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RAPID COMMUNICATION

Sensitivity of Lateral Hypothalamic Neurons to Nicotine: Origin and Possible Correlation With Nutritional Effects of Nicotine

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HIMMI, T., F. BRAHITI, J. PERRIN AND J. C. ORSINI. *Sensitivity of lateral hypothalamic neurons to nicotine: Origin and possible correlation with nutritional effects of nicotine.* PHARMACOL BIOCHEM BEHAV 44(1) 217–200, 1993. – Single-unit activity was recorded extracellularly in the lateral hypothalamus of anesthetized rats. A number of neurons responded to intravenous nicotine, but most failed to respond similarly to local nicotine or systemic administration of a peripheral acting agonist. This finding suggests that these neurons respond indirectly to systemic nicotine through afferent pathways originating in central nicotinoceptive cells. The incidence of response was significantly greater in the cells sensitive to moderate changes in blood glucose. This finding suggests that the effects of peripheral nicotine on food intake and body weight are partly mediated by "glycemia-sensitive neurons" in the lateral hypothalamus.

Nicotine Blood glucose Tetramethylammonium Lateral hypothalamus Microiontophoresis Feeding behavior

BODY weight gain is lessened by tobacco use in humans (31), and by nicotine administration in rats (21,29). This effect has been attributed to higher energy consumption (9,17) and depressed appetite for sweet-tasting food (8). Some authors have claimed that nicotine has a more general anorectic effect (15,21,32), but this point is controversial (4,29). These observations suggest that nicotine may act on central neurons involved in feeding behavior.

The lateral hypothalamic area (LHA) is one of the essential brain structures in the control of food intake, and particularly glucoprivic eating: Bilateral selective lesion of LHA cell bodies abolish the feeding response to 2-deoxy-D-glucose (7), and a number of LHA units are excited by intracerebroventricular injection of 2-deoxy-D-glucose and tonically activated during meals (14). Based on evidence that IV nicotine causes many LHA neurons to modify their spontaneous activity, we previously speculated that the effects of nicotine on feeding behavior are partly mediated by these cells (12).

The present study was carried out to further assess the sensitivity of LHA neurons to systemic nicotine by attempting to determine whether responses are mediated by a direct or indirect mechanism. Direct action is a reasonable hypothesis since the permeability of blood-brain barrier to this compound (5,22,30), as well as the existence of nicotinic receptors in the hypothalamus (6) have been clearly established. However, indirect action has been observed in other areas of the brain (10), and it cannot be ruled out that LHA cells might be affected through afferent pathways originating in central or peripheral nicotinoceptive elements. We also attempted to determine whether IV nicotine acts preferentially on "glycemia-sensitive" LHA neurons; that is, the cells responsive to moderate changes in blood glucose level, because they would be more likely to have a nutritional function (11). Brief accounts of some of the data presented have been reported elsewhere (3).

METHOD

Experiments were performed on adult male Sprague-Dawley rats weighing 334 ± 63 g (SD), anesthetized by continuous infusion of ketamine hydrochloride into the left femoral vein (Imalgène 500, Rhône-Mérieux, rate 40–70 μg/min). A previous study (11) showed that this method of anesthesia has no significant effect on blood glucose level. Rectal temper-

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ature, which was monitored using a thermistor, was maintained at 37 ± 0.5°C using an electric blanket.

All systemically administered compounds were dissolved in 0.15 M-NaCl and injected into the right jugular vein. The effective dose of nicotine hydrogen tartrate (Sigma Chemical Company, St. Louis, MO; 30–40 μg/Kg) had been previously determined (12). The doses of mecamylamine (Sigma, 0.5 mg/Kg) and tetramethylammonium (TMA, Sigma, 0.1 mg/Kg) were similar to those used in other electrophysiological studies (10,19). Transient hyperglycemia was induced by a 0.4 ml injection of 10% glucose (11).

Seven barrel glass capillary pipettes were used for microiontophoretic application of nicotine using calibrated DC currents (Bionic Instrument Units, Briis-Sous-Forges, France). The outer barrels were filled with 0.5 M-nicotine hydrogen tartrate (pH 3.5, DC resistance 10–30 MΩ), and 0.2 M-NaCl for current equilibration and control tests of possible current effects (DC resistance 10–40 MΩ). Retaining currents were routinely used. Extracellular action potentials were recorded through either a single micropipette or the central barrel of the multibarrelled electrode filled with 3 M-NaCl (DC resistance 4–14 MΩ) connected to a cathode-follower. Spikes observed on an oscilloscope were selected by a window discriminator. Frequency was measured and plotted as a histogram by means of a microcomputer (Apple II GS). Statistical analysis of data was performed as previously described (23). The electrode tip was positioned in the LHA according to stereotaxic coordinates (25). At the end of each experiment, the location of the cell studied was marked by creating a small electrolytic lesion at the tip of the electrode (18) and then the rat was perfused with a fixative solution (10% formalin). To locate the lesion, frozen serial sections (50-μm thick) were cut in a coronal plane and stained with cresyl-violet.

RESULTS

Out of 91 LHA cells tested with IV nicotine injection, 40 responded within a few minutes. The response consisted in a decrease of activity in 28 (Fig. 1), and an increase in 12 (Fig. 2).

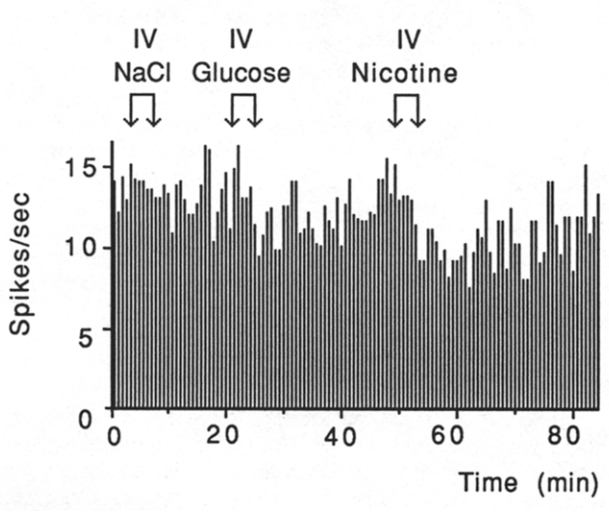

FIG. 1. Effect of IV glucose and nicotine on an LHA neuron. The cell was unaffected by a control injection of NaCl 0.43 M. The spontaneous firing rate was slightly but significantly depressed for more than 20 min after glucose injection. Nicotine injection led to an even greater depression lasting about 25 min of this "glycemia-sensitive neuron."

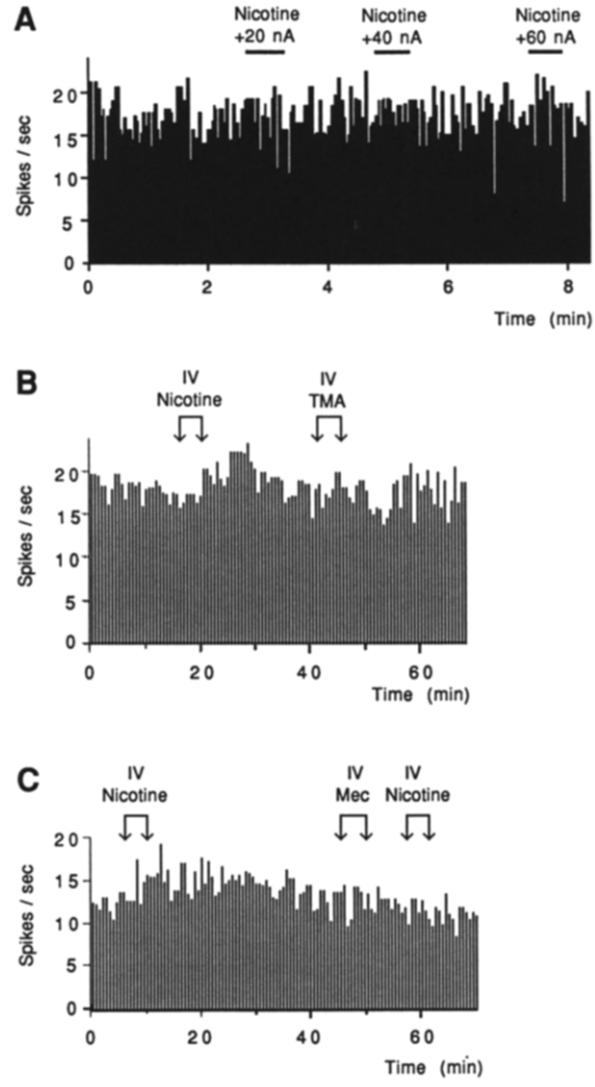

FIG. 2. Example of an LHA neuron unaffected by local nicotine (A), and consistently activated by intavenous nicotine (B, C). The peripherally acting agonist tetramethylammonium (TMA) had no effect (B). The response to nicotine was abolished by injection of mecamylamine (mec), a centrally acting antagonist (C).

Topical microiontophoretic applications of nicotine also triggered responses in many LHA neurons (20/37), with a mean threshold of 58.0 ± 24.2 nA. However, comparison of the effects of topical and systemic administration in 26 neurons revealed discordant responses in all but 2 units, which were depressed by local as well as systemic nicotine. The neuron presented in Fig. 2 responded to IV nicotine, but not to topical application. Conversely, the neuron presented in Fig. 3 responded to local application but not to systemic administration.

Since most neurons responsive to IV nicotine were unresponsive to topical application, we tried to determine whether response was due to the activation of central rather than peripheral receptors. To this end, 11 cells sensitive to IV nicotine were tested after administration of IV TMA, a quaternary nicotinic agonist unable to cross the blood–brain barrier. Only one responded to both compounds (by a decrease in activity).

The 10 others responded only to nicotine. The cell presented in the Fig. 2 was consistently activated by nicotine but was unaffected by TMA. Moreover, its response to nicotine was abolished after the administration of mecamylamine, a central acting antagonist.

To assess the possibility that the same cells were sensitive to IV glucose and nicotine, the response of 68 neurons to the two substances was investigated. The 68 neurons were localized 1.3 to 2.1 mm lateral to the sagittal plane, and their anteroposterior and dorso-ventral position is represented in Fig. 4. The proportion of cells responsive to nicotine was significantly higher among glycemia-sensitive cells than glycemia-insensitive cells (16/23 vs. 15/45, respectively; $\chi^2 = 9.13$; $p < 0.01$). In eight neurons, as the one illustrated in Fig. 1, activity decreased after glucose and nicotine administration. The proportion of cells depressed by IV nicotine was significantly higher among cells depressed by hyperglycemia than other cells tested (8/14 vs. 11/54 respectively, $\chi^2 = 7.47, p < 0.01$).

DISCUSSION

In the present study, many LHA neurons responded to IV nicotine injection but most failed to respond similarly to local nicotine. Based on this finding, we concluded that the response to IV administration was mediated by an indirect mechanism involving afferent pathways affected by nicotine. These pathways probably originate mainly from central nervous structures since almost all the cells affected by IV nicotine failed to respond to IV TMA, an agonist unable to cross the blood-brain barrier. Among the fiber tracts projecting to the LHA (1,27), the catecholaminergic pathways are likely candidates because nicotine is an activator of catecholaminergic neurons connected to hypothalamus (26).

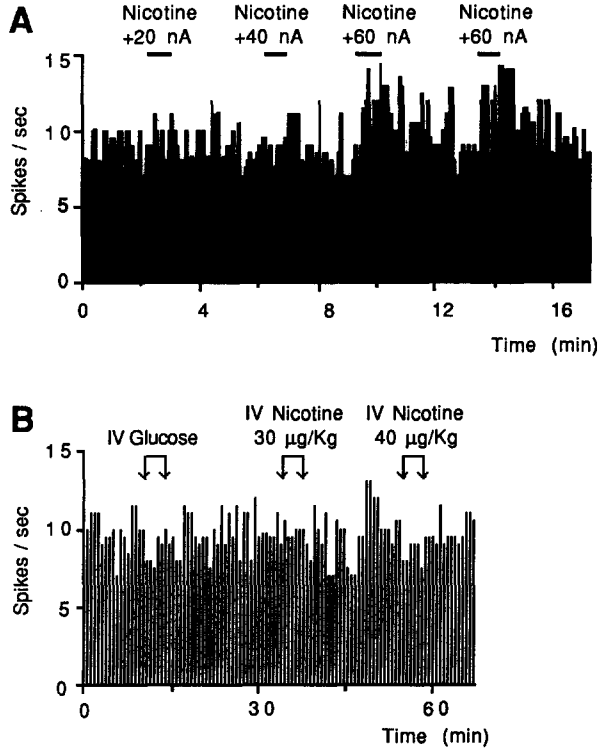

FIG. 3. Example of an LHA neuron that was excited by local nicotine (A), but showed no significant change in activity after IV glucose and nicotine (B).

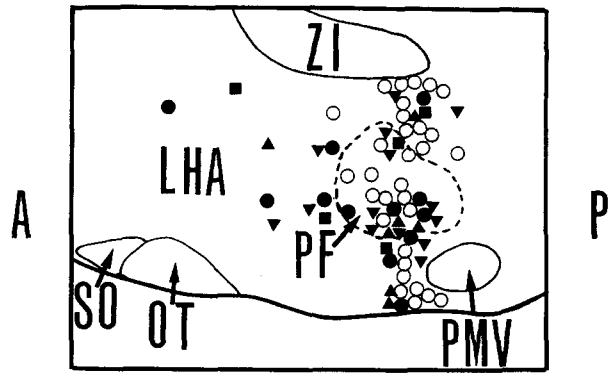

FIG. 4. Monoplanar projection on a parasagittal plane (L 1.4) of the 68 neurons tested under systemic nicotine and glucose administrations. 1) Neurons responding in the same direction to both injections (●). 2) Neurons responding in opposite directions (■). 3) Glycemia-sensitive neurons not responding to IV nicotine (▲). 4) Neurons responding exclusively to systemic nicotine (▼). 5) Unaffected neurons (○). LHA = lateral hypothalamic area; OT = optic tract; PF = perifornical part of the LHA; PMV = premammillary ventral nucleus; SO = supraoptic nucleus; ZI = zona incerta; A and P = anterior and posterior.

A number of neurons responsive to local nicotine failed to respond similarly to IV administration, probably because the intracerebral concentration obtained was below the sensitivity threshold of most nicotinoceptive cells in the LHA. This possibility is consonant with the existence of a low density of high-affinity nicotine binding sites and a high density of low-affinity nicotine binding sites in the hypothalamus (6).

A significantly greater proportion of glycemia-sensitive LHA cells than glycemia-insensitive LHA cells responded to systemic nicotine injection. This finding suggests that the response to nicotine is not due to disturbances in overall status. Nor can the response be considered as a side effect of nicotine-induced hyperglycemia: glycemic changes such as those sometimes observed after nicotine administration or cigarette smoking (2,28) were never detected in our preliminary experiment using the same doses and conditions as here (12). Since glycemia-sensitive cells are likely related to nutritional functions, their greater sensitivity to systemic nicotine is consistent with our hypothesis that some mediate the effects of nicotine on food intake and/or body weight.

Assuming that glycemia-sensitive neurons in the LHA are affected by adrenergic signals (24), it can be speculated that their response to glucose and nicotine is partly mediated by epinephrine and/or norepinephrine afferents arising in the caudal brainstem, and particularly in another feeding-related structure: the solitary tract nucleus. The metabolic activity of this nucleus, which sends direct projections to the LHA (27), is affected by serum levels of glucose and nicotine (13,16), and its neurons respond to local acetylcholine ejections (20). Further experiments are under way to investigate this hypothesis.

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