Behavioral Neuroscience
1999, Vol. 113, No. 5, 1049–1061

0735-7044/99/$3.00

Infusion of Neurotoxic Doses of *N*-Methyl-D-Aspartate Into the Lateral Hypothalamus in Rats Produces Stomach Erosions, Hyperthermia, and a Disruption in Eating Behavior

Jesus Landeira-Fernandez
Pontificia Universidade Católica
and Universidade Estácio de Sá

Carlos V. Grijalva
University of California, Los Angeles

The present study examined whether damage to intrinsic lateral hypothalamic (LH) neurons induced by microinfusions of *N*-methyl-D-aspartate (NMDA) would produce effects similar to those seen after electrolytic LH lesions. In Experiment 1, rats receiving electrolytic (1.2 mA anodal current, 10 s) LH lesions displayed motor impairments, whereas those receiving NMDA (20 μg/μl) infusions did not. Both electrolytic lesions and NMDA infusions were associated with eating deficits, hyperthermia, and gastric erosion formation 24 hr after surgery. In Experiment 2, either 20 μg/μl or 10 μg/μl NMDA destroyed LH cells and produced dose-dependent gastric mucosal erosions as well as similar increases in body temperature. These results indicate that an alteration in the acute activity of intrinsic LH neurons plays a role in the production of gastric mucosal injury and hyperthermia and lend support to other studies implicating a role of LH neurons in eating behavior.

The lateral hypothalamus (LH) comprises a large and diffuse area in the diencephalon that has been shown to be involved in ingestion, digestion, autonomic regulation, and general metabolic responses, as well as in sensory and motor functions (see Bernardis & Bellinger, 1993; Grijalva, Lindholm, & Roland, 1989; Grijalva & Novin, 1990). For example, experimental lesions of the LH are associated with drinking and feeding deficits and body weight reduction (Anand & Brobeck, 1951; Teitelbaum & Epstein, 1962), homeostatic disturbances involving olfaction (Scott & Leonard, 1971), salivation (Epstein, 1971; Schallert, Leach, & Braun, 1978), gastrointestinal properties (Grijalva, Lindholm, Schallert, & Bicknell, 1976; Lindholm, Shumway, Grijalva, Schallert, & Ruppel, 1975) and temperature regulation (Corbett, Wilterdink, & Keesey, 1985; DeRyck & Teitelbaum, 1978; Harrell, de Castro, & Balagura, 1975; Grijalva, 1980; Grijalva & Lindholm, 1980; Lennie, Hirvonen, McCarthy, & Keesey, 1995; Monda, Amaro, Sullo, & De Luca, 1996; Refinetti & Carlisle, 1987), as well as motor, sensorimotor, and activational deficits (Baillie & Morrison, 1963; Levitt & Teitelbaum, 1975; Marshall & Teitelbaum, 1974; Balagura, Wilcox, & Coscina, 1969).

One of the main goals of the present study was to further examine the role of the LH in the production of gastric mucosal erosions. Previous work has shown that bilateral electrolytic lesions of the LH area alter gastric acid secretion (Taché, Grijalva, Gunion, Walsh, & Novin, 1982), gastric mucosal barrier properties (Grijalva, Deregnaucourt, Code, & Novin, 1980; Namiki, Egawa, Inoue, Tominaga, & Takamura, 1994), and gastric contractility (Garrick, Grijalva, & Trauner, 1993) and induce gastric mucosal erosions (Grijalva & Lindholm, 1980; Grijalva, et al., 1976; Lindholm et al., 1975; Roland & Grijalva, 1993; Schallert, Whishaw, & Flannigan, 1977; Tordoff et al., 1984). Although these findings reinforce the view that the LH area is importantly involved in gastrointestinal functions (Grijalva, Lindholm & Novin, 1980; Grijalva & Novin, 1990), it remains to be determined whether changes in stomach physiology and, ultimately, the development of stomach erosions following LH damage are mediated by alterations in neuronal function or by the interruption of axonal fibers of passage that course through the LH area. The studies cited above destroyed both intrinsic LH neurons and fibers of passage with electrolytic current.

Among a large variety of chemical messengers, the excitatory amino acid glutamate appears to be a primary neurotransmitter in the LH area (Stanley, Butterfield, & Grewal, 1997; Stanley, Ha, Spears, & Dee, 1993; Stanley, Willett, Donias, Dee, & Duva, 1996; Stanley, Willett, Donias, Ha, & Spears, 1993). The receptor complex of excitatory amino acids has been classified into two major categories: *N*-methyl-D-aspartate (NMDA) and non-NMDA receptors. The NMDA receptor includes a number of sub-

Jesus Landeira-Fernandez, Departamento de Psicologia, Pontifica Universidade Católica, Rio de Janeiro, Brazil, and Curso de Psicologia, Universidade Estácio de Sá, Rio de Janeiro, Brazil; Carlos V. Grijalva, Department of Psychology, University of California, Los Angeles (UCLA).

This research was supported by a UCLA Academic Senate Faculty Research Grant and a grant from the Brazilian Secretary for Science and Technology. Portions of these data were presented at the 23rd Annual Meeting of the Society for Neuroscience, Washington, DC, November 1993. We thank Javier Rios-Jimenez for his laboratory assistance.

Correspondence concerning this article should be addressed to Carlos V. Grijalva, Department of Psychology, Franz Hall, University of California, Los Angeles, California 90095-1563. Electronic mail may be sent to grijalva@psych.ucla.edu.

types, including the NMDA, kainate, quisqualate or α-amino-3-hydroxy-5-methylisoxazole-4-propionate (AMPA), L-2-amino-4-phosphonobutyrate (AP4), and trans-1-aminocyclopentane-1,3-dicarboxylic (ACPD) receptor subtypes (Monaghan, Bridges, & Cotman, 1989). At least seven non-NMDA receptor subtypes appear to exist (Dingledine, 1991). Prolonged stimulation of excitatory amino acid receptors of either the NMDA or non-NMDA types eventually results in cell death (Rothman & Olney, 1987). Ibotenic acid (IBO) and NMDA are NMDA agonists, and kainic acid (KA) is a kainate agonist. These three agonists have been used as neurotoxins to produce neuronal death with relative sparing of axonal fibers of passage. It is believed that two independent and additive mechanisms mediate the neuronal death produced by these neurotoxins. First, excitotoxic death is produced by an increase in sodium permeability, which results in the depolarization of the neuron, causing a continuous influx of chloride and other anions, which eventually results in a osmotic lysis. Second, neuronal death appears to be related to calcium entry through the agonist-gated and voltage-gated channel. Elevation of intracellular calcium activates lipase and protease and irreversibly damages mitochondria. These neurotoxic processes generally occur in less than 24 hr (Farber, Chien, & Mittnacht, 1981; Winn, 1991).

Although KA, IBO, and NMDA share similar cellular mechanisms, it appears that these chemicals have different action profiles when used as neurotoxic agents. For example, lesions produced by KA often result in neuronal damage distal to the site of infusion (Kohler & Schwarcz, 1983; Peterson & Moore, 1980). Damage distal to the site of infusion does not seem to be present when IBO or NMDA is used (Hastings, Winn, & Dunnett, 1985; Markowska, Bakke, Walther, & Ursin, 1985). However, in comparing IBO with NMDA, it appears that NMDA might be a better neurotoxin for selective destruction of LH neurons. It has been reported that IBO infusions into the LH produce an increase in dopamine (DA) levels in the striatum (Winn, Tarbuck, & Dunnett, 1984), whereas similarly placed NMDA infusions do not alter DA concentration (Winn et al., 1990). Moreover, there is evidence showing that IBO may induce axon demyelination in the region of local infusion (Coffey, Perry, Allen, Sinden, & Rawlins, 1988; Stellar, Hall, & Waraczynski, 1991). Damage of the myelin sheath alters the ability of the axon to carry the neural impulse, possibly by changing its conduction properties. On the basis of these results, we decided to use NMDA as a preferred neurotoxin in producing selective lesions within the LH.

Winn and colleagues (A. J. M. Clark, Clark, & Winn, 1990; J.M. Clark, Clark, Bartle, & Winn, 1991) showed that bilateral infusion of NMDA into the LH produces an array of feeding deficits without causing any motor impairment. Additionally, Winn et al. (1990) did not observe gastric mucosal damage in NMDA-treated rats. However, rats in this study were keep alive for 10–13 weeks after surgery. Gastric erosions produced by electrolytic LH lesions occur within 3–24 hr after LH destruction (Grijalva, Deregnaucourt, et al., 1980; Grijalva & Lindholm, 1980), and the severity of the erosions increases over 4 days postoperatively (Lindholm et al., 1975). After this period, gastric erosions tend to heal and eventually disappear, as long as the rats are provided with intragastrically infused nutrient or if feeding resumes (Grijalva, 1993). From these findings, it would appear that the effects of either electrolytic or excitatory neurotoxin–induced LH lesions on the occurrence of gastric erosions are mediated by an acute process related to these lesioning methods, including early sustained excitation of the neurons, rather than to the actual loss of the cells (Roland & Grijalva, 1993).

This study compared the effects of bilateral NMDA infusions or electrolytic lesions in the LH on the occurrence of gastric erosions. Pilot studies from our laboratory indicated that a 20 μg/μl dose of NMDA and a 10-s, 1.2 mA anodal electrolytic current produces a similar area of damage within the LH. In addition, because both NMDA–LH infusions and electrolytic lesions lead to feeding deficits, but only electrolytic lesions produce motor impairments, eating and motor behaviors were also examined to confirm these preliminary findings. Finally, electrolytic LH lesions produce an increase in body temperature; however, it is not known whether NMDA infusions in the LH produce similar temperature changes. Therefore, the present study compared core body temperature in rats receiving either NMDA infusions or electrolytic lesions in the LH.

General Method

Animals

Experimentally naive male albino Sprague-Dawley (Bantin Kingman, Fremont, CA) rats weighing 220–320 g were used. They were individually housed in a colony room with a 12:12-hr light–dark cycle and had ad-lib access to food pellets (Purina Laboratory Chow) and tap water. The rats were handled daily for at least 1 week prior to the onset of the experiments.

Experiment 1 included three groups of rats matched for initial body weight. The groups were as follows: electrolytic LH lesion (ELEC, $n = 9$), NMDA–LH infusion (NMDA, $n = 7$), and control (CONT, $n = 7$). Three rats in the ELEC group and 2 rats in the NMDA group were excluded from the experiment because of death during surgery or lesion misplacement. The final group sizes were $n = 6$ for the ELEC group, $n = 7$ for the NMDA group, and $n = 7$ for the CONT group.

In Experiment 2, 25 naive male rats were divided into four groups matched for initial body weight. Two of the four groups received either 20 ($n = 9$) or 10 μg/μl ($n = 9$) bilateral injections of NMDA in the LH. A third group (THAL, $n = 4$) was bilaterally injected with 20 μg/μl of NMDA into the thalamus, 1.5 mm above the LH area. A CONT group ($n = 3$) received bilateral vehicle injection in the LH.

Surgery and Pre- and Postoperative Measurements

All of the rats were food, but not water, deprived overnight (14–16 hr) prior to surgery. Rats receiving stereotaxic surgery were anesthetized with sodium pentobarbital (Nembutal, 65 mg/kg, ip) and then mounted in a stereotaxic instrument (Model 900, Kopf Instruments, Tujunga, CA). After surgery, the rats were returned to their home cages and were food deprived for an additional 24 hr.

Following the postoperative deprivation period, two food pellets were placed on the floor of the home cage and the rats were tested for food intake behavior for 2 hr. The rats also were given a cage-climbing test to access motor ability on three separate occasions: 1 day prior to surgery, the day of surgery just prior to anesthetization, and 24 hr after surgery. The cage-climbing test (Grijalva & Lindholm, 1980) consisted of placing the rat on a inverted cage with its head and forepaws over the horizontal surface and its hindlimbs over the vertical surface. The presence of motor impairment was defined as the inability to climb to the horizontal position during a 30-s period. Core body temperature was measured with a rectal probe attached to a thermometer (Yellow Springs Instruments, Yellow Springs, OH) immediately prior to anesthetization for stereotaxic surgery and 24 hr after surgery. Following the 24-hr temperature measure the rats were sacrificed as described below.

Determination of gastric mucosal damage. At the conclusion of each experiment (24 hr postoperatively), the rats were given a lethal injection of sodium pentobarbital (0.5 ml, ip) and were decapitated. A ligature was placed around the duodenum, a cannula was inserted into the stomach via the esophagus, and 3 cc of 10% formalin was infused into the stomach. Following the formalin infusion, the esophagus was ligated. Approximately 10 min later, the stomach was opened along the great curvature, rinsed with water, spread on a flat surface, and fixed with 10% formalin. The stomachs were stored in formalin for several weeks, and after this time the gastric mucosa was examined with a binocular dissection microscope at 8×. One eyepiece was fitted with a reticle, permitting gastric lesions to be quantified in terms of total area (mm^2). Any discontinuity in the gastric mucosa was considered a gastric erosion. All stomachs were examined by an independent rater who was unaware of the experimental conditions.

Lesions of the LH. Bilateral electrolytic LH lesions were made by passing a 1.2 mA anodal current for 10 s through a stainless steel insect pin (size 00) insulated with baked epoxylite except for the cut tip. A cathode clamped to the tail completed the circuit. The current was delivered by a lesion-generating device (Stoelting, Chicago, IL). With reference to Paxinos and Watson's (1986) rat brain atlas, stereotaxic coordinates were 2.4 mm posterior to bregma, 1.9 mm lateral to each side of midline, and 7.8 mm ventral to dura of the brain, with the head mounted in the horizontal plane. These same stereotaxic coordinates were used for bilateral infusions of 1 μl of NMDA into the LH (Sigma Chemicals, St. Louis, MO). The NMDA was dissolved in 0.1 M of phosphate buffer solution (PBS; adjusted to 7.4 pH) and administered in doses of 20 μg/μl in Experiment 1 and 10 or 20 μg/μl in Experiment 2. The drug was made fresh on the day of the experiment. Drug infusion was made by a glass micropipette connected to a 10-μl Hamilton microsyringe using a polyethylene tubing. The Hamilton syringe was mounted on a injection pump set up to deliver 1 μl of the drug over 15 min. Following the infusion, the micropipette was kept in place for an additional 5 min to prevent the drug from diffusing up the micropipette track. Before and after each injection, the flow at the tip of the micropipette was verified by turning the pump on until a droplet appeared. Control subjects were infused with an equal volume of PBS vehicle alone.

Histology

The brains of all rats were removed and stored in 10% formalin for about a week. The brains were sectioned using the cryostatic method at 50–60 μm, and every third section was mounted on gelatin-subbed slides and stained with thionin. The extent of affected brain area (lesion cavity and/or apparent region of chromatolysis or visible cell loss) was evaluated with reference to the Paxinos and Watson (1986) rat brain atlas. All histological examinations of brain sections were done without knowledge of group status. To permit comparison of the affected brain region among groups, corrected for shrinkage due to histological preparation, we superimposed mounted and stained sections with a microprojector onto corresponding structures representing tracings from coronal atlas plates. Magnification was adjusted until projected structures adjacent to the lesion corresponded with the atlas structures. Outlines of the region displaying apparent cell loss or chromatolysis observed in the representative sections through the affected brain region were drawn to scale onto tracings of the brain sections taken at appropriate anterior–posterior levels. The anterior–posterior extent of the brain region affected was estimated by calculating the divisions of the atlas figures that were incorporated within the lesion outline. The dorsal–ventral and medial–lateral extent of the lesion was measured with the atlas scale. An estimation of the volume of brain tissue affected was obtained by multiplying the anterior–posterior, dorsal–ventral, and medial–lateral extents. Data obtained from rats with misplaced lesions (i.e., lesions falling outside the LH proper or asymmetrical lesions in which only one side of the LH was affected) were excluded from statistical analysis.

Cell Count and Cell Type

To derive at an ordinal index of the amount of cell death produced by NMDA infusions, we counted cells in the left and right infusion sites in the region that exhibited the least cell density (i.e., midpoint of the infusion site) or equivalent area in the control group. One eyepiece was fitted with a grid reticle (8 × 8 subdivisions), and histological brain sections meeting the criteria stated above were examined at a magnification of 100×. The grid covered an area of 1.0 × 1.0 mm, and all stained cells defined as a blue dot within the field of view were counted. The counting procedure did not discriminate between remaining neurons or glial cells and, therefore, was used as a conservative estimate of the magnitude of total cell loss following a given infusion procedure. The procedure for counting cells was adapted from one previously described by Turski, Bressler, Rettig, Loschmann, and Wachtel (1991). Although it was assumed that neurons would be those primarily affected by NMDA infusions, it has been shown that some types of glial cells contain NMDA receptors (Müller, Grosche, Ohlemeyer, & Kettenmann, 1993; López-Colomé, Ortega, & Romo-de-Vivar, 1993). Thus, toxic doses of NMDA have the potential of killing a proportion of both neurons and glial cells.

Statistical Analysis

We used an analysis of variance (ANOVA) to compare group differences. We used Duncan's multiple range test or the Newman–Keuls test to determine significant differences between specific groups. The results for gastric mucosal damage are expressed as means (±*SEM*). Because group means and variances tended to covary, and the distributions were also positively skewed, we conducted a log $(X + 1)$ transformation on the gastric erosion scores for each rat for the purpose of statistical analysis.

Results

Experiment 1

Histology. Visual inspection of the LH damage in the remaining rats killed 24 hr following surgery indicated that NMDA and electrolytic lesions were bilaterally symmetrical and destroyed most of the medial and posterior portions of the LH. Although the placement of both types of lesions was similar, different neural tissue damage was obvious. Electrolytic lesions included a cavity in the center of the lesion plus a region of chromatolysis surrounding the cavity. On the other hand, the NMDA lesion did not produce any cavities and the region of the infusion site tended to vary more than that produced by the electrolytic lesion. However, mean estimates of the total volume of brain tissue involved were not significantly different between the ELEC group and the NMDA group. Mean volume was 5.55 ± 0.25 mm^3 for the ELEC group, which included the lesion cavity and region of chromatolysis; 5.96 ± 0.45 mm^3 for the NMDA group; and 0.35 ± 0.13 mm^3 for the CONT group, which primarily involved the tip of the cannula tract.

A representative histological section of the rostrocaudal midpoint of the electrolytic LH lesion is presented in Figure 1. The lesions were bilaterally symmetrical and involved a majority of the LH area. The damage was generally confined to the region bordered by the rostrocaudal extent of the ventromedial hypothalamic nucleus and frequently encroached on the ventromedial tip of the internal capsule, the fornix, the fields of Forel, and the zona incerta. Chromatolysis generally extended 0.3–0.5 mm around the LH and occasionally extended to the border of the ventromedial thalamus and the ventromedial hypothalamus. Figure 2 shows a representative histological section from a subject bearing NMDA infusions. The involvement of the LH and adjacent structures following NMDA infusion was similar to that produced by electrolytic lesions. Two of the 7 rats in the NMDA group displayed apparent cell loss extending to portions of the ventromedial nucleus of the thalamus. A representative photomicrograph of a CONT subject infused with PBS is shown in Figure 3. No apparent neural damage was found in any of the CONT subjects, with the exception of the presence of the cannula tract.

To obtain the ordinal index of the extent of cellular involvement following NMDA infusions, we counted cells in each side of the brain in the region of maximal expression of apparent cell loss and an equivalent area in the brains of the CONT-group rats. High-power (100×) magnification of the LH area from a rat in the NMDA group and 1 in the CONT group is shown in Figures 4 and 5, respectively. As shown in Figure 6, there was no difference in cell count between the left and right LH area in either of the groups. However, a reliable difference between the NMDA group and the CONT group in both sides of the LH area was found:

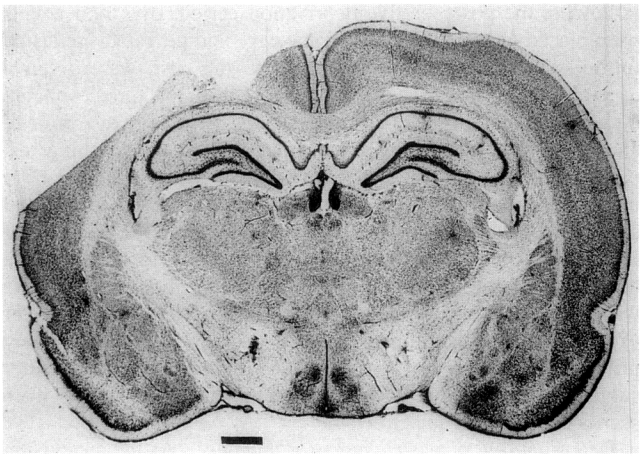

Figure 2. Histological section taken through the midpoint of a representative *N*-methyl-D-aspartate infusion (20 μg/μl) in the lateral hypothalamic area (thionin stain). Scale bar = 1 mm.

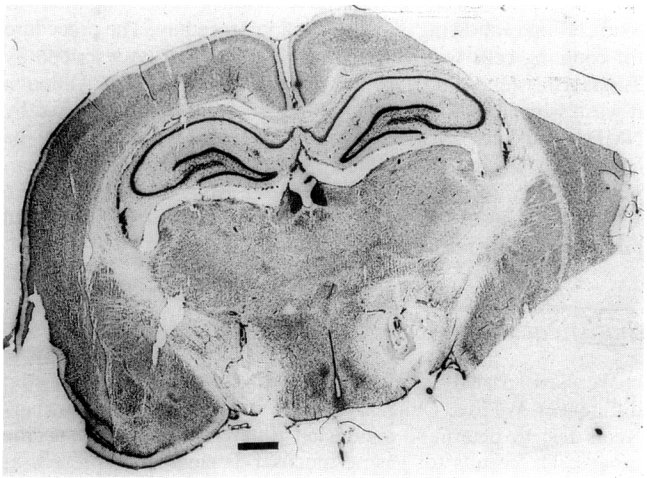

Figure 1. Histological section taken through the midpoint of a representative electrolytic lateral hypothalamic lesion (thionin stain). Scale bar = 1 mm.

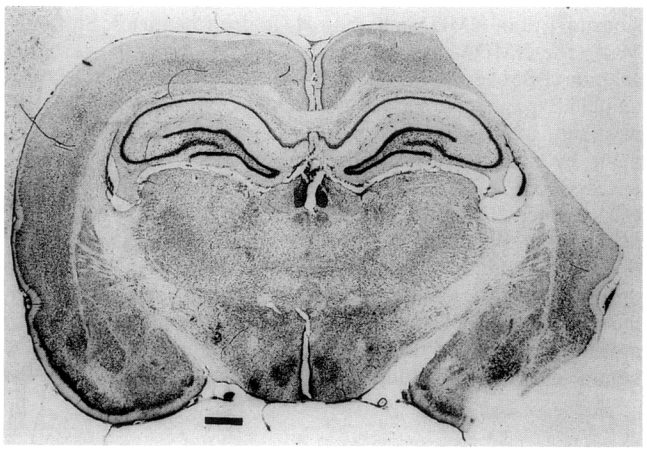

Figure 3. Histological section through the midpoint of the lateral hypothalamic area in a control-operated rat (thionin stain). Scale bar = 1 mm.

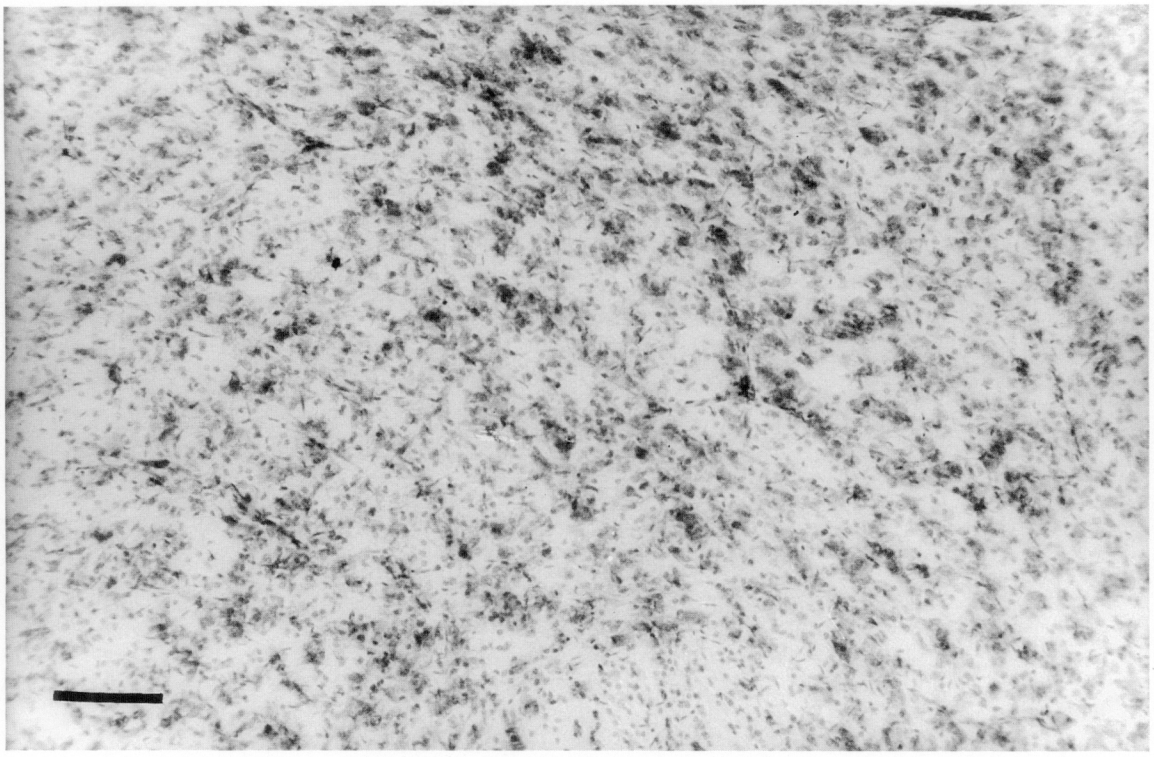

Figure 4. High-power photomicrograph from same tissue shown at low power in Figure 2. Microphotography was taken at the central region of the *N*-methyl-D-aspartate infusion (approximately 1 × 1-mm quadrant). Scale bar = 100 μm.

left side, $F(1, 11) = 21.45, p = .000$; right side, $F(1, 11) = 21.64, p = .0007$. Relative to CONT subjects, NMDA-infused rats experienced about 37% (left side = 37.5%, right side = 37.3%) total cell loss in the LH. Thus, approximately 63% of LH cells remained after NMDA infusions; however, a majority of these cells fit the criteria for glial cell classification (Bloom & Fawcett, 1975). For example, the remaining cells were typically small, round, and darkly stained, with no visibly distinct nucleus, cytoplasm, or dendritic processes at 100× magnification. This is in contrast to the appearance of cells possessing these characteristics in CONT subjects. Nevertheless, the present analysis did not permit the quantification or determination of the type of neuron affected by the NMDA infusion. Because the LH has a large number of cells with NMDA receptors (Monaghan et al., 1989), a large portion of these cells were undoubtedly affected.

Temperature. Core body temperature measures taken just before surgery and 24 hr after surgery are presented in Figure 7. Because there was no difference in temperature between groups before surgery, we analyzed the data between groups pre- and postoperatively. A paired *t*-test revealed a significant increase in temperature in both the ELEC group, $t(5) = 35.29, p = .000$, and the NMDA group, $t(6) = 3.17, p = .019$, but not in the CONT group.

Behavioral analysis. All of the rats in the ELEC group consistently displayed stereotypical motor activity for about 2–4 hr after surgery characterized by hyperactivity, persistent scratching at the corner of the cage, and obstinate progression (Grijalva & Lindholm, 1980; Levitt & Teitelbaum, 1975). This enhanced motor activity eventually disappeared, giving way to a clear-cut hypoactivity that lasted until the rats were sacrificed. NMDA-infused rats exhibited seizures approximately 1 hr after surgery that lasted for about 3 hr. After recovery from seizures, these subjects exhibited normal behavior and were indistinguishable from vehicle-treated subjects. The occurrence of seizures after NMDA injection in the LH has been previously reported (A.J.M. Clark et al., 1990).

Motor impairment, measured by the cage-climbing test, was detected only in the ELEC group. Four of the 6 ELEC rats (66%) did not climb the cage within 30 s, whereas all of the other rats successfully passed the cage-climbing test. Indeed, electrolytic lesions reliably produced motor deficit as compared with CONT rats ($z = 2.59, p < .05$).

None of the rats in the ELEC and NMDA groups ate during the 2-hr feeding test, whereas all of the rats in the CONT group clearly engaged in feeding behavior. The occurrence of eating was also confirmed by the presence of food in the stomach postmortem. Although both NMDA and ELEC rats did not show eating responses, the rats in these two groups displayed different reactions toward the food pellets. ELEC rats completely ignored the presence of the food pellets in their home cage, whereas NMDA-infused

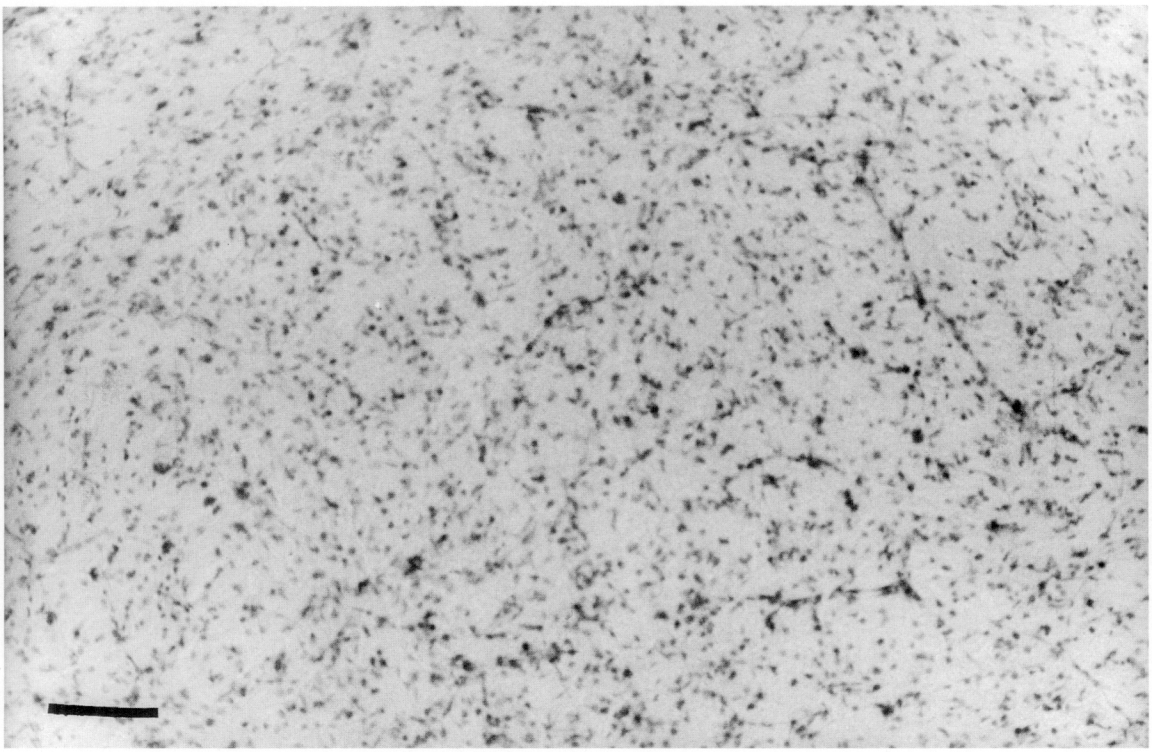

Figure 5. High-power photomicrograph from same tissue shown at low power in Figure 3. Microphotography was taken at the central region of the control phosphate buffered saline infusion (approximately 1 × 1-mm quadrant). Scale bar = 100 μm.

rats tended to handle the food pellets but made no attempt to ingest the pellets.

Gastric erosions. The presence of gastric mucosal injury was observed in the antrum and glandular portions of the stomach and was scored as total area in both of these portions. Figure 8 shows the mean gastric mucosal damage in the three groups. The extent of gastric erosion was significantly different among groups, $F(2, 18) = 4.51$, $p = .0267$. Post hoc tests revealed that the ELEC and NMDA groups did not differ from each other but that both groups had more gastric erosions than did the CONT group (ELEC vs. CONT, $p < .001$; NMDA vs. CONT, $p < .05$).

Experiment 2

The results of Experiment 1 indicate that either damage or acute hyperactivation of intrinsic LH neurons with a relatively high dose of NMDA leads to the development of gastric erosion formation, hyperthermia, and a disruption of eating behavior. However, because the dose of NMDA that was used tended to spread to adjacent structures, especially into the areas dorsal to the LH, it is unclear whether or not the effects observed after NMDA infusion were directly due to interruption of intrinsic LH neurons or due to interruption of neurons in adjacent structures. To restrict the infusion site to the LH proper, we reduced the NMDA dose by half. As a separate control, some rats received NMDA (20 μg/μl) in the thalamic area dorsal to the LH. A previous study showed that electrolytic lesions of the ventral thalamus do not induce gastric mucosal damage (Lindholm et al., 1975).

Histology. Seven rats were excluded from the experiment due to death during surgery or lesion misplacement. The final number of rats per group were as follows: $n = 6$ for the 20 μg/μl NMDA group, $n = 7$ for the 10 μg/μl NMDA group, $n = 2$ for the THAL group, and $n = 3$ for the CONT group. The 2 rats in the THAL group had lesions restricted to the thalamic area, which included parts of the ventromedial, ventrolateral, ventral posterolateral, and ventral posteromedial nuclei of the thalamus. Neither the THAL nor the CONT group exhibited neuronal damage in the LH area. Brain lesions of the LH group microinjected with 20 μg/μl of NMDA were similar to the NMDA group in Experiment 1. The area of apparent cell loss was relatively large and consistently extended to the dorsal region of the LH. The area of involvement in the group microinjected with 10 μg/μl of NMDA was less than in the 20 μg/μl NMDA group and tended to be more restricted to the LH area. The mean volume of the affected brain tissue was 6.37 ± 0.51 mm^3 for the 20 μg/μl NMDA group, 2.57 ± 0.52 mm^3 for the 10 μg/μl NMDA group, and 0.20 ± 0.05 mm^3 for the CONT group, which, as in the Experiment 1, primarily involved the tip of the cannula tract. These differences were significant, $F(2,$

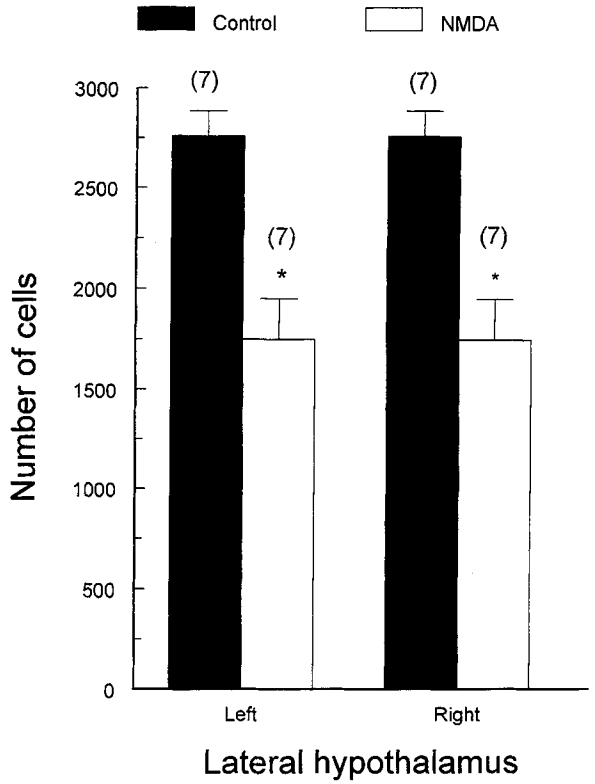

Figure 6. Mean (±*SEM*) cell count in the left and right lateral hypothalamus under high-power magnification 24 hr after *N*-methyl-D-aspartate (NMDA; 20 μg/μl) or control vehicle (phosphate buffered saline) infusion. Numbers in parentheses indicate the number of subjects. *$p < .005$ (compared with control).

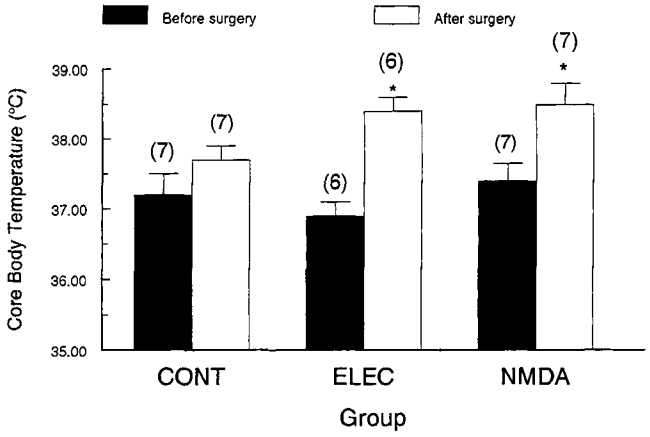

Figure 7. Mean (±*SEM*) core body temperature before (solid bars) and after (open bars) electrolytic (ELEC) lesions and *N*-methyl-D-aspartate (NMDA; 20 μg/μl) infusions in the lateral hypothalamus. Numbers in parentheses indicate the number of subjects. CONT = control. *$p < .001$ (compared with the before-treatment condition).

17) = 41.62, $p < .00001$, both the 20 and 10 μg/μl NMDA groups had a greater lesion volume than did the CONT group ($ps < .01$), and the 20 μg/μl NMDA group had a greater lesion volume than did the 10 μg/μl NMDA group ($p < .01$). The affected brain area in the rats in the 10 μg/μl NMDA group involved the majority of the LH and frequently encroached on the ventral tip of the internal capsule, the fields of Forel, the ventromedial portion of the zone incerta, and a small area of the ventromedial nucleus of the thalamus.

Brain cells were counted in the region of maximal expression of the infusion site or in an equivalent area in CONT subjects. No difference in cell count in the LH area was found between rats in the THAL group and the CONT group. Thus, the data from these two groups were pooled. The mean cell counts in the LH is presented in Figure 9. As indicated by the figure, loss of the LH cells following NMDA infusion was dose related. The 10 μg/μl dose of NMDA destroyed about 40% of intrinsic LH cells, whereas the 20 μg/μl dose destroyed about 60% of LH cells. As in Experiment 1, cells remaining following NMDA infusions appeared to be predominantly glial cells. An ANOVA indicated an overall difference among the groups: LH right side, $F(2, 15) = 51.00$, $p < .0001$; LH left side, $F(2, 15) = 47.60$, $p < .001$. Post hoc tests revealed a significant difference between the two NMDA groups compared with the CONT group in both sides of the LH ($ps < .005$). Moreover, a significant difference between the 20 and 10 μg/μl NMDA groups in both sides of the LH area was also found ($ps < .005$), indicating that the extent of cell loss was related to the NMDA dose.

Temperature. Core body temperatures, pre- and postoperatively, are shown in Figure 10. There were no group differences in body temperature before surgery, but the 20 and 10 μg/μl NMDA groups displayed statistically reliable increases in body temperature after surgery ($ps < .01$). No difference in temperature was found between the two NMDA groups. These results replicate the finding in Experiment 1 and additionally show that both doses of NMDA produce similar increases in body temperature.

Behavioral analysis. All of the subjects that were given NMDA injections (20 μg/μl NMDA–LH, 10 μg/μl NMDA–LH, and 20 μg/μl NMDA–THAL) exhibited seizure activity to varying degrees. Nonsystematic observations gave the impression that subjects in the 20 μg/μl NMDA–LH group had more severe seizures than did subjects in the 10 μg/μl NMDA–LH and 20 μg/μl NMDA–THAL groups. Three to 5 hr after surgery, seizures tended to disappear and subjects regained normal behavior. No signs of motor impairment were found 24 hr after surgery, and all of the subjects in each of the groups passed the cage-climbing test.

Consistent with the results in Experiment 1, all of the rats microinjected with 20 μg/μl of NMDA into the LH refused to eat 24 hr after surgery. Four of the 7 rats in the 10 μg/μl NMDA–LH group also did not eat. The remaining 3 rats in this group displayed some interest in eating by nibbling

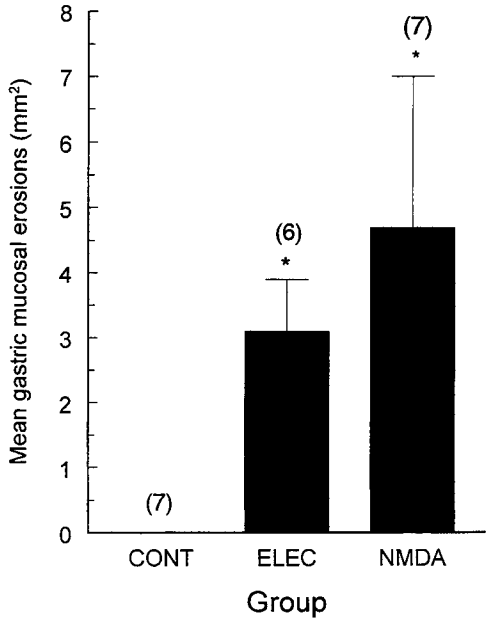

Figure 8. Mean (±*SEM*) total area of gastric mucosal erosions in rats with bilateral electrolytic (ELEC) lesions, *N*-methyl-D-aspartate (NMDA; 20 μg/μl) and control (CONT) vehicle infusions. Numbers in parentheses indicate the number of subjects. *$p < .005$ (compared with CONT group).

slightly on the food pellets, but their feeding behavior was clearly different from the robust eating of rats in the CONT group. The 2 rats in the THAL group, which received 20 μg/μl NMDA infusions restricted to the thalamic nuclei, displayed eating responses similar to those of the rats in the CONT group. However, 2 other rats in the THAL group, which were excluded from the study because of large lesions involving other structures, including part of the LH, did not show any ingestive behavior.

Gastric erosions. Gastric erosions occurred in the antrum and glandular portions of the stomach and were scored and analyzed as in the Experiment 1. The 2 rats in the THAL group exhibited no apparent gastric mucosal damage, and their scores were pooled with the rats in the CONT group. The mean total area of gastric mucosal damage for the groups treated with PBS or NMDA (20 or 10 μg/μl) are presented in Figure 11. An ANOVA revealed a significant difference among groups, $F(2, 17) = 13.40, p = .0004$). The 20 and 10 μg/μl NMDA groups differed significantly from each other ($p < .05$), and both of these groups differed significantly from the CONT group (20 μg/μl NMDA vs. CONT, $p < .001$; 10 μg/μl NMDA vs. CONT, $p < .05$). These results replicate the findings of the first experiment, in which 20 μg/μl NMDA injected into the LH led to the development of gastric erosions 24 hr after surgery. In addition, smaller NMDA infusions more restricted to the LH area also consistently induced gastric erosions, but to a lesser degree.

Discussion

Bilateral electrolytic lesions of the LH area induce a host of behavioral, physiological, and metabolic disorders (Bernardis & Bellinger, 1993; Epstein, 1971; Grijalva et al., 1989; Grijalva & Novin, 1990; Powley, Opsahl, & Weingarten, 1980; Teitelbaum & Epstein, 1962). However, electrolytic lesions destroy not only cell bodies but also disrupt axonal fibers of passage that course through the LH region. This is particularly problematic because LH neurons are diffusely scattered among a larger number of fibers that compose the medial forebrain bundle. Therefore, it is unclear to what extent the changes seen after LH lesions are due to the direct disruption in the function of intrinsic LH neurons or due to the interruption of the fibers of passage. The present study addressed this issue by examining the effect of neurotoxic doses of NMDA into the LH, which presumably acts on cell bodies without affecting fibers of passage.

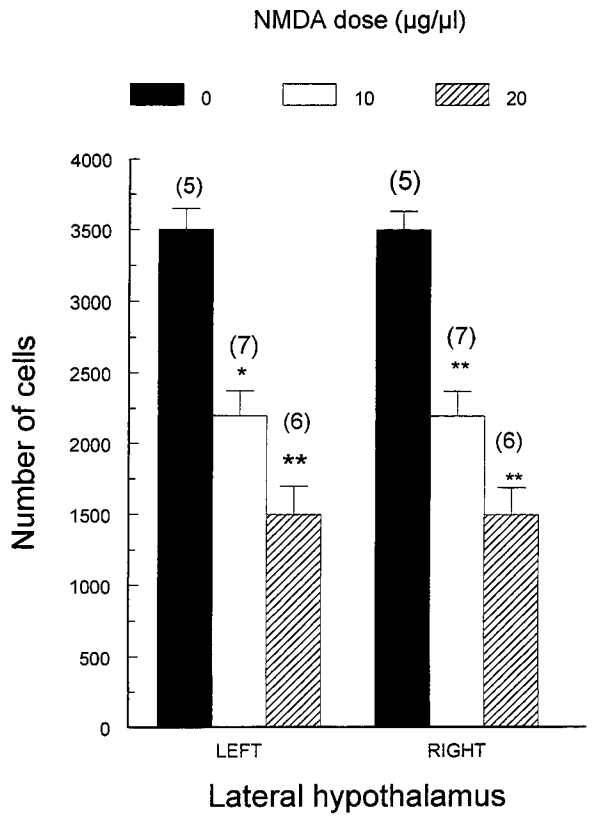

Figure 9. Mean (±*SEM*) cell count in the left and right lateral hypothalamus (LH) under high-power magnification 24 hr after microinjections of *N*-methyl-D-aspartate (NMDA; 10 or 20 μg/μl) or phosphate buffered saline (0 μg/μl). Numbers in parentheses indicate the number of subjects. The 0 μg/μl bar represents pooled data from 3 rats receiving vehicle infusions into the LH and 2 rats receiving NMDA infusions into the thalamus but without involvement of the LH. *$p < .05$ (in comparison with the 0 and 20 μg/μl doses). **$p < .01$ (in comparison with the 0 μg/μl dose).

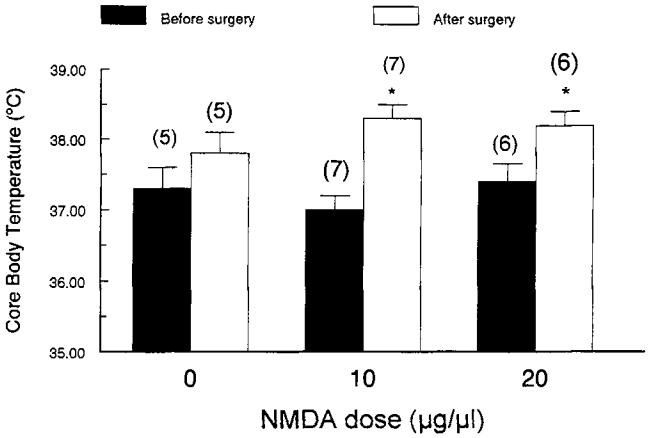

Figure 10. Mean (±*SEM*) core body temperature before and after microinjections of *N*-methyl-D-aspartate (NMDA, 10 or 20 µg/µl) or phosphate buffered saline (0 µg/µl of NMDA) into the lateral hypothalamus (LH). Numbers in parentheses indicate the number of subjects. The 0 µg/µl bar represents pooled data from 3 rats receiving vehicle infusions into the LH and 2 rats receiving NMDA infusions into the thalamus but without involvement of the LH. $*p < .005$ (compared with the before-treatment condition).

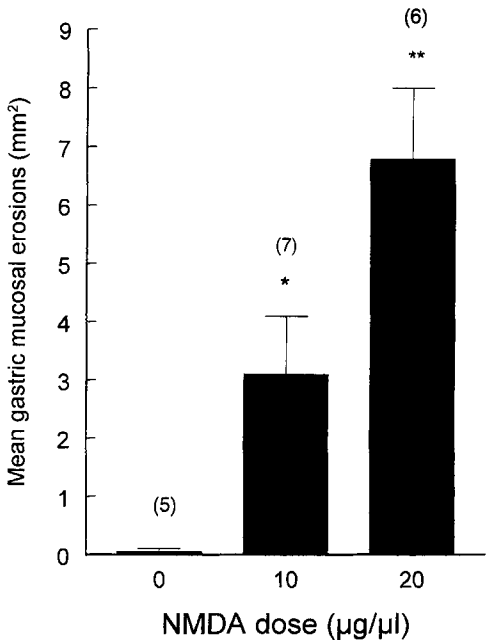

Figure 11. Dose-related gastric erosions induced by microinjections of *N*-methyl-D-aspartate (NMDA; 10 or 20 µg/µl) into the lateral hypothalamus (LH). Each column represents the mean (±*SEM*) total area of gastric mucosal erosions. Numbers in parentheses indicate the number of subjects. The 0 µg/µl bar represents pooled data from 3 rats receiving vehicle infusions into the LH and 2 rats receiving NMDA infusions into the thalamus but without involvement of the LH. $*p < .05$ (in comparison with the 0 and 20 µg/µl doses). $**p < .001$ (in comparison with the 0 µg/µl dose).

This study demonstrates that neurotoxic doses of NMDA into the LH produced many, but not all, of the changes seen after electrolytic LH lesions. For example, NMDA–LH infusions induced initial feeding deficits, hyperthermia, and gastric erosion formation, as reported after electrolytic LH lesions, but did not appear to alter gross motor behavior, as measured by the cage-climbing test. These findings provide strong evidence that interruption of intrinsic LH neurons contribute to many of the dysfunctions that typically characterize the classic LH lesion syndrome. One interesting question is whether some of the changes following NMDA–LH infusions are due primarily to initial acute hyperexcitation of the neurons and whether other changes are due to the subsequent death of the cell following the period of hyperexcitation. As discussed below, there is reason to believe that at least the formation of gastric erosions and possibly hyperthermia are the result of the acute excitatory effects on LH neurons prior to their death and/or the temporary activation of adjacent neurons, which are otherwise spared by the lesion (present study; Roland & Grijalva, 1993).

The results of the present study showed that both electrolytic LH lesions and NMDA infusions in the LH induce gastric erosion formation 24 hr after surgery and that NMDA induces erosion formation in a dose dependent manner (see Experiment 2). These findings indicate that the incidence of gastric erosions is related to a disruption of intrinsic LH neurons and may be mediated by the acute excitatory or activational effects of these lesioning procedures. It has previously been shown that electrolytic LH lesions lead to the development of gastric erosions within as little as 3 hr (Grijalva, Deregnaucourt, et al., 1980); however, erosions tend to disappear within 1 week after LH lesions with adequate postoperative care and nutrition (Grijalva, 1993). Furthermore, rats that recovered from NMDA–LH-induced lesions are erosion free 3–5 weeks after surgery (Winn et al., 1990). Support for the "hyperactivation hypothesis" comes from other studies showing that LH electrical or chemical stimulation increases gastric acid secretion (Carmona & Slangen, 1973; Misher & Brooks, 1966), and immediately after either electrical stimulation or electrolytic lesions of the LH there is an increase in stomach motility and acid secretion (Folkow & Rubinstein, 1965; Garrick et al., 1993; Glavcheva, Manchanda, Box, & Stevenson, 1972; Taché et al., 1982). Other lines of evidence indicate that it is not cell death per se but the hyperactivation of the cells that precede death that causes the formation of gastric erosions. For example, axonal transections produced by knife cuts placed lateral, anterior, or posterior to the LH do not lead to erosion formation (Grijalva, Novin, & Bray, 1980; Landeira-Fernandez & Grijalva, 1999); however, parasagittal knife cuts placed lateral to the LH block the occurrence of gastric erosions produced by bilateral NMDA infusion into the LH. This finding suggests that cuts placed lateral to the LH interrupt the outflow of laterally projecting nerve fibers from LH, which, in turn, take a descending route to the nucleus

tractus solitarius and the dorsal motor nucleus of the vagus (Berk & Finkelstein, 1982; Hosoya & Matsushita, 1981; Ricardo & Koh, 1978). The acute effects of LH lesions on gastric erosion formation appear to be related to vagally mediated increases in gastric acid secretion and gastric contractility (Garrick et al., 1993; Grijalva, Deregnaucourt, et al., 1980). On the other hand, the chronic effects of the lesions may result in a reduction in the neuromodulation of the gut and may actually lead to resistance in the development of subsequent gastric erosion provoked by stress-inducing procedures by eliminating the facilitatory influence of the LH on the upper gastrointestinal tract. This idea is supported by the fact that recovered LH-lesioned rats do not exhibit gastric erosions (Winn et al., 1990) and NMDA-induced lesions of the LH attenuate the incidence of gastric erosion produced by cold-water restraint (Landeira-Fernandez, Jentjens, Machado, Carotti, & Grijalva, 1998).

Although there is strong evidence showing that LH lesions induce gastric erosions mainly by increasing parasympathetic–vagal activity, other lines of evidence demonstrate that LH lesions also produce an abrupt increase in sympathetic activity, which also contributes to erosion formation and other disorders associated with the lesion (Grijalva, Lindholm, & Novin, 1980; Tordoff et al., 1984). Recent findings indicate that hyperthermia produced by electrolytic LH lesions is related to cerebral prostaglandin synthesis and sympathetic activation of thermogenic brown adipose tissue (BAT). For example, intraperitoneal or intracerebroventricular administration of prostaglandin synthesis inhibitors attenuates fever and reduces the increased firing rate of sympathetic nerve fibers, innervating interscapular BAT produced by electrolytic LH lesions (Lennie et al., 1995; Monda, Amaro, et al., 1996; Monda, Sullo, De Luca, & Pellicano, 1996). Thus, it appears that cerebral prostaglandins are involved in the control of sympathetic discharge of BAT, which, in turn, contributes to fever production. It has been proposed that increases in sympathetic activity reduce food intake (Bray, 1991), and this idea is supported by the finding that intraperitoneal administration of lysine acetylsalicylate, a prostaglandin synthesis inhibitor, not only reduces the firing rate of BAT sympathetic fibers and attenuates the hypothermia following electrolytic LH lesions but also modifies the postoperative aphagia (Monda, Sullo, et al., 1996). Given the fact that NMDA infusions into the LH and electrolytic LH lesions produced similar increases in core body temperature in the present study, it is reasonable to propose that the acute change in temperature is due to an alteration in the function of intrinsic LH neurons rather than to the interruption of fibers of passage.

Although the present findings provide support for the view that gastric erosion formation and possibly hyperthermia induced by electrolytic LH lesions or neurotoxic doses of NMDA are related to abrupt, acute hyperexcitation of neurons prior to their death, other dysfunctions associated with LH damage may be due to the interruption of axonal fibers of passage. For example, electrolytic LH lesions not only destroy neurons but also damage the nigrostriatal-DA pathway, and motor and sensorimotor deficits displayed by LH-lesioned rats appear to be due to a chronic reduction in endogenous brain DA levels (Marshall, Richardson, & Teitelbaum, 1974; Stricker & Zigmond, 1976). As shown in the present study, rats given electrolytic LH lesions displayed impairments in cage-climbing behavior, whereas those given NMDA–LH infusions did not. This finding suggests that intrinsic LH neurons may not be involved in motor functions required for cage-climbing behavior and further suggests that axons of neural systems coursing through the LH area, responsible for motor behavior, were preserved. Furthermore, other reports indicate that DA, both centrally and peripherally, is involved gastric erosion. For example, the occurrence of gastric erosions induced by physical stressors is potentiated by DA antagonists and attenuated by DA agonists (Hernandez et al., 1984). Likewise, gastric erosions produced by electrolytic LH lesions can be prevented by the peripheral administration of the DA agonist apomorphine (Roland & Grijalva, 1993). Nevertheless, a reduction in brain DA levels is not necessary for the production of gastric erosions. For example, infusion of NMDA into the LH, which is sufficient to produce gastric erosions (present study), does not appear to affect DA levels in the striatum (Winn et al., 1984).

A final issue to be raised is the involvement of intrinsic LH neurons in eating. Stanley and colleagues (Stanley, Ha, et al., 1993; Stanley, Willett, et al., 1993; Stanley et al., 1996, 1997) have accumulated considerable evidence that endogenous LH glutamate acts to stimulate feeding through the action on NMDA receptors. These findings are supported by the present study, which shows that, like electrolytic LH lesions, neurotoxic doses of NMDA into the LH produce an initial disruption in feeding behavior. Nevertheless, the nature of both the short- and long-term feeding impairments following either LH electrolytic lesions or excitotoxic NMDA infusions is not entirely the same (present study; A.J.M. Clark et al., 1990) and reinforces the view that the full-blown LH lesion feeding syndrome (Epstein, 1971; Stricker & Zigmond, 1976) is the result of combined damage to intrinsic LH neurons and fibers of passage (Grossman, 1975; Roland & Grijalva, 1993). Although the present study showed that NMDA infusions into the LH initially reduced eating behavior, it has been reported that 20 days after surgery rats with NMDA–LH lesions do not display gross feeding deficits (A. J. M. Clark et al., 1998). Taken together, these findings suggest that the initial feeding deficits induced by NMDA infusions may be related to an acute disruption in autonomic and metabolic regulation, leading to disturbances in gastrointestinal function and thermoregulation.

References

Anand, B. K., & Brobeck, J. R. (1951). Hypothalamic control of food intake. *Yale Journal of Biology and Medicine, 24,* 123–140.

Baillie, P., & Morrison, S. D. (1963). The nature of the suppression of food intake by lateral hypothalamic lesions. *Journal of Physiology, 165,* 227–245.

Balagura, S., Wilcox, R. H., & Coscina, D. V. (1969). The effect of diencephalic lesions on food intake and motor activity. *Physiology and Behavior, 4,* 629–633.

Berk, M. L., & Finkelstein, J. A. (1982). Efferent connections of the lateral hypothalamic area of the rat: An autoradiographic investigation. *Brain Research Bulletin, 8,* 511–526.

Bernardis, L. L., & Bellinger, L. L. (1993). The lateral hypothalamic area revised: Neuroanatomy, body weight regulation, neuroendocrinology and metabolism. *Neuroscience and Biobehavioral Review, 17,* 141–193.

Bloom, W., & Fawcett, D. W. (1975). *A textbook of histology.* Philadelphia: W. B. Saunders.

Bray, G. A. (1991). Reciprocal relation between the sympathetic nervous system and food intake. *Brain Research Bulletin, 27,* 517–520.

Carmona, A., & Slangen, J. A. (1973). Effects of chemical stimulation of the hypothalamus upon gastric secretion. *Physiology and Behavior, 10,* 657–661.

Clark, A. J. M., Clark, J. M., & Winn, P. (1990). NMDA lesions of the rat lateral hypothalamus: Effects of dietary and physiological challenges. *NeuroReport, 1,* 263–266.

Clark, J. M., Clark, A. J. M., Bartle, A., & Winn, P. (1991). The regulation of feeding and drinking in rat with lesions of the lateral hypothalamus made by *N*-methyl-D-aspartate. *Neuroscience, 45,* 631–664.

Coffey, P. J., Perry, V. H., Allen, Y., Sinden, J., & Rawlins, J. N. P. (1988). Ibotenic acid induced demyelination in the central nervous system: A consequence of a local inflammatory response. *Neuroscience Letters, 84,* 178–184.

Corbett, S. W., Wilterdink, E. J., & Keesey, R. E. (1985). Heat production and body weight changes following lateral hypothalamic lesions. *Physiology and Behavior, 35,* 971–977.

DeRyck, M., & Teitelbaum, P. (1978). Neocortical and hippocampal EEG in normal and lateral hypothalamic damaged rats. *Physiology and Behavior, 20,* 403–409.

Dingledine, R. (1991). New wave of non-NMDA excitatory aminoacid receptors. *Trends in Pharmacological Sciences, 12,* 360–362.

Epstein, A. N. (1971). The lateral hypothalamic syndrome: Its implications for the physiological psychology of hunger and thirst. In E. Stellar & J. M. Sprague (Eds.), *Progress in physiological psychology (Vol. 4,* pp. 236–317). New York: Academic Press.

Farber, J. L., Chien, K. R., & Mittnacht, S. (1981). Myocardial ischemia: The pathogenesis of irreversible cell injury in ischemia. *American Journal of Pathology, 102,* 271–281.

Folkow, B., & Rubenstein, E. H. (1965). Behavioral and autonomic patterns evoked by stimulation of the lateral hypothalamic area in the cat. *Acta Physiologica Scandinavia, 65,* 292–299.

Garrick, T., Grijalva, C. V., & Trauner, M. (1993). Lateral hypothalamic lesions cause gastric injury by stimulating gastric contractility. *American Journal of Physiology, 265,* G138–G142.

Glavcheva, L., Manchanda, S. K., Box, B., & Stevenson, J. A. F. (1972). Gastric motor activity during feeding induced by stimulation of the lateral hypothalamus. *Canadian Journal of Physiology and Pharmacology, 50,* 1091–1098.

Grijalva, C. V. (1980). Aphagia, gastric pathology, hyperthermia, and sensorimotor dysfunctions after lateral hypothalamic lesions: Effects of insulin pretreatments. *Physiology and Behavior, 25,* 931–937.

Grijalva, C. V. (1993). [Effect of lateral hypothalamic lesions on gastrointestinal properties]. Unpublished raw data.

Grijalva, C. V., Deregnaucourt, J., Code, C. F., & Novin, D. (1980). Gastric mucosal damage in rats induced by lateral hypothalamic lesions: Protection by propantheline, cimetidine and vagotomy. *Proceedings of the Society of Experimental and Biological Medicine, 163,* 528–533.

Grijalva, C. V., & Lindholm, E. (1980). Restricted feeding and its effects on aphagia and ingestion-related disorders following lateral hypothalamic damage. *Journal of Comparative Physiological Psychology, 1,* 164–177.

Grijalva, C. V., Lindholm, E., & Novin, D. (1980). Physiological and morphological changes in the gastrointestinal tract induced by hypothalamic intervention: An overview. *Brain Research Bulletin, 5,* 19–31.

Grijalva, C. V., Lindholm, E., & Roland, B. (1989). Recovery of function following lateral hypothalamic damage: The effects of preoperative manipulations. In J. Schulkin (Ed.), *Preoperative events: Their effects on behavior following brain damage* (pp. 35–63). Hillsdale, NJ: Erlbaum.

Grijalva, C. V., Lindholm, E., Schallert, T., & Bicknell, E. (1976). Gastric pathology and aphagia following lateral hypothalamic lesions in the rat: Effects of preoperative weight reduction. *Journal of Comparative and Physiological Psychology, 90,* 505–519.

Grijalva, C. V., & Novin, D. (1990). The role of hypothalamus and dorsal vagal complex in gastrointestinal function and pathology. *Annals of the New York Academy of Science, 597,* 207–222.

Grijalva, C. V., Novin, D., & Bray, G. A. (1980). Alterations in blood glucose, insulin, and free fatty acids following lateral hypothalamic lesions or parasagittal knife cuts. *Brain Research Bulletin, 5* (Suppl. 4), 109–117.

Grossman, S. P. (1975). Role of the lateral hypothalamus in the regulation of food and water intake. *Psychological Review, 82,* 200–224.

Harrell, L. E., de Castro, J. M., & Balagura, S. (1975). A critical evaluation of body weight loss following lateral hypothalamic lesions. *Physiology and Behavior, 15,* 133–136.

Hastings, M. H., Winn, P., & Dunnett, S. B. (1985). Neurotoxic aminoacid lesions of the lateral hypothalamus: A parametric comparison of the effects of ibotenic, *N*-methyl-D,L-aspartate and quisqualate in the rat. *Brain Research, 360,* 248–256.

Hernandez, D. E., Adcock, J. W., Orlando, R. C., Patrick, K. S., Nemeroff, C. B., & Prange, A. J. (1984). Prevention of stress-induced gastric ulcers by dopamine agonists in the rat. *Life Science, 35,* 2453–2458.

Hosoya, Y., & Matsushita, M. (1981). Brain stem projections from the lateral hypothalamic area in the rat, as studied with autoradiography. *Neuroscience Letters, 24,* 111–116.

Kohler, C., & Schwarcz, R. (1983). Comparison of ibotenic and kainic neurotoxicity in rat brain: A histological study. *Neuroscience, 8,* 819–835.

Landeira-Fernandez, J., & Grijalva, C. V. (1999). Gastric mucosal erosion produced by NMDA microinfusions in the lateral hypothalamus: Effect of selective knife cuts. *Behavioural Brain Research, 102,* 51–60.

Landeira-Fernandez, J., Jentjens, O., Machado, M. A., Carotti, A. P., & Grijalva, C. V. (1998). Stress-induced ulceration depends on NMDA receptors located in the lateral hypothalamus. *Society for Neuroscience Abstracts, 24* (Pt. 2), 946.

Lennie, T. A., Hirvonen, M. D., McCarthy, D. O., & Keesey, R. E. (1995). Fever and acute elevation in whole-body thermogenesis induced by lateral hypothalamic lesions. *Physiology and Behavior, 58,* 237–243.

Levitt, D. R., & Teitelbaum, P. (1975). Somnolence, akinesia and sensory activation of motivated behavior in the lateral hypothalamic syndrome. *Proceedings of the National Academy of Science of the United States of America, 72,* 2819–2823.

Lindholm, E., Shumway, G. S., Grijalva, C. V., Schallert, T., & Ruppel, M. (1975). Gastric pathology produced by hypothalamic lesions in rats. *Physiology and Behavior, 14,* 165–169.

López-Colomé, A. M., Ortega, A., & Romo-de-Vivar, M. (1993). Excitatory amino acid–induced phosphoinosite hydrolysis in Müller glial. *Glia, 9,* 127–135.

Markowska, A., Bakke, H. K., Walther, B., & Ursin, H. (1985). Comparison of electrolytic and ibtoenic acid lesions in the lateral hypothalamus. *Brain Research, 328,* 313–323.

Marshall, J. F., Richardson, J. S., & Teitelbaum, P. (1974). Nigrostriatal bundle damage and the lateral hypothalamic syndrome. *Journal of Comparative and Physiological Psychology, 87,* 808–830.

Marshall, J. F., & Teitelbaum, P. (1974). Further analysis of sensory inattention following lateral hypothalamic damage in rats. *Journal of Comparative Physiological Psychology, 86,* 375–395.

Misher, A., & Brooks, F. D. (1966). Electrical stimulation of hypothalamus and gastric secretion in the albino rat. *American Journal of Physiology, 211,* 403–406.

Monaghan, D. T., Bridges, R. J., & Cotman, C. W. (1989). The excitatory amino acid receptors: Their classes, pharmacology, and distinct properties in the function of the central nervous system. *Annual Review of Pharmacology and Toxicology, 29,* 365–402.

Monda, M., Amaro, A., Sullo, A., & De Luca, B. (1996). Lateral hypothalamic lesion induces sympathetic stimulation and hyperthermia by activating synthesis of cerebral prostaglandins. *Prostaglandins, 51,* 169–178.

Monda, M., Sullo, A., De Luca, E., & Pellicano, M. P. (1996). Lysine acetylsalicylate modifies aphagia and thermogenic changes induced by lateral hypothalamic lesion. *American Journal of Physiology, 272,* R1638–R1642.

Müller, T., Grosche, J., Ohlemeyer, C., & Kettenmann, H. (1993). NMDA-activated currents in Bergmann glial cells. *NeuroReport, 4,* 671–674.

Namiki, T., Egawa, M., Inoue, S., Tominaga, S., & Takamura, Y. (1994). Gastric damage and body weight loss in LHA-lesioned rats: Effects of cytoprotective geranylgeranylacetone. *American Journal of Physiology, 266,* R503–R509.

Paxinos, G., & Watson, C. (1986). *The rat brain in stereotaxic coordinates.* Sydney, Australia: Academic Press.

Peterson, G. M., & Moore, R. Y. (1980). Selective effects of kainic acid on diencephalic neurons. *Brain Research, 202,* 165–182.

Powley, T. L., Opsahl, C. A., & Weingarten, H. P. (1980). The role of the hypothalamus in energy homeostasis. In P. J. Morgane & J. Panksepp (Eds.), *Handbook of the hypothalamus: Behavioral studies of the hypothalamus* (Vol. 3, Pt. A, pp. 211–298). New York: Marcel Dekker.

Refinetti, R., & Carlisle, H. J. (1987). A reevaluation of the role of the lateral hypothalamus in behavioral temperature regulation. *Physiology and Behavior, 40,* 189–192.

Ricardo, J. A., & Koh, E. T. (1978). Anatomical evidence of direct projections from the nucleus of the solitary tract to the hypothalamus, amygdala, and other forebrain structures in the rat. *Brain Research, 153,* 1–26.

Roland, B., & Grijalva, C. V. (1993). Gastric mucosal erosions induced by lateral hypothalamic damage: Neuronal and dopaminergic mechanisms. *Brain Research, 605,* 110–120.

Rothman, S. M., & Olney, J. W. (1987). Excitotoxicity and the NMDA receptor. *Trends in Neuroscience, 10,* 299–302.

Schallert, T., Leach, L. R., & Braun, J. J. (1978). Saliva hypersecretion during aphagia following lateral hypothalamic lesions. *Physiology and Behavior, 21,* 461–463.

Schallert, T., Whishaw, I. Q., & Flanningan, K. P. (1977). Gastric pathology and feeding deficits induced by hypothalamic damage in rats: Effects of lesion type, size, and placement. *Journal of Comparative and Physiological Psychology, 91,* 598–610.

Scott, J. W., & Leonard, C. M. (1971). The olfactory connections of the lateral hypothalamus in the rat, mouse and hamster. *Journal of Comparative Neurology, 141,* 331–344.

Stanley, B. G., Butterfield, B. S., & Grewal, R. S. (1997). NMDA receptor coagonist glycine site: Evidence for a role in lateral hypothalamic stimulation of feeding. *American Journal of Physiology, 273,* R790–R796.

Stanley, B. G., Ha, L. H., Spear, L. C., & Dee, M. G., II. (1993). Lateral hypothalamic injections of glutamate, kainic acid, D,L-α-amino-3-hydroxy-5-methyl-isoxazole propionic acid or *N*-methyl-D-aspartic acid rapidly elicit intense transient eating in rats. *Brain Research, 613,* 88–95.

Stanley, B. G., Willett, V. L., III, Donias, H. W., Dee, M. G., II, & Duva, M. A. (1996). Lateral hypothalamic NMDA receptors and glutamate as physiological mediators of eating and weight control. *American Journal of Physiology, 270,* R443–R449.

Stanley, B. G., Willett, V. L., Donias, H. W., Ha, L. H., & Spears, L. C. (1993). The lateral hypothalamus: A primary site mediating the excitatory amino acid–elicited eating. *Brain Research, 630,* 41–50.

Stellar, J. R., Hall, F. S., & Waraczynski, M. (1991). The effect of excitotoxin lesions of the lateral hypothalamus on self-stimulation reward. *Brain Research, 541,* 29–40.

Stricker, E. M., & Zigmond, M. J. (1976). Recovery of function after damage to the central catecholamine-containing neurons: A neurochemical model for the lateral hypothalamic syndrome. In J. M. Sprague & A. N. Epstein (Eds.), *Progress in psychobiology and physiological psychology* (Vol. 6, pp. 121–188). New York: Academic Press.

Taché, Y., Grijalva, C. V., Gunion, M. W., Walsh, J. H., & Novin, D. (1982). Stimulation of gastric secretion by acute lateral hypothalamic lesions and its reversal by intracisternal bombesin. *Life Science, 31,* 2485–2491.

Teitelbaum, P., & Epstein, A. N. (1962). The lateral hypothalamic syndrome: Recovery of feeding and drinking after lateral hypothalamic lesions. *Psychological Review, 69,* 74–90.

Tordoff, M. G., Grijalva, C. V., Novin, D., Butcher, L. L., Walsh, J., Pi-Sunyer, X., & VanderWeele, D. A. (1984). Influence of sympathectomy on the lateral hypothalamic lesion syndrome. *Behavioral Neuroscience, 98,* 1039–1059.

Turski, L., Bressler, K., Rettig, K. J., Loschmann, P. A., & Wachtel, H. (1991, January 31). Protection of substantia nigra from MPP+ neurotoxicity by *N*-methyl-D-aspartate antagonists. *Nature, 349,* 414–418.

Winn, P. (1991). Excitotoxins as tools for producing brain lesions. In P. M. Conn (Ed.), *Methods in neuroscience: Lesions and transplantation* (Vol. 7, pp. 16–27). San Diego, CA: Academic Press.

Winn, P., Clark, A., Hastings, M., Clark, J., Latimer, M., Rugg, E., & Brownlee, B. (1990). Excitotoxic lesions of the lateral hypothalamus made by *N*-methyl-D-aspartate in the rat: Behavioral, histological and biochemical analysis. *Experimental Brain Research, 82,* 628–636.

Winn, P., Tarbuck, A., & Dunnett, S. B. (1984). Ibotenic acid lesions of the lateral hypothalamus: Comparison with the electrolytic lesion syndrome. *Neuroscience, 12,* 225–240.

Received November 23, 1998
Revision received March 26, 1999
Accepted April 6, 1999 ■