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Enhanced feeding response to neuropeptide Y in hypothalamic neuropeptide Y-depleted rats

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Abstract

Monosodium glutamate is neurotoxic for the arcuate nucleus and more generally for all circumventricular organs when injected in newborn rats. Neuropeptide Y, a potent stimulator of food intake, is mainly synthesized in the arcuate nucleus. In the present experiment, we determined the hypothalamic status and the feeding response to intracerebroventricular neuropeptide Y in adult rats neonatally treated with monosodium glutamate. Marked neuropeptide Y decreases were measured in the arcuate nucleus and in the paraventricular nuclei in monosodium glutamate-treated rats (−40%; $P < 0.01$). Adult rats neonatally treated with monosodium glutamate weighed significantly less (−8%; $P < 0.01$) and ate less (−10%; $P < 0.01$) than the control rats. Neuropeptide Y injections in a lateral brain ventricle stimulated food intake in control and monosodium glutamate-treated rats in a dose-dependent manner ($P < 0.001$). Whatever the time after drug injection (2, 4, 6 and 8 h) and the injected dose (0.5, 1 and 5 μg), feeding responses were always greater in monosodium glutamate-treated rats (about 2 times greater starting with the lowest dose (0.5 μg): 9.3 ± 1.0 (monosodium glutamate) vs. 5.3 ± 0.7 (control) g/2 h, $P < 0.01$). Calculated minimal effective doses were also always smaller in monosodium glutamate-treated rats than in control animals ($P < 0.01$). Neuropeptide Y increased meal duration, meal size and decreased latency to initiate feeding in monosodium glutamate-treated rats ($P < 0.01$) and control rats ($P < 0.01$). For each dose of neuropeptide Y, effects were more pronounced on meal size (+70%) and meal duration (+25%) in monosodium glutamate-treated rats than in control rats. Therefore, monosodium glutamate-treated rats were more sensitive to exogenous neuropeptide Y. Decreased food intake in the monosodium glutamate-treated rats was associated with a decrease in neuropeptide Y concentrations in the arcuate-paraventricular axis. This confirms the functional role of this peptidergic pathway in eating behavior.

Keywords: Monosodium glutamate; Neurotoxicity; Meal size and duration; Hypophagia; Hypersensitivity to neuropeptide Y; Arcuate nucleus

1. Introduction

Monosodium glutamate is a well known food additive. When injected subcutaneously at high doses in newborn rodents, it damages several areas in the central nervous system where the blood-brain barrier is absent, mainly in the circumventricular organs (Olney, 1969; Lemkey-Johnston and Reynolds, 1974; Takasaki, 1978; Dawson and Lorden, 1981; Meister et al., 1989). A general loss of neurons and fibers is observed in the area postrema, arcuate nucleus and the median eminence (Takasaki, 1978; Olney, 1969; Dawson and Annau, 1983). These anatomical modifications are associated with metabolic and endocrinological disturbances in adulthood, inducing growth stunting and sexual dysfunction (Olney, 1969; Millard et al., 1982; Lorden and Caudle, 1986). A general loss of neuromodulators is observed in the medio-basal hypothalamus (Lemkey-Johnston and Reynolds, 1974; Takasaki, 1978; Dawson and Lorden, 1981; Dawson et al., 1989; Meister et al., 1989; Walaas and Fonnum, 1978). Tyrosine hydroxylase and glutamic acid decarboxylase are decreased in the arcuate nucleus (Meister et al., 1989). Dopamine but not serotonin levels are reduced in the whole hypothalamus (Lorden and Caudle, 1986). Growth hormone-releasing factor completely disappeared in the medio-basal part of the hypothalamus (Millard et al., 1982).

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Neuropeptide Y content diminished in the arcuate nucleus and the paraventricular nucleus (Abe et al., 1990) and the number of neuropeptide Y fibers originating in the arcuate nucleus is decreased in the paraventricular nucleus (Kerkerian and Pelletier, 1986).

Neuropeptide Y is very abundant in the normal brain (Chronwall et al., 1985; Allen et al., 1983). It strongly stimulates food intake (Clark et al., 1984; Stanley and Leibowitz, 1984; Morley et al., 1987a,b; Beck et al., 1992). Measurements of its hypothalamic concentrations (Sahu et al., 1988a; Beck et al., 1990a,b) or release (Kalra et al., 1990; Stricker-Krongrad et al., 1993) have shown that the stimulatory effects of neuropeptide Y on feeding behavior are mediated by neuropeptide Y neurons found in the arcuate nucleus and their projections to the paraventricular nucleus. Involvement of this axis was also found in the obese Zucker rat (Sanacora et al., 1990; McCarthy et al., 1991; Beck et al., 1990c, 1993) and was associated with fasting and refeeding in the normal rat (Sahu et al., 1988a; Beck et al., 1990a; White and Kershaw, 1990; Frankish et al., 1993).

As neonatal treatment with monosodium glutamate also induces hypophagia (Kanarek et al., 1979; Dawson and Lorden, 1981; Dawson and Annau, 1983; Dawson et al., 1989), we measured first, the feeding response to exogenous neuropeptide Y and second, the hypothalamic neuropeptide Y concentrations in the same adult rats neonatally treated with monosodium glutamate. If neuropeptide Y is really implicated in these behavioral disturbances, we hypothesized that they could be directly related to the diminution of neuropeptide Y content in the paraventricular nucleus and the arcuate nucleus, and therefore, responsiveness to exogenous neuropeptide Y may be altered.

2. Materials and methods

2.1. Animals and protocol

Neonate Long-Evans rats were subcutaneously injected with either 4 g/kg body weight monosodium glutamate (Sigma, LaVerpilliere, France) or saline on days 3, 5 and 7 after birth. Injections were counterbalanced among litters. At weaning, female rats were discarded and male rats were fed on a standard rat chow diet (UAR AO4, Villemoisson sur Orge, France). They were housed in single wire cages with food and water ad libitum in an air-conditioned room with a 12-h light/12-h dark cycle with lights on at 7:00. Six months later, 16 rats (8 controls and 8 monosodium glutamate-treated rats) were randomly chosen. At this time and during the rest of the experiment, they were fed on a well-balanced diet supplying 54% of energy from carbohydrate, 16% from protein and 30% from fat (Beck et al., 1990b). After 2 weeks of habituation, their food intakes were measured. Then, they were placed in our system for feeding behavior analysis (FeedBAC) (Stricker-Krongrad et al., 1992) and allowed to habituate for one more week. After implantation of a cannula in the brain lateral ventricle and 1 week of recovery, they were injected with neuropeptide Y to test their feeding response. After the completion of the feeding response study, the rats were killed and the brain was sampled to evaluate the hypothalamic content of endogenous neuropeptide Y.

2.2. Feeding response study

2.2.1. Surgery and injections

All rats were anesthetized with an intraperitoneal injection of chlorhydrate ketamine (130 mg/kg BW; Ketalar, Parke-Davis). They were implanted with a 27 gauge stainless steel needle (10 mm length; Terumo Corporation, Leuven, Belgium)) aimed at the right lateral ventricle. The needle was secured on the skull surface with jeweler's screws and dental cement. A stainless steel stylet was used to occlude the cannula during surgical recovery and between injections.

One week after surgery, the rats were injected either with vehicle (artificial cerebrospinal fluid (CSF)) or with three doses of neuropeptide Y (Sigma Chemicals; La Verpillière, France) dissolved in CSF (0.5, 1 and 5 μg/animal). The injections were given through a 33-gauge stainless steel needle (11 mm length) connected to an Harvard syringe pump via 20 cm of catheter tubing. They were made under controlled light ether anesthesia, which did not exceed 5 min, and then the animals had access to food. The injected volume was fixed at 5 μl and the infusion rate at 2 μl/min. All rats received all doses of neuropeptide Y in a counterbalanced order with a wash-out period of at least 48 h between injections. All injections were done between 10:00 and 11:00 h in the morning. After injections, animals were placed back in the FeedBAC system and food intake was automatically measured every 2 h until the end of the light period e.g. during the 8 h that followed injection. Before the first injection, intra-animal reliability was calculated during a minimum of 3 times 24 h in order to ensure that the animals were well-adapted to the FeedBAC system.

2.2.2. Feeding behavior recording

2.2.2.1. The FeedBAC system. Each cage included a complete automatic FeedBAC system (Feeding Behavior Analysis Computerized system) with food delivery under the animal's control. Each feeding system is composed of a food delivery device (Stricker-Krongrad et al., 1992). The well-balanced diet was mixed with water in order to obtain a paste and a plastic syringe

containing this pasty diet was connected to infrared light diodes which controlled its delivery with a step-to-step motor. It was coupled to an Apple IIe computer. Data from the feeding system were registered in the computer for treatment. Automatic analysis was performed in a second step through a specific software.

2.2.2.2. Analysis of the first meal after injections. A meal was defined as the ingestion of a minimum of 1.2 g of the pasty diet followed by at least 10 min during which no feeding occurred. The following dependent variables and derived measures were calculated as the mean of each variable for each animal: meal size (g), meal duration (min), eating rate (g/min) and latency to eat (min).

2.3. Hypothalamic neuropeptide Y content

2.3.1. Samplings

Animals were killed by decapitation 4 h after the lights went on. Trunk blood was collected in tubes containing Iniprol and EDTA. Brains were quickly removed and frozen (−70°C). Several sections of 300 μm were cut and several areas, namely the arcuate, paraventricular, ventromedial and dorsomedial nuclei and the median eminence were micropunched. Bilateral tissue samples were placed in 500 μl cold extraction solution (HCl 0.2 N/Iniprol/EDTA) and stored at −20°C until assay.

2.3.2. Assay

2.3.2.1. Neuropeptide Y. Neuropeptide Y was measured with a specific radioimmunoassay developed in our laboratory. Neuropeptide Y antibodies were produced in rabbits and cross-react slightly with human pancreatic polypeptide (0.02%) but not with rat pancreatic polypeptide, glucagon, secretin, PHI, rat corticotropin-releasing factor, vasoactive intestinal peptide, polypeptide YY and PHM-27. Standard (porcine neuropeptide Y, SIGMA, La Verpillière, France) or lyophilized unknown sample was reconstituted with assay buffer: 0.04 M phosphate buffer pH 7.4 containing bovine serum albumin (fraction V, Sigma Chemicals, La Verpillière, France), aprotinin (4000 IU/ml, IniprolR, Laboratories Choay, Paris) and sodium azide (Merck, Darmstadt). 100 μl of antiserum diluted in assay buffer and 100 μl of standard or unknown sample were preincubated for 24 h at 4°C. Then, 100 μl of ^{125}I-labelled neuropeptide Y (Amersham IM170, Les Ulis, France) were added and incubated for a further 24 h. Bound and free fractions were separated by the addition of 500 μl of a solution of 2% charcoal (Norit A, Kodak, Rochester, NY) and 0.2% dextran (T70, Pharmacia, Uppsala, Sweden) in assay buffer. The bound fraction was measured in a gamma counter coupled to a microcomputer (MDA 312 system, Kontron, Velizy, France) for the plotting of the standard curve and the calculation of the results. Under these conditions, maximal binding was 32.8 ± 1.7%. A 50% decrease of the bound activity (IC_{50}) was obtained with a concentration of 0.68 ± 0.07 ng/ml neuropeptide Y. Non specific binding was 5.9 ± 0.2%.

2.3.2.2. Plasma insulin and glucose. Plasma glucose was enzymatically measured with a commercially available kit (Boehringer-Manheim, Meylan, France). Immunoreactive insulin was measured by a single antibody-charcoal radioimmunoassay technique using commercially available kits (INSIK, CIS, Gif sur Yvette, France) and rat insulin (Novo, Copenhagen) as standard.

2.4. Statistical analysis

Drug effects were analyzed by regression analysis after semi-logarithmic linearization. Comparisons between experimental groups were done by comparison of the slopes of the drug-effect curves and the calculation of the minimal effective dose. The parameters of the first meal were analyzed with Fisher's two-way analysis of variance for repeated measures and compared one to the other with Student's *t*-test. Probability values smaller than 0.05 (two-tailed) were considered significant. Variables are presented as means ± S.E.M. (standard error of the mean).

3. Results

3.1. Body weight and food intake

Monosodium glutamate-treated rats were lighter and smaller than the control rats (346.9 ± 7.1 (monosodium glutamate) vs. 376.2 ± 9.2 (control) g; $p < 0.01$; Lee index: 0.35 ± 0.10 (monosodium glutamate) vs. 0.28 ± 0.09 (control); $P < 0.05$). They ate less than the control rats (221.0 ± 4.1 (control) vs. 200.0 ± 4.0 (monosodium glutamate) g/10 days; $P < 0.01$).

3.2. Feeding response to neuropeptide Y

3.2.1. Dose-response study

Results are shown in Fig. 1. Food intake following intracerebroventricular (i.c.v.) injection of CSF was not different in monosodium glutamate-treated rats when compared to control rats (after 2 h: 0.3 ± 0.2 (control) vs. 0.7 ± 0.6 (monosodium glutamate) g; after 6 h: 3.6 ± 0.7 (control) vs. 3.2 ± 0.7 (monosodium glutamate) g; after 8 h: 4.0 ± 0.6 (control) vs. 4.7 ± 0.7 (monosodium glutamate) g). Central neuropeptide Y injections stimulated food intake whatever the dose

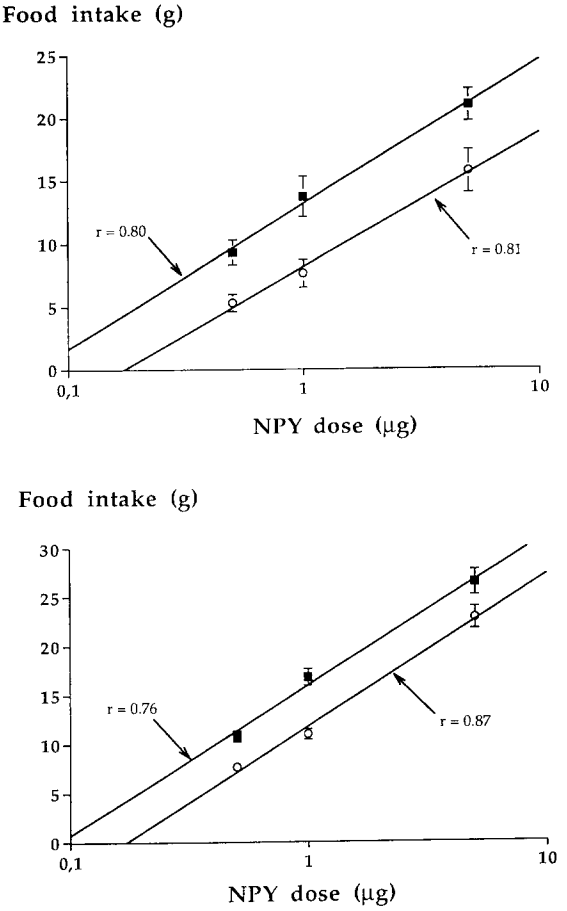

Fig. 1. Dose-response curves for food intake following intracerebroventricular neuropeptide Y (NPY) injections in monosodium glutamate-treated (square; mean ± S.E.M.; $n = 8$) and control rats (circle; mean ± S.E.M.; $n = 8$) (neuropeptide Y doses: 0.5, 1 and 5 μg; top: 2 h and bottom: 6 h following neuropeptide Y injections).

used ($F(3,60) = 16.31$; $P < 0.001$) or the time following drug injection ($F(3,60) = 17.23$; $P < 0.001$) in both monosodium glutamate-treated and control rats. Effects of neuropeptide Y on food intake were dose-dependent in monosodium glutamate-treated rats and control rats, with slopes of the dose-response curves ranging from 7.83 ± 0.82 ($P < 0.05$) to 15.41 ± 0.87 ($P < 0.05$) in monosodium glutamate-treated rats and from 8.07 ± 0.81 ($P < 0.05$) to 12.54 ± 0.75 ($P < 0.05$) in control rats. Cross-examination of slope coefficients at 2 h, 4 h, 6 h and 8 h after drug injections showed that they were not statistically different between monosodium glutamate-treated animals and control animals. Pretreatment with monosodium glutamate altered the responses to central neuropeptide Y ($F(1,62) = 19.37$; $P < 0.001$). 2 h after drug injection, the 0.5 μg dose of neuropeptide Y induced a more pronounced response in monosodium glutamate-treated rats than in control rats (9.3 ± 1.0 (monosodium glutamate) vs. 5.3 ± 0.7 (control) g; $P < 0.01$). The same phenomenom was observed with 1 μg: 13.7 ± 1.6 (monosodium glutamate) vs. 7.6 ± 1.1 (control) g ($P < 0.01$) and with 5 μg: 21.0 ± 1.3 (monosodium glutamate) vs. 15.7 ± 1.7 (control) g ($P < 0.01$). Whatever the time after drug injection and the dose injected, feeding responses were always greater in monosodium glutamate-treated rats than in control animals. Calculated minimal effective doses ranged from 0.18 ± 0.02 μg at 2 h to 0.31 ± 0.05 μg at 8 h for control rats and from 0.06 ± 0.01 μg at 2 h to 0.13 ± 0.04 μg at 8 h for monosodium glutamate-treated rats rats. Whatever the time following drug injection, minimal effective doses were always smaller in monosodium glutamate-treated rats rats than in control rats ($P < 0.001$ or less).

3.2.2. *Effects on the first meal following injection*

Results are shown in Table 1. In basal conditions (vehicle injections), meal size in monosodium glutamate-treated rats rats was smaller than in control rats ($P < 0.05$), but latency to initiate feeding was not modified (N.S.). After central neuropeptide Y injection, meal duration ($F(3,60) = 21.07$; $P < 0.001$), meal size ($F(3,60) = 18.61$; $P = 0.001$) and latency to initiate feeding ($F(3,60) = 16.28$; $P = 0.001$) were modified in monosodium glutamate-treated rats and control rats. In control rats, meal duration ($F(3,28) = 17.03$; $P <$

Table 1
Parameters of the first meal following neuropeptide Y injections in MSG-treated and control rats

NPY injected (μg)	0	0.5	1.0	5.0
Control				
Meal duration (min)	2.5 ± 0.4	15.4 ± 3.3 [a]	17.4 ± 4.2 [a]	46.0 ± 10.5 [a]
Meal size (g)	1.95 ± 0.33	2.57 ± 0.40 [a]	5.43 ± 0.79 [a]	13.01 ± 0.80 [a]
Eating rate (g/min)	0.90 ± 0.25	0.18 ± 0.02	0.34 ± 0.04	0.27 ± 0.02
Latency to eat (min)	61.0 ± 11.3	28.3 ± 2.2 [a]	13.1 ± 0.56 [a]	7.9 ± 2.1 [a]
MSG				
Meal duration (min)	3.5 ± 0.5 [c]	19.3 ± 4.3 [a]	28.2 ± 3.1 [a,b]	68.2 ± 14.1 [a,b]
Meal size (g)	1.34 ± 0.23 [c]	4.41 ± 0.69 [a,b]	7.00 ± 0.84 [a,c]	17.73 ± 2.91 [a,b]
Eating rate (g/min)	0.38 ± 0.18 [c]	0.31 ± 0.07	0.25 ± 0.02	0.33 ± 0.03
Latency to eat (min)	65.0 ± 11.4	16.4 ± 3.2 [a,c]	20.1 ± 2.1 [a,b]	15.3 ± 2.2 [a,b]

[a] $P < 0.01$ vs. control injection for the same rats. [b] $P < 0.01$ vs. control rats for the same dose. [c] $P < 0.05$ vs. control rats for the same dose.

0.01) and meal size ($F(3,28) = 15.67$; $P < 0.01$) increased and latency to initiate feeding decreased in a dose-dependent manner ($F(3,28) = 19.60$; $P < 0.01$). In monosodium glutamate-treated rats, central neuropeptide Y injections induced the same phenomena for meal duration ($F(3,28) = 13.21$; $P < 0.01$) and meal size ($F(3,28) = 21.37$; $P < 0.01$), but when modifications were observed in the latency to initiate feeding ($F(3,28) = 9.37$; $P < 0.05$), they were not dose-dependent. Differences between monosodium glutamate-treated rats and control rats for meal size were significant for each dose of neuropeptide Y ($P < 0.05$ or less). The same phenomenon was observed for meal duration except for the highest neuropeptide Y dose.

3.3. Hypothalamic neuropeptide Y, plasma insulin and glucose

Results are shown in Fig. 2. Neuropeptide Y concentrations significantly decreased in the arcuate and paraventricular nuclei in monosodium glutamate-treated rats when compared to control rats ($P < 0.002$ and $P < 0.01$, respectively). Tendencies for decreases were noted in the median eminence and in the ventromedial and dorsomedial nuclei in monosodium glutamate-treated rats, but they were not significant ($P = 0.07$; $P = 0.11$ and $P = 0.12$, respectively). No changes were observed for plasma glucose (5.89 ± 0.10 (monosodium glutamate) vs. 5.61 ± 0.10 (control) mmol/l; $P = 0.08$) and plasma immunoreactive insulin (6.49 ± 0.43 (monosodium glutamate) vs. 5.51 ± 0.47 (control) ng/ml; $P = 0.15$) when monosodium glutamate-treated rats were compared to control rats.

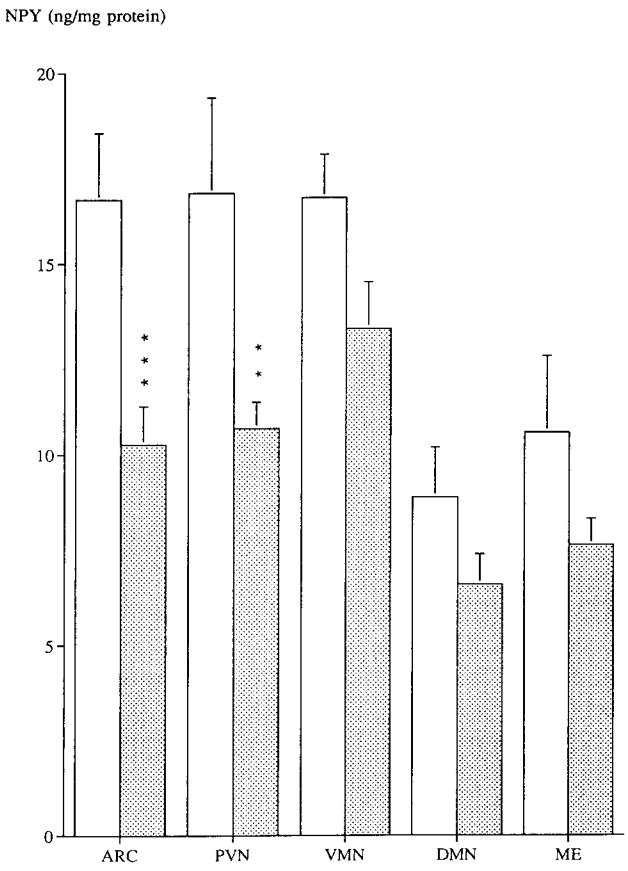

Fig. 2. Neuropeptide Y (NPY) concentrations in hypothalamic areas involved in the regulation of feeding behavior in control (white bars) and monosodium glutamate-treated rats (hatched bars) (** $P < 0.01$ and *** $P < 0.001$). ARC: arcuate nucleus; PVN: paraventricular nucleus; VMN: ventromedial nucleus; DMN: dorsomedial nucleus; ME: median eminence.

4. Discussion

In the present experiment, we studied the effects of monosodium glutamate treatment in newborn rats on different aspects of feeding behavior in relation to neuropeptide Y. In our rats, neonatal treatment with monosodium glutamate induced growth stunting in adulthood and slight hypophagia. This hypophagia was characterized by a decrease in meal size and an increase in meal duration. These effects of monosodium glutamate on food intake and growth have been currently reported in the literature (Lemkey-Johnston and Reynolds, 1974; Dawson and Lorden, 1981; Millard et al., 1982; Dawson and Annau, 1983; Lorden and Caudle, 1986; Dawson et al., 1989; Meister et al., 1989; Olney, 1989; Takasaki, 1978; Walaas and Fonnum, 1978; Arletti et al., 1993). They originate from neurochemical damage in brain circumventricular organs and mainly in the arcuate nucleus (Lemkey-Johnston and Reynolds, 1974; Dawson and Lorden, 1981; Meister et al., 1989; Olney, 1989; Takasaki, 1978; Dawson and Annau, 1983). A general loss of neuromodulators is observed in the medio-basal hypothalamus (Takasaki, 1978; Lorden and Caudle, 1986; Dawson et al., 1989; Meister et al., 1989).

Neuropeptide Y is largely affected by monosodium glutamate treatment (Abe et al., 1990; Kerkerian and Pelletier, 1986; Dawson et al., 1989), because the arcuate nucleus is the main site of its synthesis in the hypothalamus (Chronwall et al., 1985). Its concentrations indeed decreased by about 40% in the monosodium glutamate-treated rats. This decrease had repercussions in the paraventricular nucleus, where neuropeptide Y neurons of the arcuate nucleus mainly project. Indeed, neuropeptide Y levels decreased similarly in the paraventricular nucleus (35%). These results agree with the reports showing diminution of neuropeptide Y content either in the whole hypothalamus (Dawson et al., 1989) or in the paraventricular and arcuate nuclei in monosodium glutamate-treated rats (Abe et al., 1990). The paraventricular nucleus also

receives neuropeptide Y fibers originating from the brainstem where neuropeptide Y is colocalized with monoamines (Sawchenko et al., 1985). The section of these connecting fibers leads to a 50% decrease in neuropeptide Y in the paraventricular nucleus (Sahu et al., 1988b). However, whereas this 50% decrease has no effect on spontaneous eating behavior (Sahu et al., 1989), a smaller decrease due to a neurotoxic effect of monosodium glutamate in the arcuate nucleus induced a small but significant hypophagia. As previously suggested (Beck et al., 1990a,b; Stricker-Krongrad et al., 1993), this confirms that the intrahypothalamic arcuate nucleus-paraventricular nucleus axis plays a preponderant role in the regulation of feeding behavior by neuropeptide Y, whereas the medullo-hypothalamic pathway is less important.

We hypothesized that the behavorial perturbations observed in these animals could partly be related to this neuropeptide Y decrease. Actually, there are no studies dealing with hypothalamic neuropeptide Y depletion in relation to hypophagia in the same manner as neuropeptide Y abundance is associated with hyperphagia as in the Zucker rat (Beck et al., 1990c). Neuropeptide Y abundance was associated with a decrease in sensitivity to exogenous neuropeptide Y injections (Stricker-Krongrad et al., 1994) and a decrease in the total neuropeptide Y receptor number (McCarthy et al., 1991). This indicates that a down-regulation at the receptor level could exist in the obese hyperphagic Zucker rat.

In the present experiment, central injections of neuropeptide Y stimulated food intake in the neuropeptide Y-depleted rats and in the control Long-Evans rats. These effects were dose-dependent. For the control rats, this confirms the results obtained with the Sprague-Dawley rat (Stanley and Leibowitz, 1984) or with other mammals (Morley et al., 1987a; Pau et al., 1988). Neuropeptide Y induced a decrease in the latency to initiate feeding and an increase in meal duration and meal size, starting with the lowest dose injected. For the neuropeptide Y-depleted rats however, the minimal calculated dose necessary to obtain an effect on food intake was 3 times lower than in the control rats, as shown by the shift to the left of their dose-response curve. Meal size and meal duration were also greater in neuropeptide Y-depleted rats than in control rats for the same dose. Therefore, neuropeptide Y-depleted rats were more sensitive to exogenous neuropeptide Y.

An increased sensitivity is also observed when the neuropeptide Y content in the paraventricular nucleus is decreased after section of the connections with the brainstem (Sahu et al., 1989). However, this treatment failed to modify spontaneous food intake and body weight (Sahu et al., 1989). Second, it reduced noradrenaline in addition to hypothalamic neuropeptide Y (Clifton and Sawyer, 1979, 1980) because of the neuropeptide Y-noradrenaline co-localization in the brainstem neurons that project to the paraventricular nucleus (Sawchenko et al., 1985). Third, it increased the responsiveness to adrenergic agonists (Leibowitz and Brown, 1984). In rats treated with monosodium glutamate doses comparable to those of the present experiment, food intake was not different from that of control rats after systemic administration of either an α_2-adrenoceptor agonist (guanfacine) or antagonists (yohimbine, idazoxan) (Dawson et al., 1989). Therefore, it is unlikely that a deficiency of adrenergic transmitter activity, together with neuropeptide Y, contributed to the increased responsiveness of the monosodium glutamate-treated rats to neuropeptide Y. Consequently, denervation-induced hypersensitivity differs from that induced by the neurotoxic effect of monosodium glutamate on neurons of the arcuate nucleus.

Central injection studies have demonstrated that neuropeptide Y can activate the endocrine pancreas and elicits a rise in plasma glucose and insulin (Moltz et al., 1985; Abe et al., 1989). These parameters were not modified in the neuropeptide Y-depleted rats, as also previously observed by others (Tokuyama et al., 1989; Lorden and Caudle, 1986). According to the hypothesis introduced by Olney (Olney, 1969), the hypophagia observed in the monosodium glutamate-treated rats is therefore a primary defect, partly related to the hypothalamic neuropeptide Y depletion, rather than being secondary to endocrine perturbations.

In conclusion, this experiment has shown first that monosodium glutamate treatment induces, in adulthood, hypophagia as a result of a decrease in meal size. Second, that neuropeptide Y stimulates feeding behavior in monosodium glutamate-treated rats by increasing meal size and meal duration. Third, that monosodium glutamate-treated rats are more responsive to the injection of exogenous neuropeptide Y. These observations could be related to the monosodium glutamate-induced decrease in neuropeptide Y in the arcuate-paraventricular axis measured in the same animals. They imply an important functional role of this intrahypothalamic pathway in the central mechanisms controlling feeding behavior. In contrast to the obese hyperphagic Zucker rats, where a down-regulation at the neuropeptide Y receptor level is observed, the low concentrations found in the hypophagic monosodium glutamate-treated rats could up-regulate neuropeptide Y receptors, but this hypothesis needs experimental validation in binding studies.

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