Ph.D. PSYCHOLOGY

Roy A. Wise

ORGANIZATION OF EATING AND DRINKING SITES IN THE LATERAL HYPOTHALAMUS

The degree of independence between the hypothalamic neural systems participating in motivational aspects of eating and drinking in the rat is not established. Using a technique for determining the thresholds of electrical stimulation required for producing eating and drinking at several lateral hypothalamic sites in each subject, the present study found considerable anatomical overlap, but some anatomical separation, between the eating and drinking systems. Within the area of anatomical overlap no indication of structural differences was found: fibre density gradients appeared to be the same. Both eating and drinking could be elicited by electrical stimulation of almost all sites at which either response was initially elicited. The thresholds decreased during the first few days of testing. While final eating and drinking thresholds at a given placement were almost equal, initial thresholds were frequently quite different. These findings suggest an alternative to the recent controversial hypothesis that the neural organization of hypothalamic drive systems is modified by experience.

ORGANIZATION OF EATING AND DRINKING SITES IN THE LATERAL HYPOTHALAMUS

by

Roy A. Wise

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Department of Psychology
McGill University
Montreal

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INTRODUCTION

This study is concerned with the neural mechanisms underlying eating and drinking. The neural substrate that is most clearly identified with these mechanisms lies in the perifornical area of the lateral hypothalamus. Electrical or chemical stimulation of this area can produce eating and drinking in satiated animals (Grossman, 1960; Miller, 1957), while lesions in this area result in a serious disruption of eating and drinking (Anand & Brobeck, 1951; Teitelbaum & Stellar, 1954). Recently, it has been suggested that the neural systems mediating eating and drinking are only two of a larger collection of systems which mediate several species-typical behavior patterns in addition to eating and drinking, such as gnawing, copulation, nest building, grooming, hoarding, and attack (Glickman & Schiff, 1967; Roberts et al., 1967). This whole collection of systems appears to be lodged primarily in the hypothalamic portions of the medial forebrain bundle.

These hypothalamic systems play a motivational rather than a direct motor role in behavior. Actions produced by electrical stimulation at appropriate hypothalamic sites are not rigid or stereotyped: they appear only in the presence of appropriate environmental objects, and are guided by environmental cues (Levison & Flynn, 1965; MacDonnell & Flynn, 1966; Roberts & Carey, 1965; Roberts et al., 1967). Such stimulation can produce not only consummatory responses (e.g., eating), but also instrumental acts (e.g., lever pressing) which in the past have led to the appropriate consummatory objects (e.g., food) (Anderson & Wyrwicka, 1957; Coons, 1964; Miller, 1961). In addition, such stimulation can, when substituted for normally induced "drives," motivate the learning of these instrumental acts (Coons, 1964; Mendelson, 1966; Roberts & Carey, 1965;

Roberts & Keiss, 1964; Roberts et al., 1967). Eating produced by electrical stimulation of lateral hypothalamic "feeding sites" in satiated animals is similar to the eating that occurs normally following food deprivation (Coons, 1964; Tenen & Miller, 1964). It has also been suggested that lesions at feeding sites disrupt the urge to eat, but they do not destroy the animal's ability to make the consummatory movements necessary for feeding (Rogers et al., 1965). Thus these hypothalamic systems seem to mediate the same type of influences that are produced by natural motivational states or drives, such as hunger, thirst, and sexual arousal. They have, therefore, come to be considered as hypothalamic motivational systems.

Little is known regarding the structural aspects of these motivational systems. First, what is the nature of the neural or humoral inputs that affect neural activity in the hypothalamic area? Second, what are the anatomical and functional relations among the various motivational systems within this area? Third, precisely how does the activation of these hypothalamic systems affect the motor systems mediating specific responses, consummatory as well as instrumental, to specific environmental stimuli? This investigation bears on the second of these questions. My specific concern is with the relation between the lateral hypothalamic eating and drinking systems, and this thesis may be regarded as an examination of one specific question: To what extent are the eating and drinking systems independent of one another? Inasmuch as these systems are but two of a number of similar and parallel motivational systems, I shall also examine relevant evidence from studies of other hypothalamic motivational systems.

Evidence bearing on the question of the relation between the lateral hypothalamic eating and drinking systems comes from studies using three general techniques: lesions, chemical stimulation, and electrical stimulation. Investigations with each of the techniques have produced evidence which indicates that in the rat, the hypothalamic eating and drinking systems share the same anatomical locus. Within this area of anatomical overlap there may be functionally independent systems for eating and drinking, or there may be a single neural population which participates in both behaviors. Different techniques have led to conflicting views on this question. The results of studies using each of these techniques will be taken up separately.

Lesion Studies

Lateral hypothalamic lesions which produce aphagia in rats almost always produce adipsia as well (Baillie & Morrison, 1963; Epstein & Teitlebaum, 1964; Gold, 1966; Montemurro & Stevenson, 1955, 1957; Morgane, 1961a, b, c, d, e; Morrison & Mayer, 1957; Teitlebaum & Epstein, 1962; Teitlebaum & Stellar, 1954; Williams & Teitlebaum, 1959). However, some partial anatomical separation is indicated by two studies. Montemurro and Stevenson (1957) found that lateral hypothalamic lesions produced adipsia but did not disrupt voluntary food intake in three animals who were given water by stomach tube; these lesions seemed to be slightly more lateral than those that caused both aphagia and adipsia. Smith & McCann (1962) found lesion sites which produced pure adipsia; lesions producing this effect were either anterior or posterior to the level where lesions cause both aphagia and adipsia.

The possibility that two independent neural networks exist within

the lateral hypothalamic area of anatomical overlap is suggested by the lack of correlation between the rates of recovery of eating and drinking following lateral hypothalamic lesions (see Epstein & Teitlebaum, 1964, Fig. 1, p. 397; and Gold, 1966, Table 1, p. 1275). For example, one animal (Epstein & Teitlebaum, 1964, animal No. 45) recovered eating in less than three days but did not recover drinking in over 100 days, while another animal (No. 37) took much longer to recover eating (about 20 days), but recovered drinking much sooner (about 50 days). Animals with small unilateral lesions, which leave much of the lateral hypothalamic area intact (Gold, 1966), recover eating and drinking functions much more quickly than animals with large bilateral lesions (Teitlebaum & Epstein, 1962). However, the recovery patterns seem otherwise similar, with drinking recovering more slowly and with a positive, but not perfect, correlation between the recovery rates for eating and drinking (Gold, 1966). This suggests that following lateral hypothalamic lesions, recovered eating and drinking are mediated by independent neural networks within the undamaged portions of the lateral hypothalamic systems, rather than by systems in other areas. Thus the evidence from lesion studies suggests that the lateral hypothalamic systems which mediate eating and drinking are composed of functionally independent neural populations which are anatomically interwoven.

Chemical Stimulation Studies

The strongest evidence for the position that eating and drinking are mediated by functionally independent lateral hypothalamic systems has come from the effects of chemical stimulation in this area. Grossman (1960, 1962a) and others (Miller, et al., 1964; Fisher & Coury, 1962) have shown

that lateral hypothalamic application of carbachol elicits drinking while application of nor-epinephrine at the same site elicits eating. Grossman (1962b) suggests that these chemical stimulants have their effect at the synapses of an adrenergically-coded eating system and a cholinergically-coded drinking system, which are anatomically interwoven at the site of stimulation. It is logically correct to conclude that these observed effects of chemical stimulation must be mediated by different neural populations, however there are alternatives to the hypothesis that these independent populations are located at the site of application of the substances in the lateral hypothalamic area.

A recent controversy centering around the interpretation of work done by Fisher and Coury points out one of these alternatives. Fisher and Coury (1962) have used chemical stimulation at several limbic and midbrain sites to produce drinking in satiated animals. Coury (1967) has shown that many of the same structures are involved in adrenergic mediation of eating. On the basis of these findings, they (Coury, 1967; Fisher & Coury, 1962) have suggested that the hunger and thirst drives are mediated by limbic circuits, which would involve the fibres of the medial forebrain bundle as well as the lateral hypothalamic eating and drinking systems. This position is consistent with the assumption that lateral hypothalamic chemical stimulation has its effect directly on lateral hypothalamic systems. Routtenberg (1967) has challenged this view of a limbic "thirst" circuit arguing that the carbachol-induced facilitation of drinking is mediated by the nearby ventricular system rather than by limbic neural networks. His argument rests on the assumption that the chemical stimulation can diffuse far enough to change the ventricular

milieu and affect receptors lining the walls of the ventricle. The issue as to how far the chemical stimulants might spread is not resolved (Fisher & Levett, 1967; Routtenberg, 1967). Grossman (1964) has reported positive and negative sites within 0.5 mm of each other for both acetylcholine and nor-epinephrine stimulation. Coury (1967) reports differences of only 0.25 mm between positive and negative sites for both eating and drinking. Negative placements 0.25 mm from the third ventricle have been reported, while positive lateral hypothalamic sites are as far as 2.0 mm from the ventricle (Grossman, 1962a). Thus it appears that while ventricular diffusion may mediate chemical stimulation effects in other structures in the "thirst" circuit, it seems unlikely that it mediates drinking effects in the lateral hypothalamic area.

Another hypothesis is that there are neural systems which participate in eating and drinking that are separate from the classical perifornical systems, and that it is these systems which are differentially excited by lateral hypothalamic chemical stimulation. Morgane (1960, 1961a, b, c, d, e) has reported eating and drinking systems in the far-lateral portion of the lateral hypothalamic area which are functionally and anatomically distinct from the classical (mid-lateral) systems. These systems involve pallidofugal fibres (Morgane, 1961b; Gold, 1966) which pass transversely across the fibres of the medial forebrain bundle, within 0.25 mm of the classical lateral hypothalamic systems. Morgane (1961a) reports eating elicited by injection of adrenergic substances in this far-lateral region, although he could not elicit drinking by cholinergic stimulation in this area. While this suggests a neural system which might mediate lateral hypothalamic chemical stimulation effects without involving the fibres of

the classical eating and drinking systems, Morgane's results have been recently questioned by Booth (1967) on the basis of a study using more refined stimulation techniques.

Booth (1967) suggests that chemical stimulation affects neural systems which are different from both the mid-lateral, perifornical systems, which appear to be part of the larger medial forebrain bundle network, and the far-lateral systems, which seem to be pallidofugal, and are thought of as not having a purely motivational function (Morgane 1961 b, c). Booth finds that the anatomical localization of sites where very small injections of norepinephrine produce eating does not correspond with the anatomical localization of sites where electrical stimulation produces hunger-like effects and where lesions produce aphagia. Rather he finds that the effective area for adrenergic stimulation is anterior to the lateral hypothalamic sites identified by electrical stimulation and lesion techniques. Booth and Quartermain (1965) have shown behavioral differences between chemically and electrically elicited eating which provide support for the idea that the chemically and electrically induced responses are mediated by different neural systems. Thus Booth (1967) suggests that chemical stimulation using doses large enough to elicit eating from sites in the lateral hypothalamic feeding area has its effect through diffusion to an adjacent neural system which also participates in feeding.

While the differential effects of adrenergic and cholinergic stimulation in the lateral hypothalamic area were earlier taken as strong evidence that the lateral hypothalamic hunger and thirst systems are functionally independent, these recent studies of Booth and Routtenberg reopen the issue. In its present state the chemical stimulation literature

no longer provides clear cut support for Grossman's argument for the chemically coded functional independence of the lateral hypothalamic eating and drinking systems.

Electrical Stimulation Studies

The finding that sites where electrical stimulation produces eating overlap with those producing drinking, has also been taken, until recently, as support for the hypothesis that the lateral hypothalamic eating and drinking systems are functionally independent, although anatomically overlapping. Miller (1957) reports sites which mediate eating but not drinking ("pure" eating), and others that mediate drinking but not eating ("pure" drinking), as well as sites that mediate both eating and drinking. Coons (1964) reports "pure" feeders that would leave a water tube when thirsty in order to eat when electrically stimulated. Similarly, Roberts and Carey (1965) report "pure" gnawers that would leave food to gnaw wood when stimulated in the same area where eating and drinking effects are found. The distributions of the loci mediating these "pure" eating, drinking, and gnawing effects overlap not only with each other, but also with sites where none of these behaviors can be elicited (E.S. Valenstein, personal communication). This overlap of loci suggests the possibility that the lateral hypothalamic area contains several small foci for each of three independent systems mediating eating, drinking, and gnawing, and that these foci are anatomically independent of one another. Recently, however, data have been reported that conflict with these findings and suggest that more than one behavior could have been elicited from each of the reported "pure" sites, if different testing procedures had been used (Valenstein et al., 1968).

Valenstein and his co-workers have suggested that a single drive system, subject to changes in drive-specificity as a function of experience, mediates all of the motivational effects of the lateral hypothalamic area (Valenstein, Cox & Kakolewski, 1968). Their proposal is based on the finding, first reported by Mogenson and Morgan (1967), that electrical stimulation of a particular site that initially elicits only one consummatory response (e.g., eating), can come to elicit a different response (e.g., drinking) as a function of experience. In the study of Valenstein et al., animals with electrodes in the lateral hypothalamic area were electrically stimulated in a cage where food, water, and wooden wedges were available. The intensity of the stimulation was gradually raised until it reliably elicited eating, drinking or gnawing. Stimulation intensity was fixed at this level, and then the consummatory objects appropriate to the observed response were removed (e.g., if the consummatory response displayed by the animal was eating, food was removed from the cage). Stimulation was then continued on an overnight schedule with only the other two consummatory materials present (i.e., water or wooden wedges). Valenstein et al., observed that after a few nights of such stimulation experience a second stimulation-bound consummatory response, appropriate to one of the remaining consummatory objects emerged. When the animal was re-stimulated in a situation with all three stimulus objects once again present, this second consummatory response (e.g., gnawing or drinking) was found to be about as likely to occur as the original response (i.e., eating). The Valenstein group seems to interpret this finding as evidence that the hypothalamic drive systems are plastic, in that the drive-specificity of particular cell populations can be altered by learning.

The data of Valenstein et al., represent the only major findings which are not readily compatable with the view that fixed, independent neural populations mediate lateral hypothalamic involvement in eating and drinking. Some questions raised by this work will be taken up later in the context of the present investigation.

If the hypothesis that the medial forebrain bundle-lateral hypothalamic area contains several motivational systems, each mediating a particular class of species-typical response pattern is correct, it may be profitable to study hypothalamic organization in species with a richer species-typical response repertoire than is displayed by the rat. In a study of stimulation-bound behavior in the opossum, Roberts et al., (1967) have carefully studied a variety of responses produced through many electrodes systematically placed in a variety of sites in the hypothalamus. This study points out two interesting aspects of lateral hypothalamic organization. First, Roberts et al. analyzed each behavior pattern (e.g., male eating behavior) in terms of several distinct components (e.g., mounting, rubbing, biting, etc.). They not only found electrode placements which mediated "pure" effects with respect to major behavior patterns, but they also found placements which mediated various components of a particular behavior pattern, but would not mediate the complete pattern. Sometimes adjacent electrodes, spaced 1.0 mm apart, elicited behavior patterns which were different, but which shared common components. This finding suggests a high degree of specificity of function for the neural elements within these systems.

The second point is that male mating behavior was evoked by stimulation in both male and female subjects. Fisher (1956), similarly, has found that female maternal behavior can be elicited by chemical stimulation in both male and female rats. These findings argue strongly for the position that the medial forebrain bundle systems mediating sexual behavior, at least, have response specificity which is largely independent of experiential factors. While the sites mediating these sexual behaviors are anterior to the lateral hypothalamic area studied by Valenstein et al., (1968), the anatomical and functional similarities between all of the various systems mapped by Roberts et al. (1967) suggest that the response specificity of all these systems may be similarly determined.

The Present Investigation

Two major questions regarding the functional organization of medial forebrain bundle-lateral hypothalamic area motivational systems emerge from the review of these studies. The first is: To what degree do these systems overlap anatomically? While some systems seem to a large extent anatomically independent (e.g., male mating behavior and eating in the opossum), others seem to overlap extensively (e.g., eating and drinking in the rat). Even in systems with great overlap, there may be clusters of response-specific fibres rather than a completely homogeneous interweaving of the fibres of two or more systems.

The second question is this: In areas where two systems overlap, to what extent are the same fibres capable of mediating both responses? The use of the plural "systems" in this discussion has implied an independence between the fibres mediating different responses which has in fact

not been firmly established. It is quite possible that at least some of the fibres in the medial forebrain bundle-lateral hypothalamic area can play a part in more than one response, and also that the nature of the response mediated by these fibres may depend upon experiential variables.

The experiments to be described in the following pages deal with both of these questions, in the specific case of the mid-lateral hypothalamic area of the rat.

EXPERIMENT I

One approach to the question of the functional relation between the lateral hypothalamic eating and drinking systems is to compare their anatomical organizations. To the degree that structural differences can be demonstrated, functional independence may be assumed. The first question which has been asked is whether the anatomical boundaries of the two systems are different. This question has been dealt with in several of the studies reviewed earlier, although the boundaries of the two systems have not yet been clearly established. A second question that might be asked is whether there are structural differences within the boundaries of the systems. One structural difference that would indicate that the lateral hypothalamic neurons mediating eating and drinking are different would be a difference in the distributions of fibre densities in the two systems. A way to obtain data which might reflect the relative densities of the fibres of the two systems would be to measure the strength of response to electrical stimulation of fixed intensity at each of several sites, or, conversely, to measure the intensity of stimulation required to elicit a response of fixed intensity at each of several sites. This experiment was designed to determine the minimal stimulation intensity

necessary to elicit eating and drinking at each of several stimulation sites, as well as to locate the boundaries of the eating and drinking systems in the dorsal-ventral plane. Animals were implanted with electrodes that could be progressively lowered during the experiment, and thresholds for stimulation-bound eating and drinking were measured and compared at several locations in the perifornical area of each subject.

Method

Subjects

The subjects were 14 adult male albino rats of the Wistar strain, weighing 275-300 grams at the time of surgery. They were maintained in individual cages with food and water continuously available.

Apparatus

Intracranial electrical stimulation was delivered by a sixty-cycle sine-wave stimulator (for the circuit diagram see Appendix A). Current was monitored with a microammeter and adjusted with a calibrated potentiometer. The on-off pattern of stimulation was programmed by a timer which opened and closed a relay at appropriate intervals.

Subjects were tested in a box measuring 20 in. long, 10 in. deep, and 16 in. high, which had a mirror as the long back wall and a one-way vision glass as the long front wall. Water bottles were mounted on the wooden side walls, at each end, with drinking spouts extending 1/2 in. into the box at a height of 2 in. from the floor. The box was lighted during testing by a 40 watt bulb in a reflector at the top of the box, with the room lights out. Consummatory responses could be viewed either directly through the one-way glass, or as reflected in the mirror on the back wall.

Surgery

Each subject was implanted with one monopolar stimulating electrode aimed at the lateral hypothalamus, and an indifferent electrode mounted in the skull over the frontal cortex. The stimulating electrode was implanted using a stereotaxic instrument (David Kopf Instruments, Model 900) with the incisor bar located 3.2 mm above the interaural line, using target coordinates 1.5 mm left of the sagittal suture, 0.8 mm posterior to bregma, and 7.5 mm below the superior surface of the skull. Three stainless steel screws were mounted in the skull to anchor the electrode assembly, one lateral, one anterior, and one posterior to the stimulation electrode. The anterior screw served as the indifferent electrode, being connected by a stainless steel wire to a miniature male brass connector which was imbedded in a crown of dental cement which was used to anchor the electrode assembly to the skull screws. Surgery was performed under pentobarbital anesthesia and subjects were injected with 60,000 units of Benzathine penicillin G to reduce the chance of infection.

The electrode assembly and its mounting on the skull are illustrated in Figure 1. The electrode proper was a piece of straightened stainless steel wire 0.01 in. in diameter. The wire was soldered concentrically to a miniature male connector which had been threaded externally with a 2-56 thread die. After soldering, the electrode was dipped in lacquer, tested for insulation leaks, and cut to length. The electrode and pin were then screwed into a threaded nylon cylinder which had been notched to provide anchor points for the dental cement. The receptacle was 1/4 in. in diameter and 3/8 in. long. The receptacle and electrode were located stereotaxically as a unit and then cemented into place.

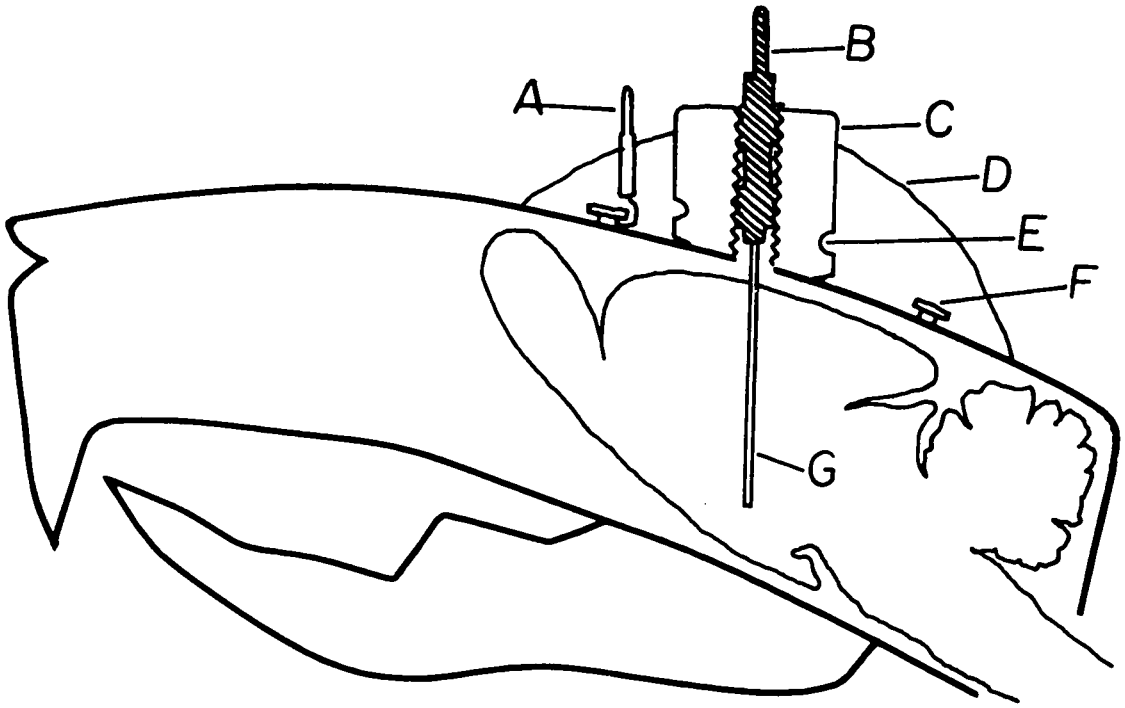


Figure 1 Schematic diagram of mounted electrode assembly for Experiment 1

A miniature connector for reference electrode
B threaded miniature connector for stimulating electrode
C threaded nylon receptacle
D crown of dental cement
E notches for anchoring nylon receptacle in cement
F skull screw
G stainless steel electrode

(See text for explanation)

Procedure

Preliminary testing. Each subject was given preliminary testing with free access to a water bottle at each end of the box, and with food spread liberally on the floor. Sine-wave stimulation was administered on a 20-seconds-on-20-seconds-off schedule. If the subject ate or drank during a period when stimulation was off, it was returned to its home cage for a period of at least 30 minutes. After each stimulation, current intensity was raised 2 to 5 μA, beginning at a level of 5 μA, unless one of the following conditions obtained: (a) a maximum of 100 μA was reached without any observed consummatory responses, (b) forced circling, cringing, jumping or some other response which seemed to be of a motor or aversive nature was observed, or (c) some consummatory response was observed. In the first two cases the animal was returned to its cage and not tested further that day. If, on the following day the same results were obtained, the electrode placement was changed as described below, and testing (as described above) was continued at the new electrode placement. If eating or drinking was observed, the subject was tested for eating and drinking thresholds.

Threshold testing. Eating and drinking thresholds were taken independently for each animal which either ate or drank in the preliminary testing. Each animal was first tested for drinking threshold in a testing box with only water available, then placed for twenty minutes in a testing box with food but no water available, and finally tested for eating threshold. Testing began at the current level best approximating the appropriate threshold as determined in the previous testing session. Stimulation trials were 20 sec. long with a 20-sec. inter-trial interval.

Current intensity was varied from trial to trial as a function of the animal's behavior on the previous trial. If on a given trial the subject ingested food or water, the current intensity was lowered 2 μA for the subsequent trial. Conversely, if a subject failed to make a consummatory response on a given trial, current intensity on the subsequent trial was raised 2 μA. In this way a series of stimulations was administered which involved current intensities varying between some level which always elicited a consummatory response and some lower level which never elicited a consummatory response. A sample record is shown in Figure 2. The first ascending series of stimulations and the first descending series were ignored in order to allow the stimulation level to be adjusted, as a consequence of the animal's behavior, from its initial value (the threshold estimate from the previous testing) to a value which reflected the threshold at the time of the test. The average intensity of the following ten stimulations was taken as the estimated threshold value for the session.

During the experiment, the electrodes were systematically lowered through the lateral hypothalamic area. When an electrode was to be lowered, the animal was placed under ether anesthesia and the exposed electrode pin was clamped in a pin vice. The pin vice was then rotated 1/4 turn, thus screwing the electrode 1/224 in., or 0.113 mm further into the brain.

Thresholds were recorded for six consecutive days at each electrode location except the first location at which stimulation-bound eating or drinking could be elicited. At this first positive site, thresholds were measured for ten consecutive days. Daily testing was found necessary to minimize day-to-day threshold variation; however, in order to

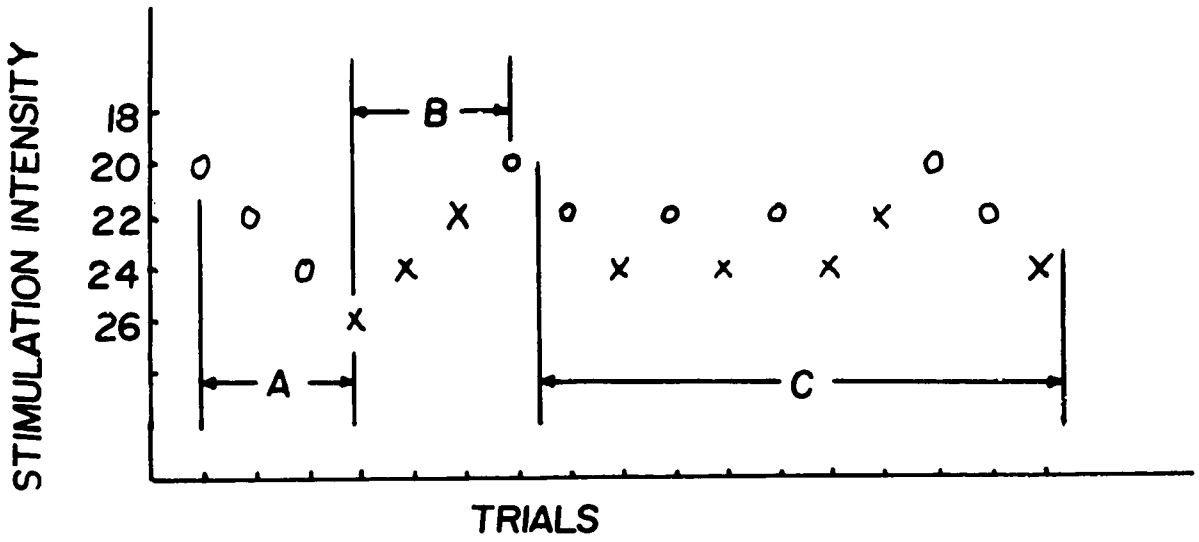


Figure 2 Sample data from a typical eating threshold determination test. O indicates a stimulation trial during which no stimulation-bound eating was observed. X indicates a trial during which food was ingested during stimulation.

A the first ascending series of stimulations
B the first descending series of stimulations
C threshold determination trials

The threshold value computed for these data is 22.6 μA. See text for explanation.

guard against residual anesthetic effects, thresholds were not measured on the day following electrode movement. Thresholds taken on the first day after this rest day tended to be unreliable, so only the data for the last five days of testing at each electrode placement were used in the analysis. After the sixth day of threshold testing at a given level, the electrode was lowered, and testing begun again at the new level, following the same procedure. This procedure was continued until the electrodes had been lowered down to an area where no stimulation-bound eating or drinking could be elicited (or, in the case where stimulation-bound behavior was never elicited from a given electrode, to a level 2.0 mm below the original electrode placement).

Histology. At the end of the experiment, subjects were sacrificed and perfused with saline followed by a 10% formol saline solution. The brains were removed, frozen, cut in 50 u sections, and stained with luxol fast blue and cresyl violet techniques (Gilbert & Nuttall, 1965). Electrode locations for each stimulation point were estimated on the basis of the distance from the bottom of the electrode tract to the placement in question, as estimated from the number of turns used to lower the electrode.

Results

Eating or drinking was elicited from six of the fourteen subjects. Of these six, complete data could not be obtained from two subjects. Subject 12M became sick and died while its electrode was still in a location that yielded eating and drinking; data regarding the lower boundary of the eating and drinking systems were not obtained from this

animal. The initial electrode location for subject 14M turned out to be within the eating and drinking systems, so that data regarding the upper boundaries of the systems could not be obtained from this animal.

Median values for the five eating thresholds and the five drinking thresholds taken at each stimulation site are shown in Figure 3. The figure also shows the range of the five eating and five drinking thresholds at each site. As mentioned earlier, threshold testing was continued for 10 days at the first site where stimulation-bound eating or drinking was observed. A systematic decline in thresholds was observed during the first few days of testing at this site, but the thresholds were quite stable during the last five days; therefore, only the data for the last five days of threshold tests at this first site are included in Figure 3. The data from the first five days of testing at this site and the implications of this decline in threshold will be discussed in the next experiment.

Large threshold changes were produced by electrode movements at the boundaries of the eating and drinking systems. At the dorsal boundaries of the systems, a movement of 0.113 mm was sufficient to change both eating and drinking thresholds from some value above 100 μA to a value which was typical of all the more ventral placements in that particular electrode tract; such a slight movement could change the threshold from above the 100 μA maximum stimulation used in the experiment, by as much as 80 μA, to a value approximating the minimum threshold for that tract. After this initial threshold drop which occurred as the electrode entered the eating and drinking systems, placement to placement threshold variation was small. As the electrodes approached the ventral boundaries of

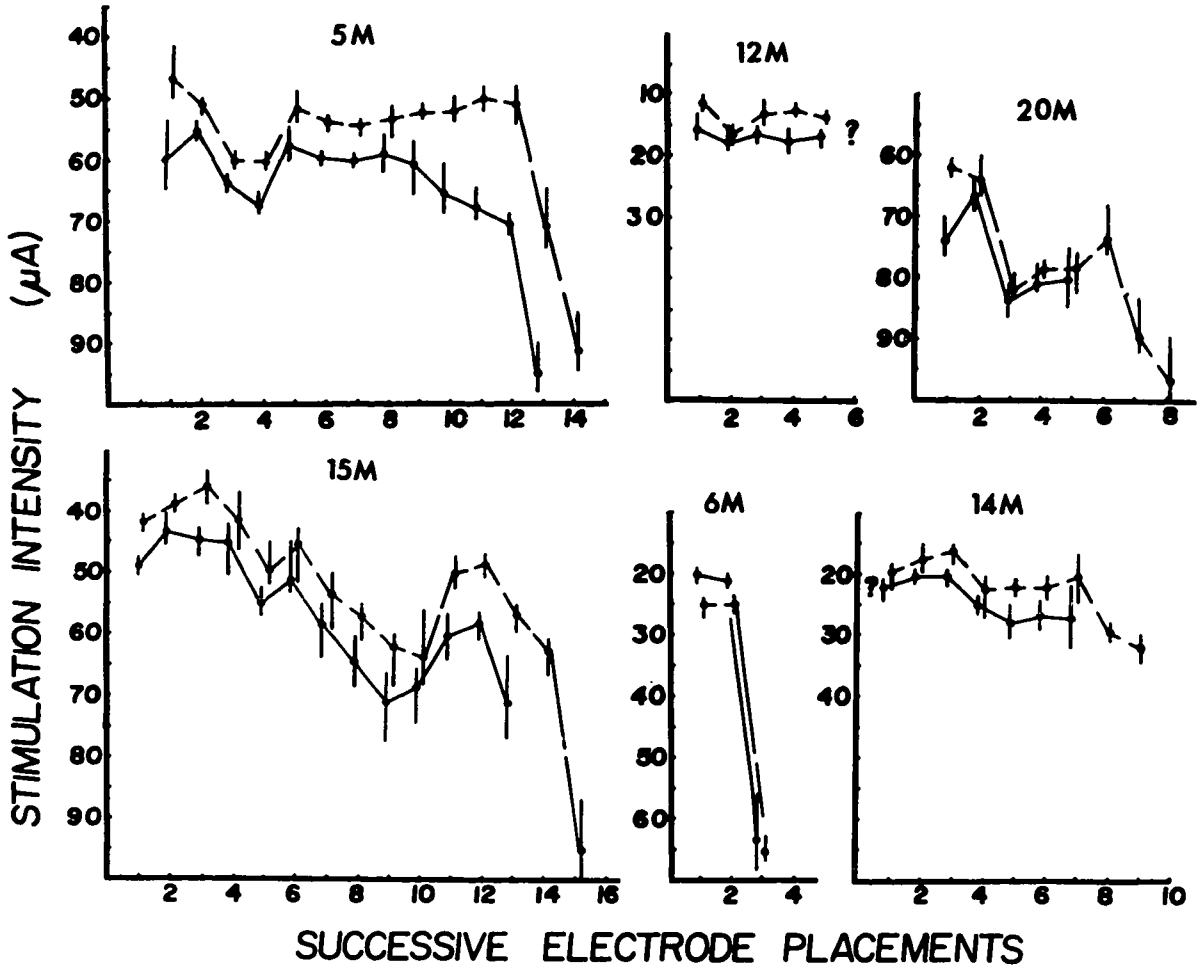


Figure 3 Median intensity thresholds for electrically elicited eating and drinking at successive lateral hypothalamic electrode placements in Experiment I. Solid lines connect successive median eating thresholds; broken lines connect drinking thresholds. Ranges for the five thresholds taken at each electrode placement are indicated by vertical lines. Electrode placements numbered (1) represent the most dorsal site where eating or drinking could be evoked. Each successive placement is 0.113 mm more ventral. Subjects are identified by the numbers at the top of each graph. Question marks at placements one and six for subjects 14M and 12M respectively indicate that thresholds are not available for these placements. For all other placements prior to placement one, and for all other placements subsequent to the last threshold shown for each animal, thresholds were higher than the 100 μA maximum stimulation level used in the experiment.

the systems, large threshold changes were again observed between placements, although two or three 0.113 mm movements were usually required to produce threshold changes of the same magnitude as were produced by one such movement at the dorsal boundary of the systems. Within the dorsal and ventral boundaries of the systems, which were separated by as much as 2.0 mm, no sites which failed to produce eating or drinking were observed in any positive electrode track ("positive" will be used to indicate electrode placements or tracks which yielded stimulation-bound eating or drinking; "negative" will not be used to imply aversive properties of stimulation, but will simply indicate placements or tracks where no stimulation-bound eating or drinking could be elicited).

While most stimulation sites yielded both eating and drinking, a few sites were found which mediated only drinking. These sites were always found to lie just ventral to the most ventral sites mediating both eating and drinking in a given electrode track.

Just as threshold variation at various locations within a track were minor, so also were variations between eating and drinking thresholds within a track. Drinking thresholds were consistently a little lower than eating thresholds in five of the subjects, and consistently a little higher in the sixth subject. In contrast to this consistency of thresholds within particular tracks of individual subjects, threshold variation between animals was great.

Histological reconstruction of positive and negative tracks is shown in Figure 4. Positive placements were all in the perifornical area with the dorsal boundaries near the level of the zona incerta and the ventral boundaries parallel to, and about 0.5 mm above, the sloping base of the

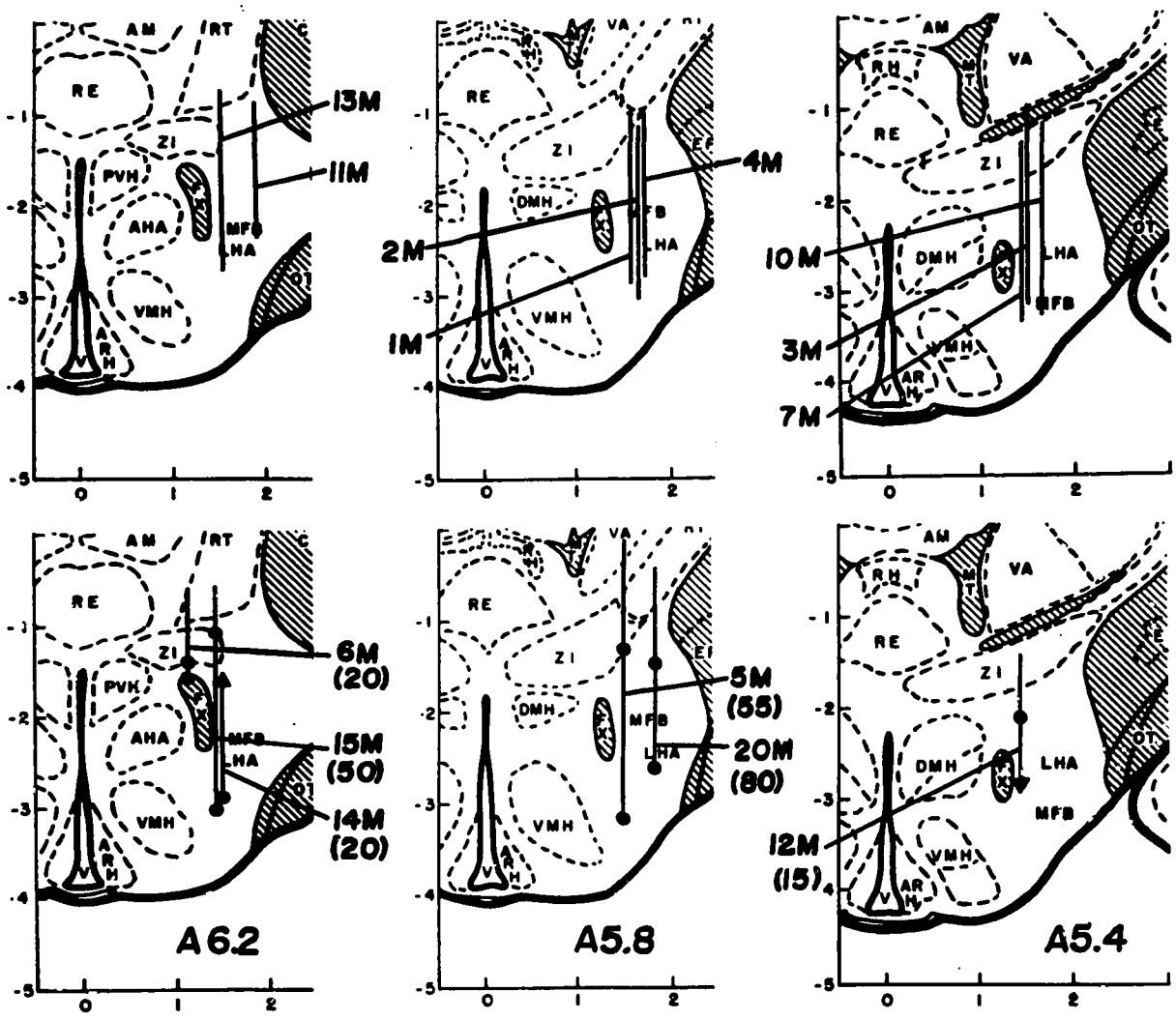


Figure 4 Histological reconstruction of electrode placements for Experiment I. Upper sections show stimulation tracks which failed to yield stimulation-bound eating or drinking. Lower sections show stimulation tracks which mediated eating and drinking effects. Positive effects were obtained from those portions of each track bounded by ●. Stimulation was not attempted above the point indicated by ▲ in Subject 14M nor below the point indicated by ▼ in Subject 12M, as noted in the text. Representative thresholds for each track are indicated in parentheses. Brain sections from de Groot (1959) with anterior planes noted.

brain. No simple anatomical distinction could be made between positive and negative placements or between placements yielding high thresholds and those yielding low thresholds.

Discussion

The dorsal and ventral boundaries of the eating and drinking systems delineated in the present experiment appear to correspond roughly to the most dorsal and most ventral placements mediating eating or drinking as reported in previous studies (e.g., Miller, 1957; Morgane, 1961a). Within animals showing positive effects, the systems appear to be continuous between these bounds, contrary to what might be expected from the results of comparisons of placements mediating different effects in different animals (Miller, 1957; E. S. Valenstein, personal communication; see also Experiment 3). In the present study no negative site was ever found, in a positive tract, within the boundaries of the eating or drinking systems.

Another finding worth mentioning is that while the eating and drinking systems are largely co-extensive, the drinking system extends slightly further ventral than does the eating system. Thus, at least some lateral hypothalamic neurons that play a role in drinking are not involved in eating.

Within the area of anatomical overlap of the two systems, no differences in threshold gradients were observed which would suggest that independent systems mediate the two responses. While some difference in threshold gradients was observed in subject 5M between the 10th and 12th weeks, the gradients were remarkably parallel in the other subjects, as they were in this subject during the first 9 weeks of testing.

Not only were the threshold gradients parallel, but they were also nearly equal in average absolute value. Drinking thresholds were slightly, though reliably, lower than eating thresholds in five of the six subjects. However, this difference in threshold is most likely due to "task" differences (e.g., a difference in the effort required for eating and drinking), or to other particular aspects of the experimental method, rather than to differences in concentration of eating and drinking cells at the electrode tip. Incidental observations in this laboratory indicated that threshold changes of this magnitude can easily be produced by changes in palatability of the food used, changes in the time of daily testing, and changes in the size of the food pellets used. Changes in one of several arbitrarily chosen details of procedure could probably eliminate any apparent difference between eating and drinking thresholds.

A perplexing problem which remains unsolved is to explain the individual differences in responses to stimulation. The negative electrode tracts shown in the upper half of Figure 4 are not anatomically distinguishable from the positive tracts shown in the bottom of the figure. Further, the individual differences in threshold between animals with positive placements do not seem correlated to anatomical locus in any simple way. Even after several nights of overnight stimulation experience, negative sites are found in some animals which appear to be within the area yielding positive results in other animals (E.S. Valenstein, personal communication). Examination of these differences between positive and negative sites in different animals leads to the hypothesis that the lateral hypothalamic area contains a very heterogeneous arrangement of eating, drinking, gnawing, and negative fibres.

However, the comparison between sites within each animal in the present study revealed a very homogeneous and continuous distribution of eating and drinking fibres throughout the lateral hypothalamic area of animals with positive placements. Further comparisons within animals, exploring the anterior-posterior and medial-lateral dimensions of these systems are needed to determine whether heterogeneity exists within animals but was not revealed by mapping the vertical dimension only, or whether there are real differences in hypothalamic structure between animals.

EXPERIMENT 2

The observations reported here as Experiment 2 do not truly constitute an independent experiment. Most of the data reported were collected in the course of Experiment 1. However, as results from some animals that did not participate in Experiment 1 are included, and as the fundamental question discussed in this section is quite different from the questions raised in Experiment 1, the present organization appeared to be more appropriate--at least, more convenient for the reader.

As noted in the Introduction, Valenstein et al. (1968) have shown that after a certain type of stimulation experience animals will perform stimulation-bound responses that were not performed before such stimulation experience. They have interpreted this finding in a way which implies that stimulation experience could alter the efferent connections between specific lateral hypothalamic neural populations and the motor systems mediating drive-specific responses. However, the hypothesis that the connections between the motivational and the motor systems are plastic is not necessary to explain their findings. The results of Experiment 1

suggest the alternate hypothesis that stable independent drive systems, which have fixed efferent connections, are simultaneously activated by electrical stimulation, and that stimulation experience merely changes the thresholds of the different systems without altering the functional organization of any. In the study of Valenstein et al., stimulation of fixed intensity was used throughout. The argument presented here is that this fixed stimulation intensity was sufficient to elicit only one response at the time of first testing, but that the stimulation thresholds changed as a result of continued testing, such that this intensity later came to be sufficient to elicit a second response as well.

Threshold changes consistent with this hypothesis occurred during the first few days of threshold testing at the first positive placement in each subject in Experiment I. Since these placements were at the dorsal boundary of the eating and drinking systems, additional animals were also tested for progressive threshold changes in an attempt to examine stimulation sites in the middle and ventral portions of the eating and drinking systems.

Method

The subjects were the six subjects with positive placements used in Experiment I, and seven additional adult albino rats of the Wistar strain weighing 275-300 grams at the time of surgery.

The seven new subjects were implanted with permanent bilateral monopolar electrodes; a skull screw over the frontal cortex served as the reference electrode. The stimulating electrodes were of the same type as those used in Experiment I. Electrodes were located stereotaxically

with target coordinates 0.8 mm posterior to bregma, 1.5 mm lateral to the midsagital suture, and 8.2 mm below the superior surface of the skull. Electrodes were soldered to miniature brass connectors and imbedded in a crown of dental cement which anchored the assembly to the skull screw used as the indifferent electrode, as well as to two additional skull screws located posterior to the stimulating electrodes. Other surgical details were the same as in Experiment I.

Subjects were given preliminary testing as in Experiment I, and those which ate or drank when stimulated were tested daily for drinking threshold and for eating threshold. Daily threshold testing was continued for 10 days and then the subjects which were used in this experiment only were sacrificed and their electrode placements histologically determined as in Experiment I.

Results

Two of the new subjects showed stimulation-bound responses on one electrode each. Daily thresholds for eating and drinking from these animals as well as for the six subjects from Experiment I are shown in Figure 5: note that the data from the animals used in Experiment I are from the first 10 days of threshold testing at the first positive electrode placement.

Thresholds for both responses decreased over the first 10 days of testing in all subjects. Final thresholds for both eating and drinking were lower than the initial thresholds for either response. The differences between initial and final thresholds varied from 1.8 μA (Subject 12M, eating) to 47 μA (Subject 15M, eating).

In histological reconstructions, the electrode placements for both the new subjects were found to be in the anterior plane 5.4 of de Groot

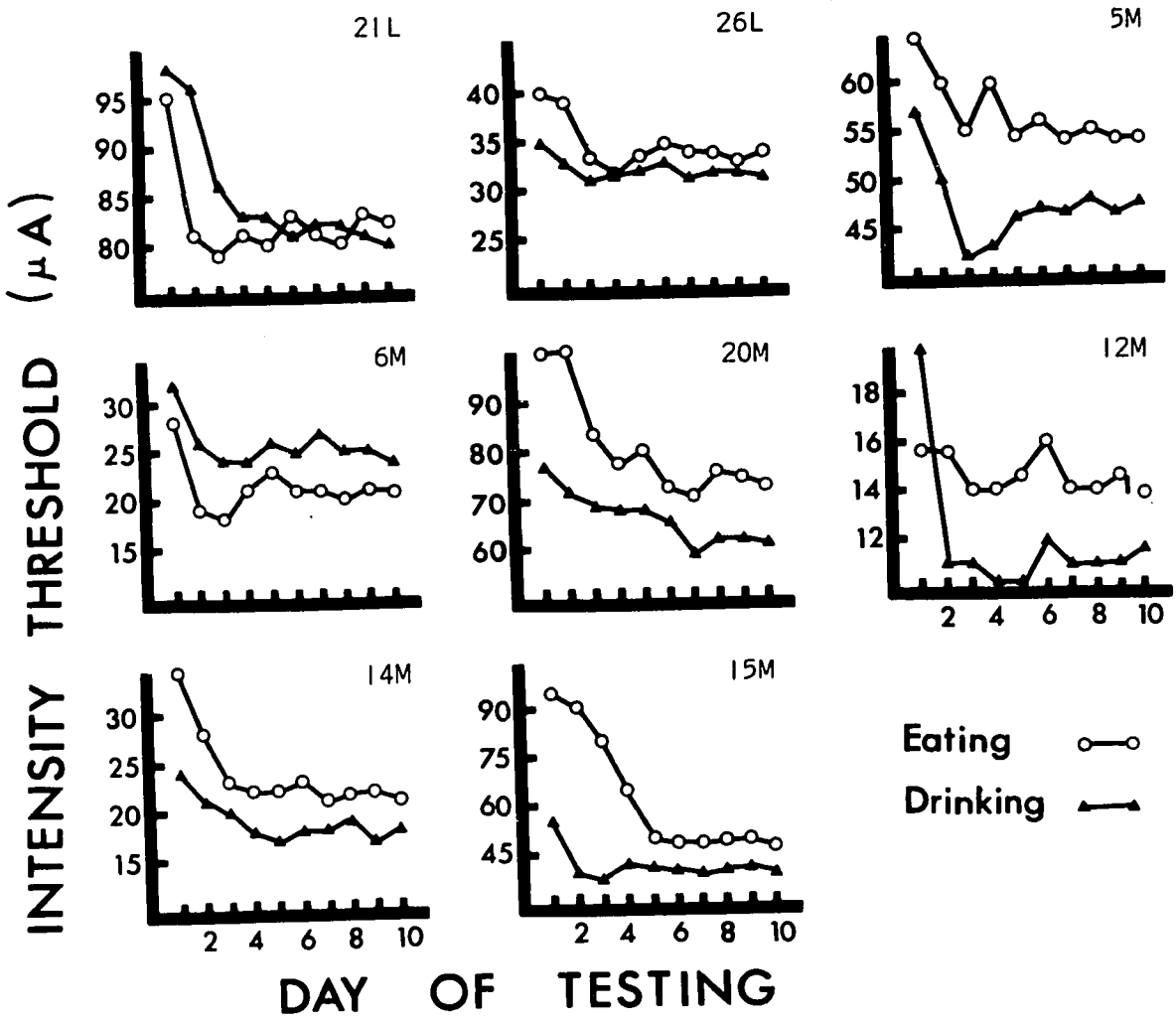


Figure 5 Daily thresholds for eating and drinking elicited by electrical stimulation of the lateral hypothalamus in Experiment 2.

(1959), just lateral to the fornix, one at the level of the mid-fornix, and the other at the ventral boundary of the fornix. Electrode placements for the six subjects from Experiment I are shown in Figure 4.

Discussion

The results show that threshold changes of sufficient magnitude to explain the findings of Valenstein et al., (1968) did occur as a result of repeated lateral hypothalamic stimulation. The fixed stimulation intensity used in the Valenstein et al. study was the level required to first elicit a response. In the present study this level was usually quite a bit higher than the "initial" threshold, which was determined after some stimulation experiences in the preliminary testing phase. Since the final thresholds of eating and drinking in the present study were always below the initial thresholds for both eating and drinking, which represent current levels below the level which would have been used in the procedure of Valenstein et al., it is apparent that these threshold reductions could explain their observation of the emergence of a response after stimulation experience, which could not be elicited before experience.

It is interesting to note that no progressive threshold changes were observed in Experiment I, except at the first positive electrode site. This suggests that rather than causing some change such as sensitization of the specific neurons activated by electrical stimulation, the stimulation experience may have some more general effect. For example, conflicting responses such as freezing or exploration which might prevent the occurrence of the consummatory response, may be elicited

by initial stimulation, but then habituate, allowing consumatory responses to emerge. A second possibility is that some general features of the testing situation, such as the location of the drinking spout are learned during the initial stimulation periods. Animals, when first given drinking experience, seem not to know where the water tube is. If they happen to be near a tube they will drink at intensities which do not produce drinking when they are further away from the tube. After several days of testing, stimulation at near-threshold intensities reliably causes the animal to go to the water spout regardless of its (the animal's) location in the testing box.

This experiment indicates one effect of stimulation experience on stimulation-bound behavior which is consistent with the hypothesis that separate systems with fixed efferent connections mediate lateral hypothalamic electrical stimulation effects. The threshold changes observed in this experiment are sufficient to explain the results of the Valenstein et al. (1968) experiment as well; there is no need for the assumption that stimulation experience produces any further changes in lateral hypothalamic systems, such as changes in the drive-specificity of cell populations at the stimulation site.

EXPERIMENT 3

Valenstein et al. (1968) have shown that, after appropriate electrical stimulation experience, more than one consummatory response can be elicited from all or almost all lateral hypothalamic electrode placements that initially appear to mediate only one response. On the basis of this finding they suggest that experience plays an important role in determining the drive-specificity of stimulation at these sites, and they imply

that the neural populations mediating eating, drinking, and gnawing in the rat may be equipotential for the mediation of all of these responses.

This experiment was designed to answer two questions regarding the findings of Valenstein et al. First, is stimulation experience a necessary condition for obtaining two or more responses to electrical stimulation from positive lateral hypothalamic sites? Experiments I and 2 suggest that more than one response might be elicited from most sites without prior stimulation experience, if proper testing techniques were employed. An aspect of testing procedure that seems important is this: If different responses have different thresholds, one may be dominant at any given stimulation intensity, and may mask the less dominant response when testing is done in a competitive situation. For example, eating may never be observed from a placement with a high eating threshold if the subject is tested with water available and that placement has a low drinking threshold. Consequently subjects should be tested independently for each response in question. Another aspect of testing procedure that seems important is that since different responses may have very different thresholds, stimulation intensities much higher than are required to elicit an initial response should be used if lower intensities have no effect.

The second question dealt with in this experiment is whether appropriate stimulation experience is a sufficient condition for the development of new responses to lateral hypothalamic stimulation. If it is, then eating, drinking, and gnawing should be obtained from all positive lateral hypothalamic stimulation sites, after appropriate stimulation experience with all of the appropriate consummatory objects. If two, but only two,

responses can be elicited from some perifornical sites, it would be difficult to maintain that this area was equipotential for eating, drinking, and gnawing.

Method

The subjects were 18 adult male albino rats of the Wistar strain, weighing 275-300 grams at the time of surgery. They were maintained in individual cages with food and water continuously available.

The stimulator and testing box used in Experiment I were again used in this experiment. Two wooden wedges, 6 in. long, with triangular cross section, having 1 1/2 in. sides, were placed in the otherwise empty testing box when gnawing tests were made. Food or water were made available for the eating or drinking tests as in Experiment I.

Subjects were implanted with monopolar, bilateral, lateral hypothalamic electrodes as described in the method section of Experiment 2.

Subjects were given preliminary testing as in Experiment I, but with wooden wedges as well as food and water available. Subjects showing stimulation-bound eating, drinking, or gnawing in preliminary testing were then tested independently for threshold of each response, prior to any further stimulation experience. Following threshold testing, subjects that did not show all three responses to stimulation were given overnight stimulation experience with the consummatory object not responded to in the threshold tests. If a subject made only one consummatory response during initial threshold testing, it was first given overnight stimulation experience with one of the ramaining objects, then given a threshold test with this object, and then this whole procedure was repeated with the remaining consummatory object.

Overnight stimulation experience was given following the procedure of Valenstein et al. (1968). Subjects were placed in the testing box for twelve hours each night with only one consummatory object available. Thirty seconds of stimulation was administered every five minutes. Intensity of stimulation was fixed at the level which first produced any of the three stimulation-bound behaviors in preliminary testing. Overnight training was continued until five nights of training were completed. Subjects were then given threshold testing for the response in question and subsequently sacrificed for histological purposes.

Results

Stimulation-bound responses from 24 of the 36 electrode placements were obtained during preliminary testing. Initial threshold testing revealed that 23 of these placements were capable of mediating more than one response: stimulation at 15 of these placements produced all three responses, while stimulation at 8 placements produced only two responses, and one placement yielded only one response. Thresholds for eating, drinking, and gnawing upon initial testing are shown in Table 1.

After overnight stimulation experience, there remained one placement which yielded drinking only, one that yielded eating and gnawing but not drinking, and five that yielded eating and drinking but not gnawing. Thresholds for the three responses after overnight stimulation experience are also shown in Table 1.

Histological reconstructions of electrode placements are shown in Figure 6. Positive placements were distributed in the perifornical area, with the placements which yielded only one or two responses all lying

TABLE I

Subject	Before			After		
	Eat	Drink	Gnaw	Eat	Drink	Gnaw
30L	22	5	24			
30R	100	50	54			
32L	32	22	27			
32R	56	32	42			
33L	28	14	no			no
33R	60	44	no			no
34L	no	no	no	no	no	no
34R	50	28	38			
35L	30	30	26			
35R	26	19	18			
36L	no	no	no	no	no	no
36R	no	no	no	no	no	no
37L	28	8	no			no
37R	25	19	no			21
40L	no	no	no	no	no	no
40R	43	37	35			
41L	28	11	17			
41R	16	11	10			
42L	35	21	no			no
42R	17	1	12			
43L	no	no	no	no	no	no
43R	35	no	34		no	
44L	39	26	no			80
44R	no	no	no	no	no	no
45L	no	no	no	no	no	no
45R	10	10	14			
46L	40	30	38			
46R	17	12	18			
47L	no	17	no	no		no
47R	no	no	no	no	no	no
50L	no	no	no	no	no	no
50R	25	24	no			no
51L	no	no	no	no	no	no
51R	no	no	no	no	no	no
52L	no	no	no	no	no	no
52R	21	18	22			

Table I Intensity thresholds in μA for electrically elicited eating, drinking, and gnawing before and after overnight stimulation experience in Experiment 3. "No" indicates that the response in question was not observed at stimulation intensities below 100 μA. Only responses not observed in initial testing were tested after overnight experience.

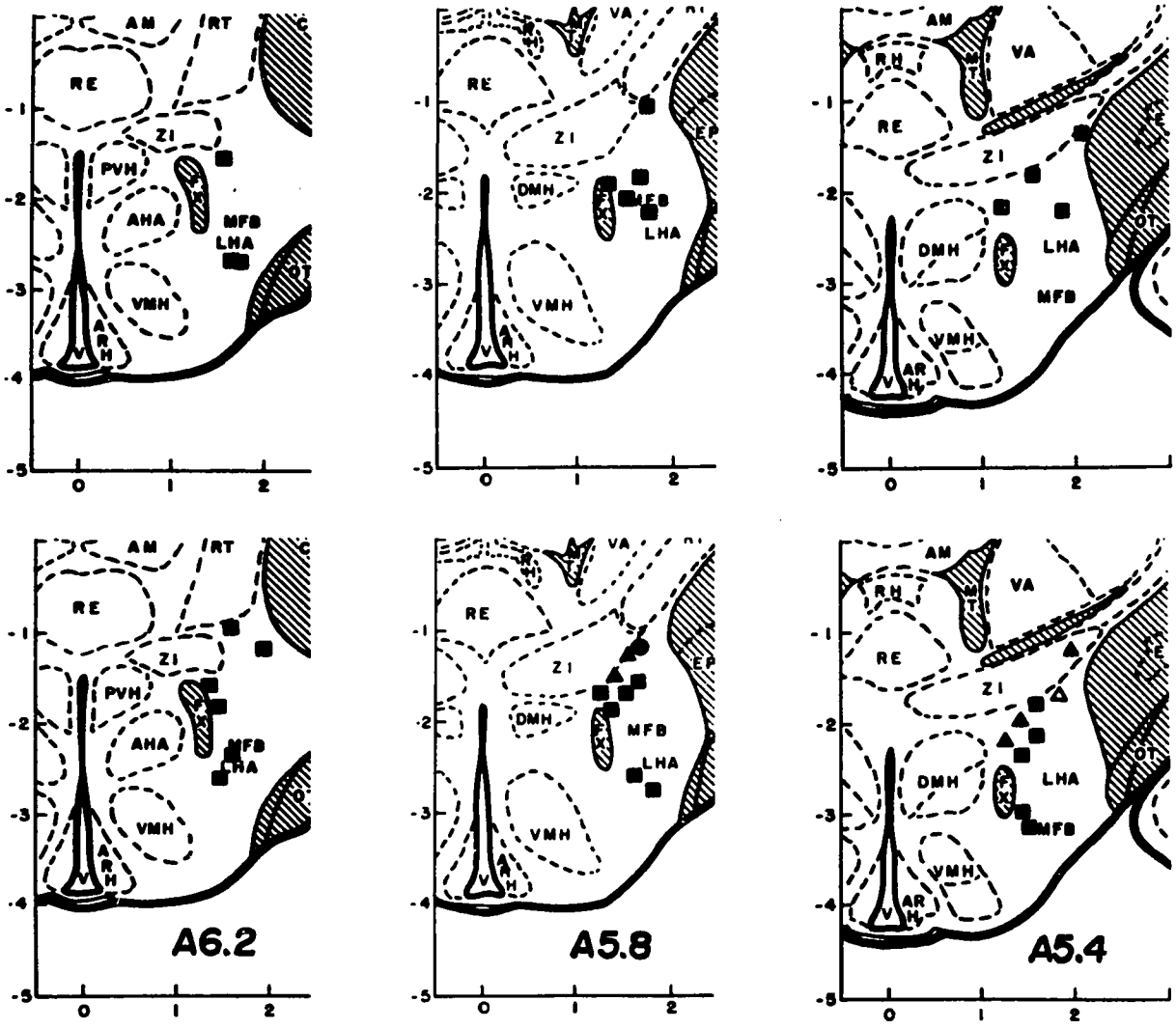


Figure 6 Reconstruction of electrode placements for Experiment 3.

- ■ Placements mediating eating, drinking and gnawing
- ● Placements mediating eating and gnawing
- ▲ Placements mediating eating and drinking
- △ Placements mediating drinking only
- □ Placements mediating negative effects.
- ▩ Negative placements

Brain sections from de Groot (1959) with anterior planes noted.

along the dorsal boundary of the eating and drinking systems as determined in Experiment I.

Discussion

The finding that only one out of twenty-four positive electrode placements failed to mediate more than one response upon initial testing indicates that no neural reorganization by stimulation experience is necessary before such results can be obtained. The study of Valenstein et al. (1968) demonstrated that none of the lateral hypothalamic sites which had been reported as mediating "pure" eating, drinking, or gnawing could be truly considered as "pure"; given their procedure (i.e., testing in a competitive situation after stimulation experience with individual consummatory objects), all or almost all of these sites would probably be capable of mediating at least two responses. The present study demonstrates that few of the lateral hypothalamic sites which appeared to be "pure" at the time of initial testing in the study of Valenstein et al. could be truly considered as "pure" either; given certain procedural changes (i.e., testing in non-competitive situations at a variety of stimulation intensities) all or almost all of these sites would probably be capable of mediating two or more responses, without the necessity of further stimulation experience. Thus it appears that the stimulation experience given the subjects of Valenstein et al. is not a necessary condition for obtaining two or more consummatory responses from one lateral hypothalamic site.

Nor does stimulation experience appear to be a sufficient condition for changing the drive specificity of lateral hypothalamic sites. The

histological consistency between the five placements which mediated eating and drinking but not gnawing suggests that there exists a dorsal portion of the perifornical area in which the gnawing system does not overlap with the eating and drinking systems. In Experiment I, a small ventral portion of the perifornical area was found which yielded drinking responses but not eating responses to stimulation. The eight sites which yielded this drinking, but failed to yield eating, did so despite months of stimulation-bound eating experience at other sites in the same animal. While this area may not be a "pure" drinking area in that it might have yielded gnawing if the appropriate test had been made, it seems unlikely that it could be "trained" to yield eating. These results seem to indicate that at least some portions of the eating, drinking, and gnawing systems are not only functionally, but also structurally independent, and that the response specificity of at least these portions of the systems cannot be modified by stimulation experience of the type given by Valenstein et al.

GENERAL DISCUSSION

The present experiments taken together support the following conclusions. First, while much of the lateral hypothalamic eating, drinking, and gnawing systems are anatomically overlapping, minor portions of some of these systems are structurally separated from at least some of the other systems. Second, the eating and drinking systems are continuous between their most dorsal and most ventral portions. Third, the fibre densities of the eating and drinking systems are homogeneous along any intersection of anterior-posterior and medial-lateral planes, within the

dorsal and ventral boundaries of the system in question. Fourth, there is no difference between the fibre density gradient of the eating system and that of the drinking system within the area of anatomical overlap. Fifth, the eating threshold at a given site is about the same as the drinking threshold at that site. Sixth, during the initial phases of electrical stimulation experience, thresholds for stimulation-bound responses can undergo marked changes. Seventh, comparison of electrode placements between animals provides a less satisfactory basis for understanding the functional anatomy of the lateral hypothalamic area than a comparison of several stimulation sites within a subject. Eighth, the neural organization necessary for the mediation of lateral hypothalamic stimulation-bound responses is established independent of stimulation experience.

The present investigation does not answer the question as to whether there are two independent fibre systems mediating eating and drinking, or whether the same fibres mediate both responses. The finding that eating and drinking thresholds are almost the same, and that even small changes in one are accompanied by similar changes in the other suggests that many of the same cells may be involved in both functions. It is readily apparent that some portions of the brain must contain separate fibres for eating and drinking, but it appears (Booth, 1967) that the chemical specificity of these systems disappears somewhere just anterior to the lateral hypothalamus. An hypothesis which would seem to be worth exploring is that the lateral hypothalamic area is an area of convergence of separate eating and drinking systems on a largely common path toward the motor system.

REFERENCES

Anand, B.K., and Brobeck, J.R. Hypothalamic control of food intake. Yale J. Biol. Med., 1951, 24, 123-140.

Andersson, B., and Wyrwicka, W. Elicitation of a drinking motor conditioned reaction by electrical stimulation of the hypothalamic "drinking area" in the goat. Acta physiol. scand., 1957, 41, 194-198.

Baillie, P., and Morrison, S.D. The nature of the supression of food intake by lateral hypothalamic lesions in rats. J. Physiol., 1963, 165, 227-245.

Booth, D.A. Localization of the adrenergic feeding system in the rat diencephalon. Science, 1967, 158, 515-517.

Booth, D.A., and Quartermain, D. Taste sensitivity of eating elicited by chemical stimulation of the rat hypothalamus. Psychonom. Sci., 1965, 3, 525-526.

Coons, E.E. Motivational correlates of eating elicited by electrical stimulation in the hypothalamic feeding area. Unpublished Ph.d. thesis, Yale University, 1964.

Coury, J.N. Neural correlates of food and water intake in the rat. Science, 1967, 156, 1763-1765.

de Groot, J. The rat forebrain in stereotaxic coordinates. Verh. Kon. Ned. Akad. Wet., B., 1959, 52, 1-40.

Epstein, A.N., and Teitlebaum, P. Severe and persistant deficits in thirst produced by lateral hypothalamic damage. In Thirst, M.J. Wayner, (Ed.), Pergamon; New York, 1964, pp 395-406.

Fisher, A.E. Maternal and sexual behavior induced by intracranial chemical stimulation. Science, 1956, 128, 228-229.

Fisher, A.E., and Coury, J.N. Cholinergic tracing of a central neural circuit underlying the thirst drive. Science, 1962, 138, 691-693.

Fisher, A.E., and Levitt, R.A. Drinking induced by carbachol: thirst circuit or ventricular modification? Science, 1967, 157, 839-841.

Gilbert, J.R. and Nuttall, B.J. Manual of techniques for neuropathology, Montreal Neurological Institute: Montreal, 1965.

Glickman, S.E., and Schiff, B.B. A biological theory of reinforcement. Psychol. Rev., 1967, 74, 81-109.

Gold, R.M. Aphagia and adipsia produced by unilateral hypothalamic lesions in rats. Amer. J. Physiol., 1966, 211, 1274-1276.

Gold, R.M. Aphagia and adipsia following unilateral and bilaterally asymmetrical lesions in rats. Physiol. Behav., 1967, 2, 211-220.

Grossman, S.P. Eating or drinking elicited by direct adrenergic or cholinergic stimulation of hypothalamus. Science, 1960, 132, 301.

Grossman, S.P. Direct adrenergic and cholinergic stimulation of hypothalamic mechanisms. Amer. J. Physiol., 1962a, 202, 872-882.

Grossman, S.P. Effects of adrenergic and chlinergic blocking agents on hypothalamic mechanisms. Amer. J. Physiol., 1962b, 202, 1230-1236.

Grossman, S.P. Some neurochemical properties of the central regulation of thirst. In Thirst, M.J. Wayner, (Ed.) Pergamon: New York, 1964, pp 487-511.

Levison, P.K., and Flynn, J.P. The object attacked by cats during stimulation of the hypothalamus. Animal Behaviour, 1965, 13, 217-220.

MacDonnell, M., and Flynn, J.P. Sensory control of hypothalamic attack. Animal Behaviour, 1966, 14, 399-405.

Mendelson, J. Role of hunger in T-maze learning for food by rats. J. comp. physiol. Psychol., 1966, 62, 341-349.

Miller, N.E. Experiments on motivation. Science, 1957, 126, 1271-1278.

Miller, N.E. Learning and performance motivated by direct stimulation of the brain. in Sheer, D.E. (Ed.) Electrical stimulation of the brain. University of Austin Press: Austin, 1961. pp 387-396.

Miller, N.E., Gottesman, K.S., and Emery, N. Dose response to carbachol and norepinephrine in rat hypothalamus. Amer. J. Physiol., 1964, 206, 1384-1388.

Mogenson, G.J., and Morgan, C.W. Effects of induced drinking on self stimulation of the lateral hypothalamus. Exp. Br. Res., 1967, 3, 111-116.

Montemurro, D.G., and Stevenson, J.A.F. The localization of hypothalamic structures in the rat influencing water consumption. Yale J. Biol. Med., 1955, 28, 396-403.

Montemurro, D.G., and Stevenson, J.A.F. Adypsia produced by hypothalamic lesions in the rat. Canad. J. Biochem. Physiol., 1957, 35, 31-37.

Morgane, P.J. Medial forebrain bundle and hypothalamic "feeding centers". Fed. Proc., 1960, 19, 292.

Morgane, P.J. Evidence of a "hunger motivational" system in the lateral hypothalamus of the rat. Nature, 1961a, 191, 672-674.

Morgane, P.J. Alterations in feeding and drinking behavior of rats with lesions in globi pallidi. Amer. J. Physiol., 1961b, 201, 420-428.

Morgane, P.J. Electrophysiological studies of feeding and satiety centers in the rat. Amer. J. Physiol. 1961c, 201, 838-844.

Morgane, P.J. Distinct feeding and hunger motivating systems in the lateral hypothalamus of the rat. Science, 1961d, 133, 887-888.

Morgane, P.J. Medial forebrain bundle and "feeding centers" of the hypothalamus. J. comp. Neurol., 1961e, 117, 1-26.

Morrison, S.D., and Mayer, J. Adipsia and aphagia in rats after lateral subthalamic lesions. Amer. J. Physiol., 1957, 191, 248-254.

Roberts, W.W., and Carey, R.J. Rewarding effect of performance of gnawing aroused by hypothalamic stimulation in the rat. J. comp. physiol. Psychol., 1965, 59, 317-324.

Roberts, W.W., and Keiss, H.O. Motivational properties of hypothalamic aggression in cats. J. comp. physiol. Psychol., 1964, 58, 187-193.

Roberts, W.W., Steinberg, M.L., and Means, L. W. Hypothalamic mechanisms for sexual, aggressive, and other motivational behaviors in the oppossum. Didelphis Virginiana. J. comp. physiol. Psychol., 1967, 64, 1-15.

Rogers, W.L., Epstein, A.N., and Teitlebaum, P. Lateral hypothalamic aphagia: motor failure or motivational deficit? Amer. J. Physiol., 1965, 208, 334-342.

Routtenberg, A. Drinking induced by carbachol: thirst circuit or ventricular modification? Science, 1967, 157, 838-839.

Smith, R.W., and McCann, S.M. Alterations in food and water intake after hypothalamic lesions in the rat. Amer. J. Physiol., 1962, 203, 366-370.

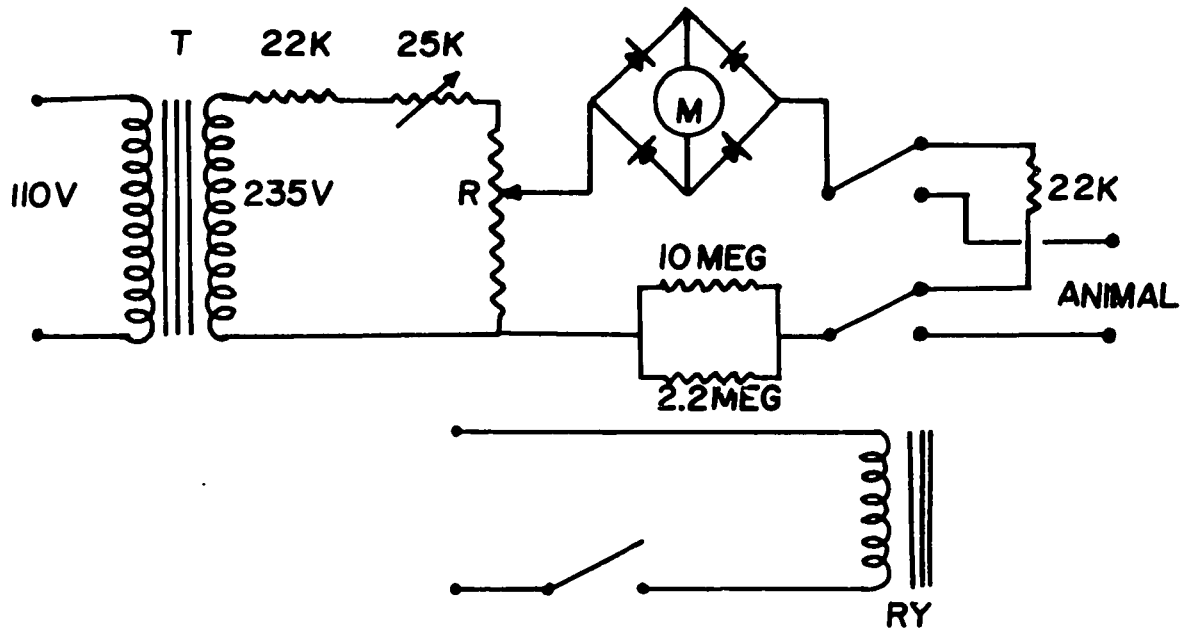
Teitelbaum, P., and Epstein, A.N. The lateral hypothalamic syndrome: recovery of feeding and drinking after lateral hypothalamic lesions. Psychol. Rev., 1962, 69, 74-90.

Teitlebaum, P., and Stellar, E. Recovery from the failure to eat produced by hypothalamic lesions. Science, 1954, 120, 894-895.

Tenen, S.S., and Miller, N.E. Strength of electrical stimulation of lateral hypothalamus, food deprivation and tolerance for quinine in food. J. comp. physiol. Psychol., 1964, 58, 55-62.

Valenstein, E.S., Cox, V.C., and Kakolewski, J.W.K. Modification of motivated behavior elicited by electrical stimulation of the hypothalamus. Science, 1968, 159, 1119-1121.

Williams, D.R., and Teitlebaum, P. Some observations on the starvation resulting from hypothalamic lesions. J. comp. physiol. Psychol. 1959, 52, 458-465.



Appendix A Circuit diagram for brain stimulator

T	Hammond 270Z
R	Borg 2201B 100K, 10 turn
RY	KRP 11A 117V AC
M	Simpson 2327 No. 27 0-100 μA