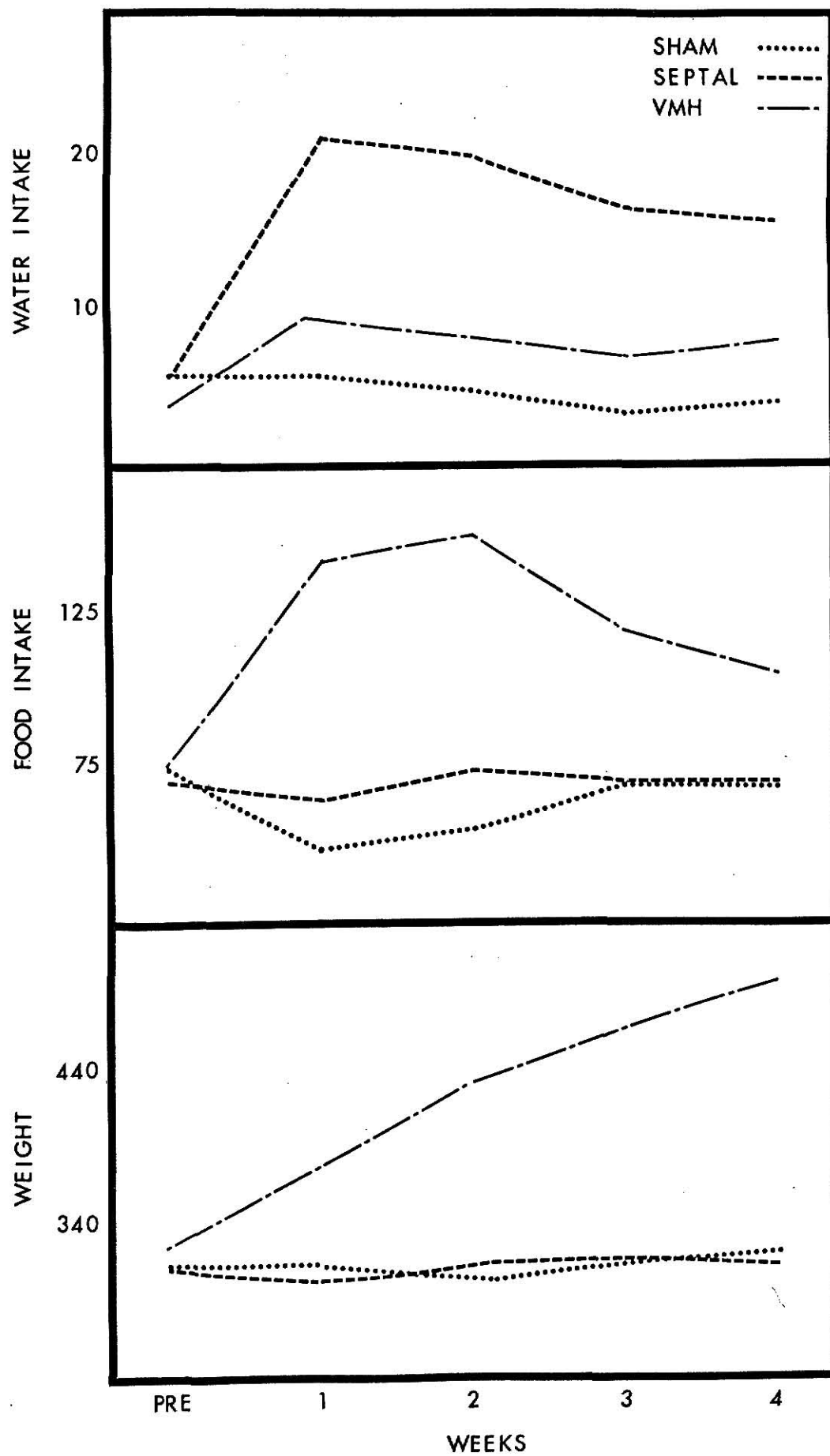
For the post-operative comparisons, the analysis of variance indicated a significant effect for lesion ($F=59.9$, 2/17). Individual comparisons showed that there are significant differences for only the VMH group: VMH rats consumed more than sham rats ($t=11.89$, 13) and septal rats ($t=11.46$, 13). Again significant effects were found for post-operative time ($F=3.95$, 3/41, $p<.05$) and time X lesion ($F=11.8$, 6/51). Inspection of the center panel of Figure 1 shows that the effects of time are due to changes in food intake for the VMH group.

In addition, comparisons of daily intake for the 7 day period were made among VMH-dynamic, VMH-static and sham rats. VMH rats consumed less in the static than in the dynamic stage ($t=4.39$, 8). However, compared to sham rats, static rats still had greater intakes ($t=5.11$, 8).

A measure of the amount of food consumed during the day and during the night was derived by multiplying the average meal size by the average number of meals. Amount consumed was computed separately for day and night intake for each subject.

Figure Caption

Figure 1. Mean body weight (grams) and daily food (ml) and water (ml) intake for one preoperative and four postoperative weeks.



Group means for day and night intake are presented in Figure 2. Sham rats consumed only 25% of their daily intake during the day and consumed the remaining 75% of their daily intake during the night. Septal rats consumed 35% in the daytime and 65% in the nighttime; so while septal rats also consumed most of their intake during the night, they consumed significantly more of their intake during the day than shams ($t=5.48$, 13). In contrast to shams and septals, both dynamic and static VMH rats (combined dynamic and static) consumed more of their total daily intake during the day than shams ($t=6.23$, 13) and septals ($t=6.04$, 18). Water Intake--The analysis of variance for water intake showed significant effects for: lesion ($F=4.02$, 2/17, $p<.05$) and pre-post ($F=4.49$, 1/17, $p<.05$). Individual comparisons showed that both septal rats ($t=4.78$, 18) and VMH rats ($t=2.16$, 8, $p<.05$) increased water consumption over preoperative levels.

Analysis of variance for postoperative effects showed significant differences for the lesion ($F=3.86$, 2/17, $p<.05$). Individual comparisons showed that septal rats drank more than both the sham ($t=4.78$, 13) and VMH ($t=2.84$, 13) rats, while VMH rats drank more than the sham rats ($t=1.92$, 8, $p<.05$). There were no effects for postoperative time.

Water to food ratios were computed to assess the increased water intake of the VMH rats taking into account their increased food intake. The average water (ml)/food(ml) ratio for the sham group was .077, while for the VMH group it was .066. It appears then, that the water intake increases for the VMH group are proportional to food increases. However, the average water to

food ratio for the septal group was .26, indicating polydipsic effect.

Summary--Rats with sham lesions show no pre to postoperative changes in body weight, food intake or water intake. Although septal rats show no differences from either their own preoperative levels, or from postoperative sham levels for body weight or food intake, septal rats did show a significant increase in water consumption over both preoperative and postoperative sham levels. Rats with VMH lesions show significant increases in body weight, food intake and water intake relative to preoperative as well as postoperative levels of the sham and septal groups. In contrast to sham and septal rats who consume most of their intake during the nighttime, VMH rats eat equal amounts during the day and the night.

Meal Patterns: meal size, meal frequency and post meal interval.

The 24 hour daily intake was broken down to examine the distribution of food intake. Data was analyzed by meal size, meal frequency and post-meal interval. For each animal, an average measure of daily meal size, meal frequency and post meal interval was obtained separately for day and night. Daily averages were used in the computation of all statistical tests. All tests were made separately for day and night.

In addition, frequency histograms were constructed for meal size and post-meal interval data. Meals and intervals for every other postoperative day were used for sham and septal groups. Meals and intervals for 7 days during the dynamic and 7 days

during the static stages were used for the dynamic and static groups. Meal sizes for each animal were sorted into 32.5 ml categories, e.g. the first category contained meals of 0-.5ml, the second of .51-1.0ml and the thirty-second of 15.51-16.0. The frequency obtained for each animal for a particular category was added together to determine the lesion group frequency. This group frequency for each category was converted to a percentage of total meals occurring for that lesion group. Histograms for post meal intervals were similarly constructed. There were 15 categories of 10 minute intervals each.

Neither the sham or septal group showed any changes in meal size, meal frequency or post meal interval over the 4 postoperative weeks; effects over time for these groups will not be further discussed.

Figure 2 summarizes the averaged daily measures for day and night for all 4 groups. Sham rats consume equal sized meals during the day as at night, but they eat more meals at night ($t=19.05$, 8). In keeping with the greater meal frequency at night, sham rats have shorter post-meal intervals at night than during the day ($t=19.21$, 8). Figure 3 indicates the distribution of meal sizes, while Figure 4 shows the distribution of post-meal intervals during both the day and the night.

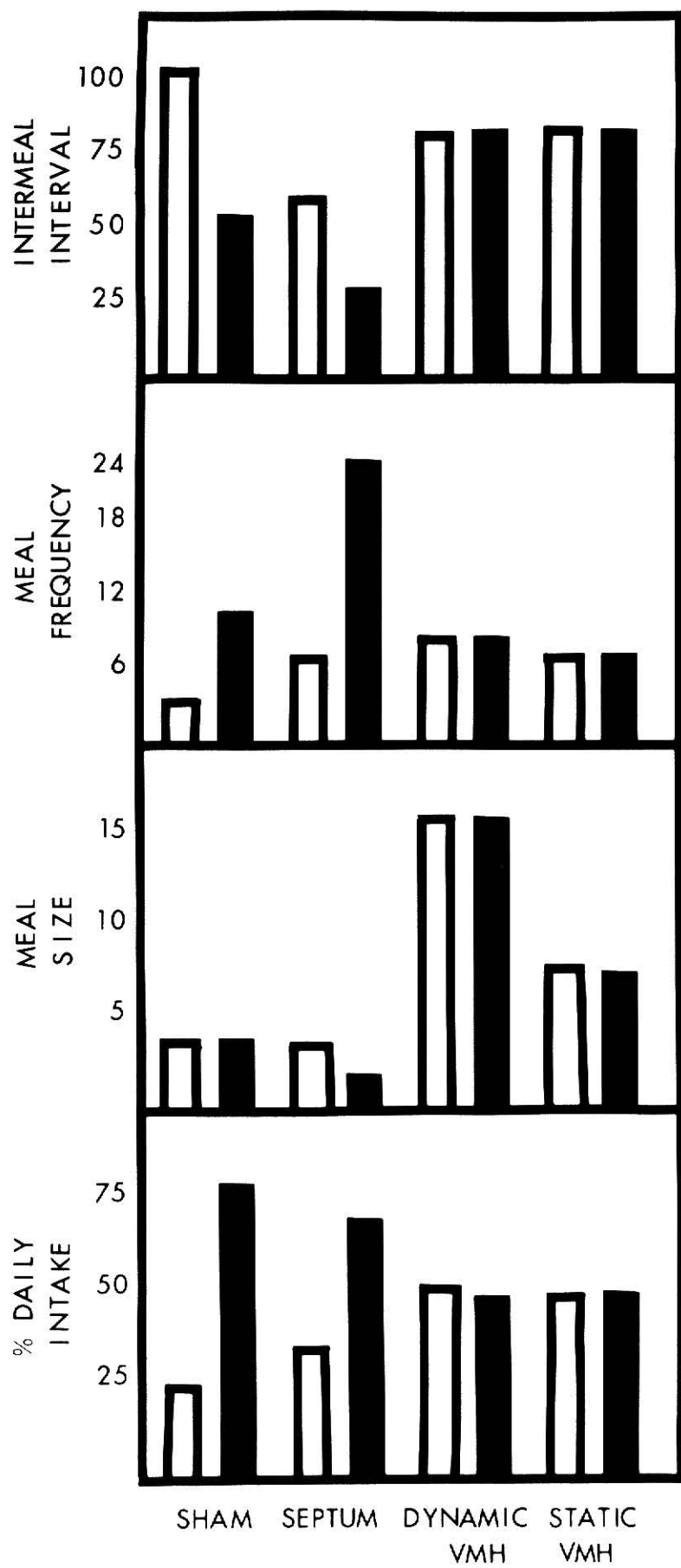
Septal Pattern--In contrast to shams, septal rats eat smaller meals at night than during the day ($t=7.63$, 18). Correspondingly, septal rats eat more frequently at night than during the day ($t=10.57$, 18); they also have shorter post-meal intervals at night than during the day ($t=10.79$, 18).

Figure Caption

Figure 2. Mean measures of percentage total daily intake, meal size (ml), meal frequency and post-meal interval (min.) for the daytime and nighttime for each lesion group.

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Meal size for septal rats is smaller than for shams during both the day ($t=4.11$, 13) and the night ($t=12.24$, 13). In comparison to sham rats, septal rats show higher meal frequencies during both the day ($t=8.48$, 13) and at night ($t=7.0$, 13), as well as shorter post-meal intervals during the day ($t=9.74$, 13) and night ($t=10.86$, 13).

The smaller meal size of the septal group is most easily seen in Figure 3 by comparing the nighttime histograms for the sham and septal groups. For the sham group, only 19% of the night meals are less than 2ml; however, for the septal group, 76% of the night meals are less than 2ml. The shorter post meal intervals at night of the septal group can similarly be seen in Figure 4. For shams, 23% of the post meal intervals are less than 40 minutes while for septals, 83% are less than 40 minutes.

VMH-Dynamic--As shown in Figure 1, dynamic VMH rats consume more per day than sham rats. The increased intake is expressed as larger meal sizes for dynamic rats compared to sham rats at night ($t=17.22$, 8) and during the day ($t=15.87$, 8). In addition, the increase in meal size is equal for both the day and the night, as shown in Figure 2. Also contributing to the increase in intake is a failure of the dynamic rats to decrease the overall frequency of these large meals. Although dynamics eat fewer meals at night ($t=3.42$, 8) they eat more meals during the day ($t=14.17$, 8) than shams. Correspondingly, relative to shams, dynamics show longer post meal intervals at night ($t=12.04$, 8), but shorter post meal intervals during the day ($t=5.01$, 8). Day/night differences in meal frequency and post-meal interval characteristic of the shams,

Figure Caption

Figure 3. Percentage frequency histogram for meal sizes. Percentages are plotted for day and nighttime data for each lesion group.

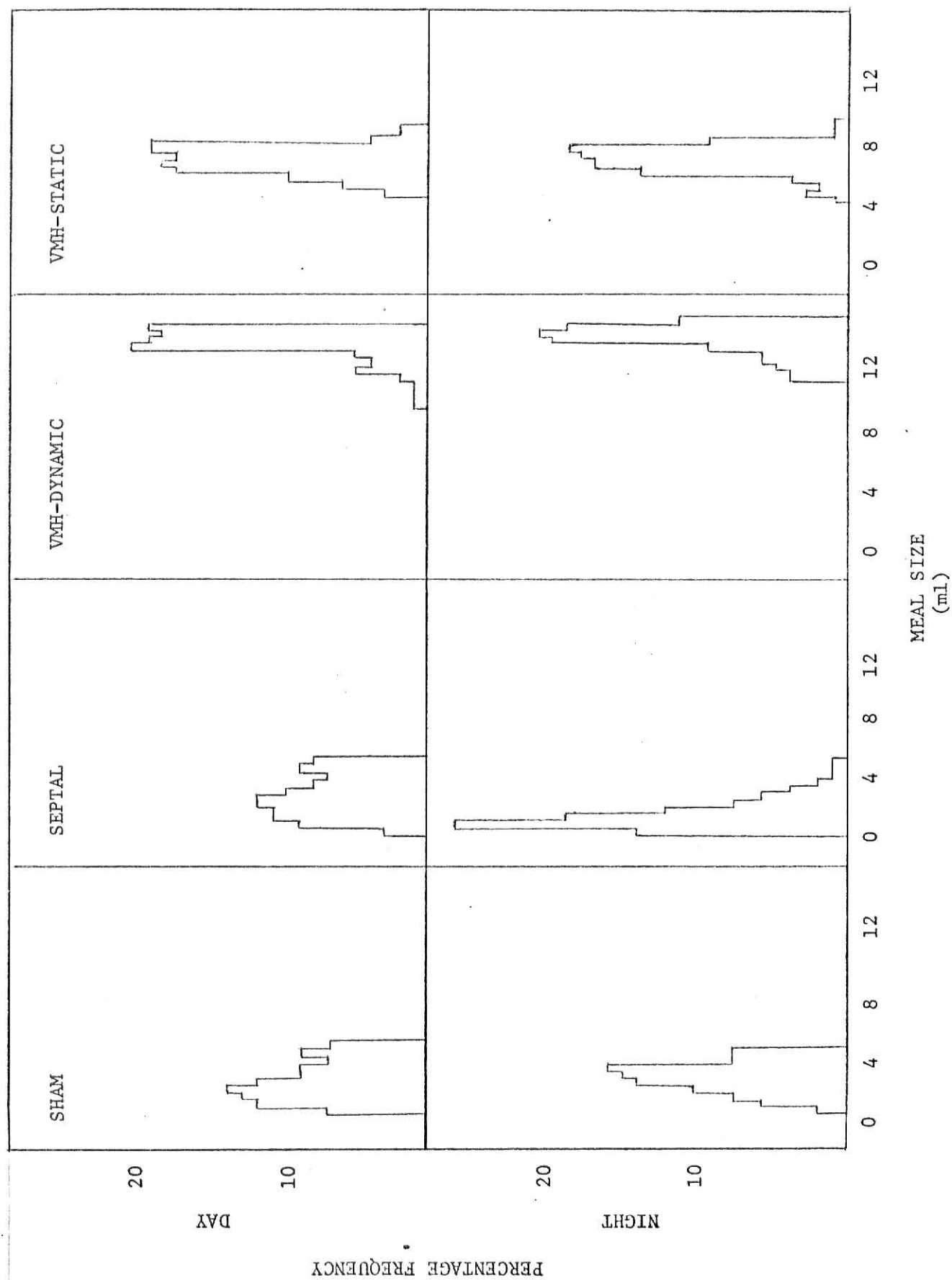
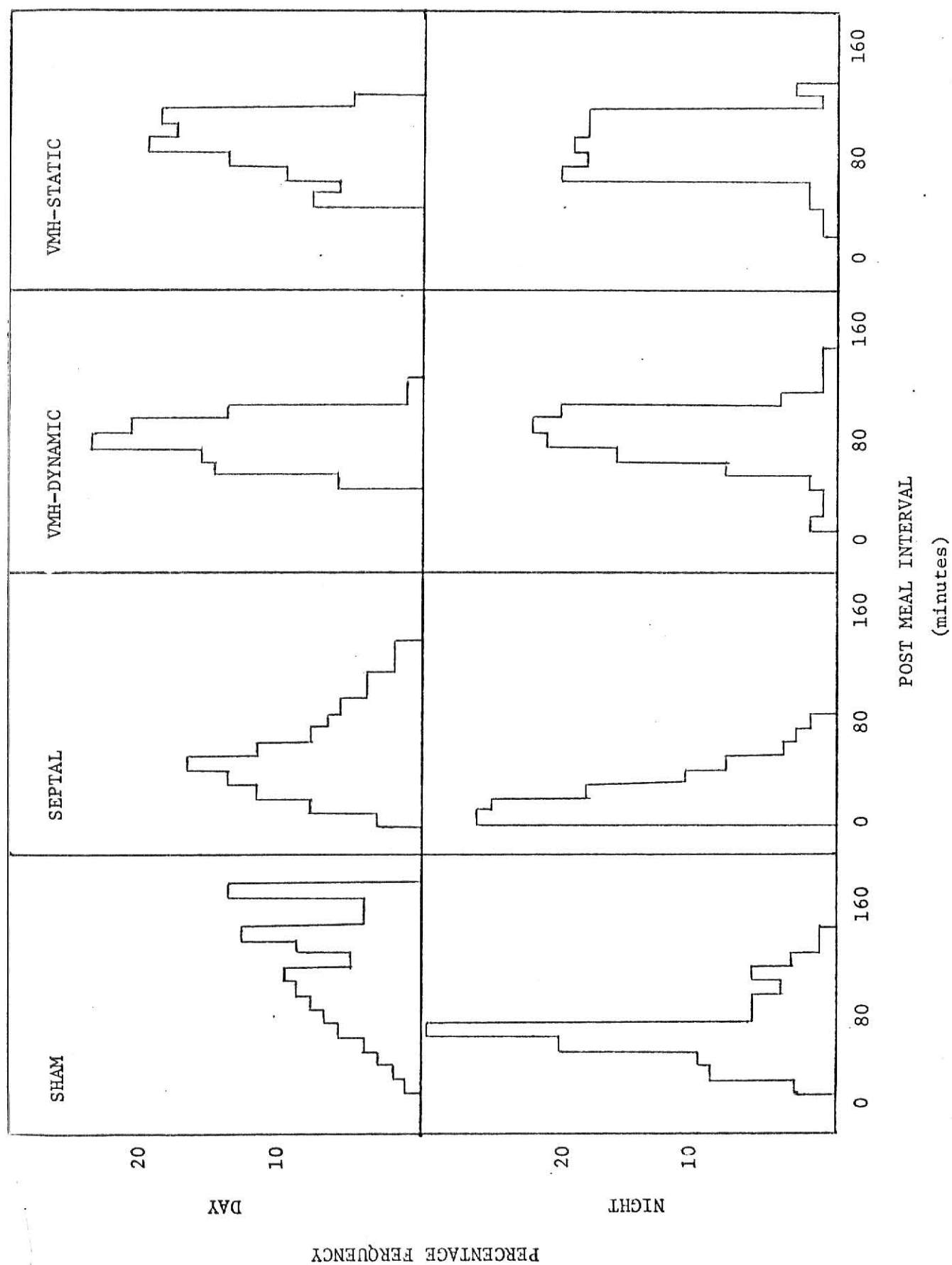


Figure Caption

Figure 4. Percentage frequency histogram for post-meal intervals. Percentages are plotted for day and nighttime data for each lesion group.



are not shown by dynamic rats: meal frequency and post-meal intervals are equal during the day and the night.

Figure 3 illustrates the consistency of the large meals as well as the similarity of the distribution of meal sizes during the day and the night. It should be noted that there is no overlap of the dynamic and sham meal size distributions. In Figure 4 the similarity of the day and night post-meal intervals for the dynamic rats is evidenced by the large overlap of the day and night distributions.

VMH-Static--When VMH rats reach the static stage, their overall intake decreases from the dynamic levels but does not return to preoperative levels. As in the dynamic stage, rats in the static stage show no differences in meal size, meal frequency or post-meal interval during the day compared to during the night as shown in Figure 2. Meal size, like daily intake, decreases from the dynamic to the static stage ($t=10.53, 8$). Although meal frequency decreases are reliable from the dynamic to the static stage during both the day ($t=5.52, 8$) and the night ($t=3.63, 8$), post-meal intervals during the day and night show no changes.

Compared to sham rats, static rats still show greater meal sizes both at night ($t=18.68, 8$) and during the day ($t=14.2, 8$). Meal frequency is greater ($t=11.25, 8$) and post-meal interval is less ($t=4.43, 8$) than for shams during the day. At night however, meal frequency is less ($t=6.85, 8$) and post-meal interval is greater ($t=11.04, 8$) than for shams.

In Figure 3 the shift toward smaller meals from the dynamic to the static stage is shown; it should be noted, however, that

static rats still consume meals much larger than shams. Figure 3 also illustrates the similarity of meal size during the day and night. In Figure 4 it can be seen that the post meal interval distributions for dynamic and static rats do not differ. The large overlap between the day and the night distributions for both dynamic and static rats is indicative of the loss of the day/night cyclicity that is characteristic of the sham meal patterns.

Correlations of Meals and Post-Meal Intervals--Table 1 presents the correlation coefficients between meal size and the post-meal interval. For the sham and septal groups, coefficients were determined for both pre and postoperative data. For the dynamic and static VMH groups, coefficients were calculated using data from 7 postoperative days.

There were no significant differences between pre and postoperative groups or among any postoperative groups. Pre-operatively both sham and septal groups showed positive correlations between meal size and post-meal interval. Post-operative correlations for the sham and septal groups indicate that the lesions did not systematically affect the relationship between meal size and post-meal interval. The magnitude of the correlation also does not seem to be related to differences produced by the day/night cycle.

Comparisons of the postoperative correlations for the VMH rats indicate no day/night differences. In addition, the VMH correlations do not differ from the sham correlations. There also is no change from the dynamic to the static stage.

TABLE 1

Correlations of Meal Size and Post-Meal
Interval for Day and Night

Subjects	Day		Night	
	Pre	Post	Pre	Post
Sham				
1	+.68 ^a	+.59 ^b	+.27 ^a	+.44 ^b
2	+.21	+.25	+.18	+.35
3	+.25	+.54	+.73	+.54
4	+.86	+.55	+.61	+.63
5	+.50	+.34	+.42	+.36
$\bar{X}$	+.50	+.45	+.45	+.46
SD	+.27	+.14	+.22	+.12
Septal				
1	+.30 ^a	+.57 ^b	+.47 ^a	+.71 ^b
2	+.36	+.25	+.59	+.42
3	+.45	+.05	+.46	+.70
4	+.67	+.11	+.57	+.39
5	-.03	+.13	+.59	+.49
6	+.62	+.45	+.61	+.75
7	+.70	+.55	+.16	+.49
8	+.07	+.22	+.49	+.52
9	+.20	+.28	+.45	+.59
10	+.47	+.52	+.33	+.35
$\bar{X}$	+.38	+.31	+.47	+.54
SD	.24	.19	.13	.14
VMH	Dynamic	Static	Dynamic	Static
1	+.42 ^c	+.51 ^d	+.37 ^c	+.45 ^d
2	+.22	+.30	+.26	+.31
3	+.25	+.21	+.41	+.38
4	+.48	+.53	+.59	+.62
5	+.22	+.24	+.42	+.42
$\bar{X}$	+.31	+.35	+.42	+.43
SD	.12	.15	.11	.11

- a. Correlation based on 7 preoperative days.
b. Correlation based on 14 alternated postoperative days.
c. Correlation based on 7 postoperative days during the dynamic stage.
d. Correlation based on 7 postoperative days during the static state.

Histological Analyses

Histological examination of the brains indicated that the septal lesions were in general quite large. The average lesion destroyed all of the medial and most of the lateral septum. There was infrequent minor sparing of the dorsal lateral nucleus. The anterior extent was to the septo-cortical junction while the posterior extent was to the descending columns of the fornix. There was major damage to the nucleus triangularis and nucleus fimbrialis. Occasionally the lesion destroyed the descending columns of the fornix with minor damage to the stria medullaris. Extraneous damage was minimal while in most cases the lesion destroyed all septal nuclei.

Lesions of the ventromedial hypothalamus were total and very symmetrical. In addition to VMH destruction, there was major arcuate nucleus damage. Infundibular damage was insignificant and there was no intrusion of the lateral hypothalamus. There was occasional minor damage to the dorsal medial and periventricular nuclei.

DISCUSSION

Water Intake--Water intake following VMH lesions was higher than preoperative levels as well as sham postoperative levels.

Although few report measures of water consumption in VMH rats, the available data is consistent. The present results, in agreement with Strominger (1947) and Singh and Meyer (1968) showed that water increases were proportional to food increases. This indicates that the increased water consumption is secondary to the increased food consumption on the liquid diet as well as the pellet (Strominger, 1947) and the powdered diets (Singh and Meyer, 1968). The only conflicting study (Kakolewski et al, 1971) that reports increases in the water to food ratio, also reports damage to neural tracts involved in the regulation of the water balance hormone, ADH.

Increases in water intake following septal lesions have been widely reported (Harvey and Hunt, 1965; Blass and Hanson, 1970; Kisseleff, 1969; Wishart and Mogensen, 1970; Kondo and Lorens, 1971; Lorens and Kondo, 1971; Dickinson, 1973). In agreement with these results, reliable increases in water consumption were also found in the present study. This is particularly noteworthy since in all previous work, powdered or pellet chow was used, in contrast to the liquid diet used in this study. Unlike the VMH rat, the septal rat is polydipsic without concomitant increases in food intake.

Food Intake: Daily Intake and Meal Patterns--Rats with sham or septal lesions showed no changes in body weight or food intake over the 5 week testing period. Rats with VMH lesions, however,

showed the classic hyperphagic-obesity syndrome by overeating and rapidly gaining weight.

There has been disagreement among previous studies about changes in body weight and food intake over the postoperative testing period in sham rats. Although most have reported stable food intake, body weights were found to be either stable or steadily increasing. In the present study, both body weights and food intake were stable following sham lesions. This is in agreement with previous findings for female albino sham rats on both a plain powdered diet (Kondo and Lorens, 1971) and a powdered diet mixed with oil (Gold, 1970). However, Snowdon and Wampler (1974) also using female albino sham rats, found steadily increasing body weights using a palatable liquid diet similar to that used in the present study. These conflicting results may be due to the strain of rat or the method of food presentation. The current study used hooded while the Snowdon and Wampler study used albino rats. Although the diet was similar in both studies, Snowdon and Wampler used an operant feeding mechanism while in the present study the diet was available from a drinking spout.

Rats with lesions of the septum also showed no changes in food intake or body weight over the 5 week testing period. In addition, septal rats maintained body weights and intake comparable to sham rats. Only a few studies have dealt directly with ad lib feeding of rats with septal lesions (e.g. Kondo and Lorens, 1971) and most available weight and intake data are only secondarily reported (e.g. Beatty and Schwartzbaum, 1967). In addition there is little consistency in the strain (albino vs hooded), sex

or diet used. Correspondingly, varying results have been obtained. In the only study using females, Kondo and Lorens (1971) report an overall increase in food intake and a transitory increase in body weight in albino septal rats fed a powdered diet. Studies using male albino rats fed on a pellet diet show that septal lesions produce a decrease in body weight (Beatty and Schwartzbaum, 1967; Flarhety and Hamilton, 1971). However, Kondo and Lorens (1971) found no differences between albino septal male rats and their sham controls on a powdered diet. Based on this scanty literature, it is difficult to determine if males with septal lesions generally maintain lower body weights and females higher body weights. The weight gain observed by Kondo and Lorens (1971) in females may perhaps be unique to albinos fed a powdered diet. However, since septal rats over-respond to palatable foods (Beatty and Schwartzbaum, 1967, 1968), it does seem that the sweet liquid diet used in the current study would enhance any tendency in the female septal rat to become obese. It is therefore concluded that females with septal lesions are not hyperphagic and maintain normal body weights.

Lesions of the VMH produced the classic hyperphagic-obesity syndrome. In the dynamic stage of the syndrome, lasting 3-5 weeks postoperatively, the rats were hyperphagic and rapidly gained weight. In the static stage, food intake drastically decreased and body weight stabilized near 500 grams, in agreement with previous reports (Teitelbaum, 1958; Thomas and Mayer, 1968; Gold, 1970; Kakolewski et al, 1971). It should be noted that most previous

studies have used only albino rats, and the present results replicate these using hooded rats.

Measures of daily food intake and body weight have failed to differentiate the effects of septal lesions from sham lesions. More detailed descriptions of the VMH syndrome as well as the effects of septal lesions were obtained by analyzing the daily food intake into its component meals and post-meal intervals during the day and night.

During the daytime sham rats consumed only 25% of their daily total intake; the remaining 75% was consumed during the nighttime. Similar differences in day and nighttime feeding in sham rats were also reported by Balagura and Coscina (1968) and LeMagnen (1971). The lower intake during the day is produced by eating a fewer number of meals. At night rats eat the same sized meals as during the day, but eat more frequently. This increase in meal frequency is in agreement with Balagura and Coscina (1968), Balagura and Davenport (1971) and LeMagnen (1971). However, in contrast to the present results, these studies showed that meal sizes were larger at night than during the day. Since these studies all used pellet rather than liquid diets, the discrepancies may merely be due to differences in the diets.

One way that rats with VMH lesions increase their daily food intake is by eating more during the day. VMH lesions caused a permanent loss of the diurnal cyclicity of intake: VMH rats ate equal amounts during the day and the nighttime, in contrast to shams who ate less during the day than at night. Loss of the

diurnal cyclicity has also been reported by Balagura and Davenport (1970) and Kakolewski et al, (1971).

An examination of daily food intake broken down into meals and post-meal intervals showed that during the dynamic stage, rats consumed very large meals without compensating by increasing the interval between the meals. This confirms earlier results obtained using both liquid and pellet diets (Teitelbaum and Campbell, 1968; Thomas and Mayer, 1968).

Although there was no overall change in the number of meals consumed, the daytime frequency increased while the nighttime frequency decreased; meal sizes were equally large during the day and the night. Therefore, the amount consumed during the day and the nighttime was the same, since meal sizes and meal frequencies were equal. While this shift is in agreement with Teitelbaum and Campbell (1958), Balagura and Davenport (1970) report increases in daytime meal frequency but no change in nighttime frequency.

When the rats reached the static stage of the hypothalamic syndrome, decreases in daily intake were produced by a drastic reduction in meal size and a slight reduction in meal frequency. However, the diurnal feeding cycle remained disturbed: as in the dynamic stage, intake and meal frequency were the same during the day and the night.

The analysis of the daily intake into component meals and intervals as well as the distribution of intake during the day and the nighttime showed effects of VMH lesions previously undetected in more global measures. It has been repeatedly

shown that increases in food intake following VMH lesions are transient and weight stabilizes 3-5 weeks postoperatively. In the present study, it was shown that changes in meal size parallel changes in daily intake; i.e., during the dynamic stage when intake is high rats eat very large meals, but during the static stage when daily intake decreases, rats eat smaller meals. However, the disruption in the diurnal cyclicity of intake is permanent. While intake and meal size decrease from the dynamic to the static stage, VMH rats eat the same amount during the day and the night during both the dynamic and static stages. Formerly the most salient characteristic of the VMH rat was reported to be increases in food intake and subsequent obesity. However, the analysis presented here shows that a more reliable and perhaps basic effect of the VMH lesion is the disruption of the circadian feeding rhythms.

The analysis of daily food intake into component meals and post-meal intervals has also shown effects of the septal lesion undetected in the 24 hour intake measures. Daily food intake was similar for septal and sham rats; in addition septal rats also consumed most of their daily intake during the nighttime. However, an examination of the distribution of meals and intervals showed that septal lesions produce reliable changes in feeding patterns. Septal rats consumed much smaller, more frequent meals than sham rats.

According to LeMagnen (1971) orosensory cues are the major determinant of meal size. The VMH rat who overresponds to orosensory cues, shows increases in meal size. Previous studies

have shown that the septal rat, like the VMH rat, is also over-responsive to orosensory cues (Beatty and Schwartzbaum, 1967; 1968). It was therefore predicted that the septal rat would also show increases in meal size. However, this prediction was not supported, since following septal lesions rats consumed smaller, not larger meals.

Snowdon and Epstein (1970) have reported two other cases in which rats show the pattern of small frequent meals characteristic of septal rats. (1) Rats in which orosensory cues were eliminated by intragastric feeding consumed small frequent meals. (2) Rats in which the sensory input from the esophagus and stomach was reduced by severing the vagus nerve, also showed the small frequent meal pattern. In both cases rats maintained stable intake, but their meal sizes were greatly reduced.

This evidence suggests that the small frequent meal pattern is indicative of an animal that is less responsive than normal to orosensory information. Yet it has been shown that septal rats over-respond to orosensory cues (Beatty and Schwartzbaum, 1967; 1968). In view of this contradiction, it is possible that the septal rat's altered sensitivity to orosensory information is not responsible for the small frequent meal pattern. In support of this possibility is evidence presented by Snowdon (1970) which links the vagotomized rat's small meal patterns to a motor not a sensory deficit.

Snowdon (1970) suggests that meal size is determined by the palatability of the diet. Once in the stomach, the meal empties into the intestine at a rate depending on the osmotic and nutritive

properties of the food as well as the volume of the meal. Snowdon found that the stomach emptying time was highly correlated with the post-meal interval. This implies that another meal is initiated when the previous meal completely passes from the stomach to the intestine. Snowdon demonstrated that in vagotomized rats, liquid diets emptied more rapidly than normal from the stomach to the duodenum. Humans with vagotomies also show this rapid stomach emptying and report accompanying epigastric discomfort and nausea (Roberts, 1967). Small frequent meals eliminated these uncomfortable sensations. Snowdon suggests that the vagotomized rats may eat small meals to avoid sensations caused by the rapid stomach emptying. Small meals empty faster than normal sized meals, and consequently, another meal is initiated sooner, producing the small frequent meal pattern. Thus, Snowdon has accounted for the small frequent meal pattern of vagotomized rats by demonstrating a gastrointestinal motor control deficiency.

Although there are no data available concerning possible gastrointestinal motor deficiencies in septal rats, the behavioral similarity between septal and vagotomized rats is suggestive. The data do not discount hypotheses concerning the septal's over-responsiveness to orosensory cues. However, since it is clear that such hypotheses cannot account for the small frequent meal pattern, an alternative explanation of the septal meal pattern could involve the motor effects of septal lesions. Investigations of the gastrointestinal motor changes following septal lesions may indicate causes for the alterations in the meal pattern.

Correlation of Meal Size and Post-Meal Interval--When LeMagnen and Tallon (1966) first reported the correlation between meal size and the following post-meal interval, it appeared that this relationship would provide a sensitive measure of food regulation. In his investigation of this relationship, LeMagnen (1971) demonstrated the importance of the diurnal control of food regulation. Daytime feeding was characterized by post-meal intervals that were longer than intervals in the nighttime. LeMagnen showed that the relationship, or correlation, between meal size and meal interval is similar during the day and the nighttime, but the magnitude of the intervals differed. The correlations reported were high (.75) and many other investigators began using the correlations as a measure of food regulation (e.g. Snowdon, 1969; Balagura and Coscina, 1969).

Recently the validity of these correlations have been questioned (Panksepp, 1973). Panksepp (1973) has pointed out that the Vincent transformation used by LeMagnen and Tallon (1966) and adopted by others (e.g. Snowdon, 1969), produces spuriously high correlations. A correlation of +.06 obtained with raw data was increased to .35 after application of the Vincent transformation. Panksepp (1973) argued that there is no reliable correlation between meal size and post-meal interval when using raw data. However, it must be noted that Panksepp used an unusual day/night cycle. His rats were maintained on a 12 hour day/night cycle, than were tested in constant light. Since it has been demonstrated that the feeding patterns are at least partially controlled by the light conditions (Siegel, 1961),

it is not surprising that Panksepp's rats failed to show stable feeding patterns. Although Panksepp's procedure leads one to doubt the validity of his nonsignificant correlations, he has shown that the transformations of meal size and interval previously used, did distort the magnitude of the correlations.

The present study has shown that positive correlations are obtained using raw data. In the present study as well as that of Snowden and Wampler (1974), correlations were consistently positive and in the vicinity of .40-.50. Although others have reported near zero correlations, these studies used pellet rather than liquid diets (Balagura and Coscina, 1969; Panksepp, 1973).

Another reason that reported correlations are low is that most studies have failed to compute separate correlations for the day and nighttime (e.g. Thomas and Mayer, 1968; Snowden and Wampler, 1974). As mentioned previously, LeMagnen demonstrated that since the meal patterns differ during the day and the night combining meals and intervals over the 24 hour period will mask the correlation between meal size and the following meal interval.

Although the correlations provide useful information about the relationship between meal size and interval, they have not been very useful in differentiating the effects of brain lesions. In the present study the magnitude of the correlations was not changed as predicted following septal lesions. In addition, postoperative coefficients for shams, septals and VMH groups did not differ, although there were other dramatic differences in the meal patterns of these groups. At least for these lesion groups,

the analysis of the meal sizes, intervals and diurnal cyclicality proved to be more sensitive measures than the correlations in assessing the pattern of food regulation.

FOOTNOTES

1. The modified recipe for 3000 ml of diet was: 26 oz. evaporated milk, 400 ml 50% sucrose, 2 ml Polyvisol vitamins, 100 ml Kaopectate, 40 ml 10% Formalin, 120g. powdered egg and 1250 ml water.

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EFFECTS OF SEPTAL OR VENTROMEDIAL HYPOTHALAMIC
LESIONS ON THE DIURNAL FEEDING PATTERNS OF FEMALE RATS

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Measure of food intake and body weight were compared in female rats with sham, ventromedial hypothalamic (VMH) or septal lesions. In addition, the pattern of food intake during the day and the night was analyzed into meal sizes and post meal intervals. Postoperatively VMH rats increased their food intake and became obese. The increase in food intake during the dynamic stage was produced by two deviations from the shams. In contrast to shams who eat most of their intake during the night, VMH rats increased their daytime eating so that they consumed equal amounts during both the day and the nighttime. They also increased the size of each meal without correspondingly increasing the length of the post-meal interval. When VMH rats reached the static stage, they decreased their intake by eating smaller meals; however, they still consumed equal amounts during the day and the nighttime. It was concluded that the loss of the diurnal cyclicity of intake is a more permanent characteristic of the VMH rat than the increase in food intake. Postoperatively sham and septal rats were comparable in that both maintained stable food intake and body weight over the testing period. However, the analysis of meal patterns showed that septal rats ate very small frequent meals relative to sham rats. Since this type of feeding pattern is also seen in animals with gastrointestinal motor deficits, it is suggested that the motor aspects of feeding should be investigated in rats with septal lesions.