ELSEVIER

European Journal of Pharmacology 386 (1999) 247–252

ejp

www.elsevier.nl/locate/ejphar

Neonatal monosodium glutamate treatment alters rat intestinal muscle reactivity to some agonists

Sergey N. Sukhanov [a], Iracema S. de Andrade [b], Miriam S. Dolnikoff [b], Alice T. Ferreira [a,*]

[a] *Department of Biophysics, Universidade Federal de São Paulo, Escola Paulista de Medicina, Rua Botucatu 862, Sao Paulo, SP CEP 04023-062, Brazil*
[b] *Department of Physiology, Universidade Federal de São Paulo, Escola Paulista de Medicina, Rua Botucatu 862, Sao Paulo, SP CEP 04023-062, Brazil*

Received 19 October 1999; accepted 22 October 1999

Abstract

The following study is an investigation of the changes in the contractile reactivity of visceral muscles in response to agonists and alterations in metabolic parameters after neonatal rat treatment with monosodium-L-glutamate. This treatment markedly sensitizes ileum and colon preparations to adenosine-5′-triphosphate (ATP) stimulation and also increases the colon activity to acetylcholine ($p < 0.05$). Response to bradykinin remained unchanged, while ileum activity to angiotensin II was characterized by a reduction in the maximal tension (E_{max}) and an increase in the EC_{50} ($p < 0.05$) value. The responses of nonintestinal muscle preparations from monosodium-glutamate-treated rats to both ATP and bradykinin did not show a significant difference when compared to the controls. This treatment diminished food intake, feces excretion and increased plasma insulin, nonesterified fatty acids and triglyceride concentrations ($p < 0.001$). These results suggest that the changes in intestinal muscle activity, in response to agonists, can be due to metabolic alterations as well as the monosodium glutamate action on enteric neurons and/or smooth muscle receptors.

Keywords: Monosodium glutamate; Smooth muscle; Adenosine-5′-triphosphate; Acetylcholine; Bradykinin; Angiotensin II

1. Introduction

The administration of monosodium L-glutamate to neonatal rats is known to induce anatomo-physiological disturbances which can be associated with several neuroendocrine, metabolic and behavioral abnormalities (Dolnikoff et al., 1988; Wong et al., 1997; Stricker-Krongrad et al., 1998). Marked neuronal loss was shown in the hypothalamic arcuate nucleus (Legradi et al., 1998), in the medial preoptic area (Desjardins et al., 1992) and in the circumventricular organs (Rogulja et al., 1987). Monosodium glutamate treatment alters cholinergic neurotransmission by decreasing acetyltransferase activity (Ortuno-Sahagun et al., 1997) and increasing the number of muscarinic receptors in the cerebral cortex (Beas-Zarate et al., 1994). On the other hand, when compared to normal rats, the immunoreactive somatostatin levels in the pancreas and antral region of the stomach doubled in the monosodium-glutamate-treated rats (De Paolo and Steger, 1985), while in the duodenum, jejunum and colon, no variation was observed. Actually, little is known about the physiological properties of the rat intestine following monosodium-glutamate-induced disturbances in the central nervous system.

This study was designed to examine if there is any change in the responses of different visceral muscle preparations to acetylcholine, adenosine-5′-triphosphate (ATP), bradykinin and angiotensin II following neonatal treatment with monosodium glutamate. Food intake, feces excretion and some metabolic parameters were also evaluated.

2. Materials and methods

2.1. Animals

Male newborn Wistar rats were used. Monosodium-L-glutamate was administered daily throughout the first 10 days of life (4 g/kg body weight) (Olney, 1969; Remke et al., 1988). The protocol for the use of the research animals was approved by the University Committee. The animals became obese after being treated with monosodium glutamate. Isotonic NaCl solution was injected in the control

* Corresponding author. Tel.: +55-11-576-4526; fax: +55-11-571-5780.
E-mail address: alice@biofis.epm.br (A.T. Ferreira)

0014-2999/99/$ - see front matter
PII: S0014-2999(99)00751-7

rats (1.25 g/kg body weight). The rats were kept in polystyrene boxes (lean and obese separate) under controlled light (12 hours light/dark phase [0700–1900 h]) and temperature conditions (22 ± 1°C), with free access to food and water. At the age of 75 days, control rats (298 ± 5 g, $n = 6$) and monosodium-glutamate-treated rats (295 ± 5 g, $n = 6$) were pair-weighed and placed in individual metabolic cages. The daily food intake and feces excretion were measured during the following days. After 15 days, the animals were decapitated and trunk blood was collected. Plasma was separated by centrifugation at $15{,}000 \times g$ and 4°C and kept at -20°C until the analysis for nonesterified fatty acids (Miles et al., 1983), triglycerides (Bucolo and David, 1973), cholesterol (Allain et al., 1974) and insulin. Glucose was assayed by a glucose oxidase kit (Boehring Mannheim, Mannheim, Germany) and insulin using the rat-specific radioimmunoassay kit (Novo Industry, Copenhagen, Denmark).

2.2. Procedures

The distal ileum and distal colon were dissected, quickly freed of any connective tissue and placed in Tyrode solution (137 mM NaCl/2.7 mM KCl/12 mM $NaHCO_3$/0.36 mM NaH_2PO_4/0.53 mM $MgCl_2$/1.36 mM $CaCl_2$/5.5 mM glucose). For experiments with vas deferens, the epididymal and prostatic portions were tested separately.

The preparations from control and monosodium-glutamate-treated groups of rats were mounted isometrically in a 5-ml organ bath at 37°C filled with Tyrode solution and continuously bubbled with air. The preparations were equilibrated in this solution for 45 min with buffer changes every 15 min, the resting tension being 1 g. At the beginning of each experiment, the response to KCl (40 mM) was tested as a control. Preparations from the control and monosodium-glutamate-treated rats were challenged with single concentrations of various drugs. The response to agonists was recorded during 1.5 min and expressed as a percentage of a 40-mM KCl-induced contraction for the establishment of dose–response curves. A short (3.5 min) interval with repeated renewals of the bathing solutions was allowed to elapse between one challenge and the next. For angiotensin II, a 20-min interval between applications was used. Only this interval provided reproducible responses and was thus selected to avoid tachyphylaxis (Paiva et al., 1974).

2.3. Drugs

The following drugs were used: monosodium-L-glutamate, ATP (disodium salt) from Sigma, St. Louis, MO, USA and acetylcholine chloride from Merck Industries, Quimicas, RJ, Brazil. The bradykinin and angiotensin II used in the present study were synthetized in our laboratory as previously described (Paiva et al., 1974). Stock solutions of all drugs were prepared in 0.9% NaCl. Salts for the Tyrode solution were of analytical grade and were obtained from Merck Industries, Quimicas, RJ, Brazil.

2.4. Data analysis

The EC_{50} values were calculated from the dose–response curves and expressed as $\log EC_{50}$ (Flemming et al., 1972) for the statistical analysis. Values of maximum tension developed under agonist stimulation (E_{max}) were obtained from the same curves and shown in the text as a percentage of a 40-mM KCl-induced contraction. Nonlinear regression fit (sigmoidal dose–response, variable slope, without weight) was carried out using the Graf Pad Prism v.2.01 computer program (Graf Pad Software, San Diego, CA, USA). All the values found in the text, figures and Table 1 are expressed as means $\pm$ S.E.M., and n is the number of observations. Significance was tested in all experiments according to the nonparametric two-tailed Mann–Whitney U-test ($p < 0.05$) or the Student's unpaired t-test ($p < 0.001$) as required.

3. Results

In this study, the animals were pair-weighed to avoid body weight influence. Therefore, the control and monosodium glutamate groups were found to have similar body weight (296.5 ± 5 g). As shown in Table 1, total food intake and feces excretion diminished significantly ($p < 0.001$, t-test, $n = 6$) in rats following the treatment with monosodium glutamate. Plasma insulin, nonesterified fatty acids and triglyceride concentrations were higher in monosodium-glutamate-treated rats vs. control rats ($p < 0.001$) while there were no differences found in plasma glucose and cholesterol concentrations between both groups of animals.

Intestinal muscle preparations from monosodium-glutamate-treated and control rats showed a dose-dependent response to ATP, acetylcholine, bradykinin and angiotensin II (Figs. 1A–D and 2A–D).

Table 1
Some metabolic parameters of monosodium-glutamate-treated and control rats
Values presented are means $\pm$ S.E.M.; $n = 6$ rats/group.

Parameters	Control	Monosodium glutamate
Body weight (g)	298 ± 5	295 ± 5
Food intake (g/100 g/day)	12.07 ± 0.2	8.77 ± 0.2*
Feces excretion (g/100 g/day)	1.98 ± 0.07	1.41 ± 0.06*
Glucose (mM/l)	7.6 ± 0.2	7.8 ± 0.3
Insulin (nM/l)	389.8 ± 20.6	531.0 ± 25.0*
Nonesterified fatty acids (mM)	284.0 ± 32.0	448.0 ± 26.0*
Triglycerides ($g \times 10^{-3}$/dl)	87.5 ± 4.1	134.0 ± 11.8*
Cholesterol ($g \times 10^{-3}$/dl)	87.8 ± 2.9	90.9 ± 3.7

* Significantly different vs. control rats ($p < 0.001$) (Student's t-test).

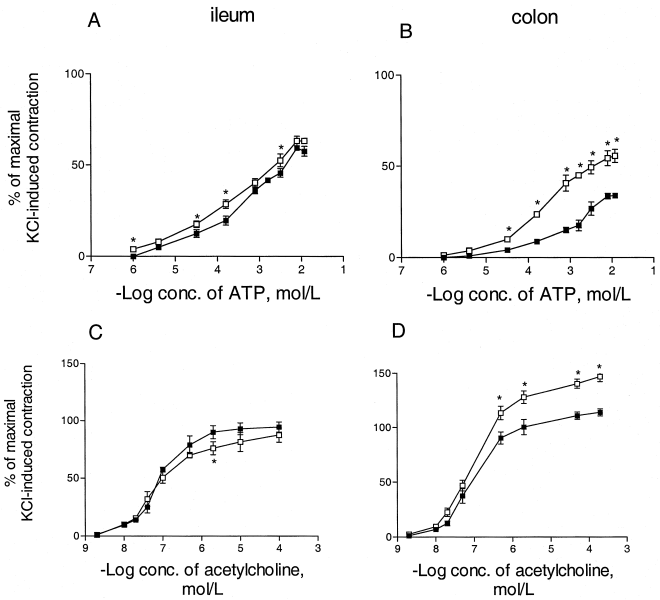

Fig. 1. Dose–response curves of ATP (A, B) and acetylcholine (C, D) for ileum (A, C) and colon (B, D) muscle preparations from control (■) and monosodium-glutamate-treated (□) rats. Agonists were applied in a near-cumulative manner during 1.5 and 3.5 min interval between application with intensive washing. ATP — adenosine-5′-triphosphate. The contractile responses to agonists were expressed as percentage of the maximum response elicited by 40 mM KCl at the beginning of experiment. Each symbol and vertical bar represents the means ± S.E.M. of 9–12 experiments. $^{*}p < 0.05$ (Mann–Whitney *U*-test). For details, see text.

Dose–response curves of ATP and acetylcholine for ileum and colon preparations from monosodium-glutamate-treated rats and control rats are shown in Fig. 1A,B where it can be seen that ATP curves for both ileum and colon obtained for monosodium-glutamate-treated rat preparations shift to the left as compared to controls. A

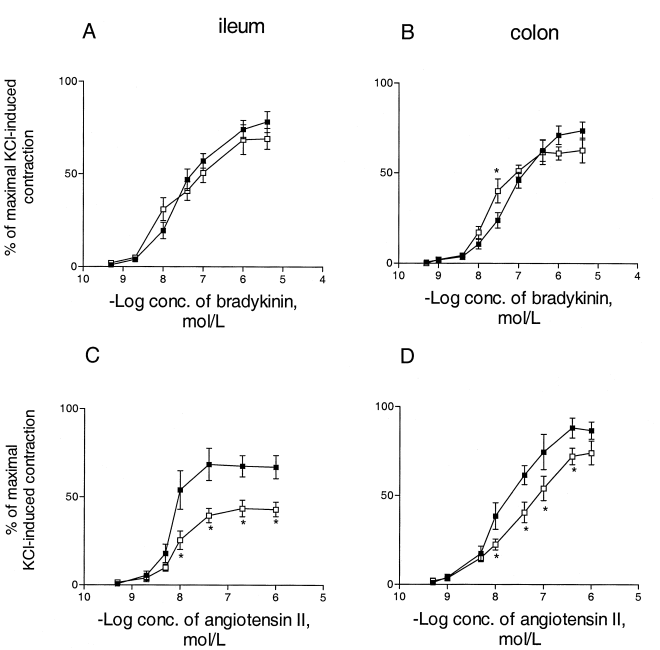

Fig. 2. Dose–response curves of bradykinin (A, B) and angiotensin II (C, D) for ileum (A, C) and colon (B, D) muscle preparations from control (■) and monosodium-glutamate-treated (□) rats. In experiments with bradykinin, a near-cumulative protocol (1.5 min for drug application; 3.5 min between injections with intensive washing) was used. The same protocol but with longer intervals (20 min) between applications was used for angiotensin II to avoid tachyphylaxis. The contractile responses to agonists were expressed as percentage of the maximum response elicited by 40 mM KCl at the beginning of experiment. Each symbol and vertical bar represent the means ± S.E.M. of 6–10 experiments. $^{*}p < 0.05$ (Mann–Whitney *U*-test). For details, see text.

significant decrease ($p < 0.05$) in the EC_{50} value of this agonist regarding ileum (from $0.54 \pm 0.13 \times 10^{-3}$ M, $n = 9$ in control to 0.23 ± 0.0510^{-3} M, $n = 10$ in monosodium-glutamate-treated rats) and colon preparations (from $0.84 \pm 0.33 \times 10^{-3}$ M, $n = 10$ to $0.28 \pm 0.08 \times 10^{-3}$ M, $n = 11$) was observed. The maximum tension developed by the ileum preparations from monosodium-glutamate-treated rats under ATP stimulation did not change as compared to controls, while the E_{max} of ATP-induced responses of the colon was significantly ($p < 0.05$) potentiated in monosodium-glutamate-treated rats ($53 \pm 5\%$, $n = 11$) with respect to the controls ($30 \pm 3\%$, $n = 10$).

Acetylcholine produced no difference in EC_{50} and E_{max} values in the ileum between monosodium-glutamate-treated rats and control rats (Fig. 1C,D). However, EC_{50} of acetylcholine in colon preparations decreased markedly from $1.2 \pm 0.3 \times 10^{-7}$ M, $n = 10$ in controls to $0.5 \pm 0.1 \times 10^{-7}$ M, $n = 10$ in monosodium-glutamate-treated rats and E_{max} was increased from $120 \pm 11\%$, $n = 10$ to $143 \pm 14\%$, $n = 10$, respectively ($p < 0.05$).

Dose–response curves of bradykinin and angiotensin II for intestinal muscle preparations from control and monosodium-glutamate-treated rats are shown in Fig. 2. This treatment induced a nonsignificant increase in EC_{50} of bradykinin in ileum (from $43 \pm 9 \times 10^{-9}$ M, $n = 9$ to $60 \pm 8 \times 10^{-9}$ M, $n = 7$) as well as in colon preparations (from $47 \pm 7 \times 10^{-9}$ M, $n = 7$ to $62 \pm 9 \times 10^{-9}$ M, $n = 10$). Maximal responses to bradykinin also showed a nonsignificant decrease in the ileum from monosodium-glutamate-treated rats (from $78 \pm 6\%$, $n = 8$ to $69 \pm 6\%$, $n = 6$) and colon (from $77 \pm 9\%$, $n = 6$ to $61 \pm 7\%$, $n = 9$) as compared to controls (Fig. 2A,B).

The dose–response curves of angiotensin II for ileum and colon preparations from monosodium-glutamate-treated rats were shifted to the right (Fig. 2C,D). The EC_{50} of angiotensin II increased significantly ($p < 0.05$) in the ileum (from $7.0 \pm 2.1 \times 10^{-9}$ M, $n = 9$ to $17.0 \pm 1.8 \times 10^{-9}$ M, $n = 10$) and was nonsignificant in the colon (from $12.2 \pm 3.7 \times 10^{-9}$ M, $n = 9$ to $17.8 \pm 5.5 \times 10^{-9}$ M, $n = 10$) preparations. The maximum tension developed in the ileum preparation from monosodium-glutamate-treated rats under angiotensin II stimulation ($41 \pm 8\%$, $n = 10$) was smaller than that of the controls ($66 \pm 10\%$, $n = 9$).

In order to compare the effect of neonatal monosodium glutamate treatment on intestinal and nonintestinal muscle preparations in response to ATP and bradykinin, we carried out experiments with prostatic and epididymal portions of the rat vas deferens. Bisected nonintestinal muscle preparations from monosodium-glutamate-treated and control rats responded to agonists in a dose-dependent manner. Monosodium glutamate treatment induced no differences in the EC_{50} value of ATP for prostatic ($4.2 \pm 0.6 \times 10^{-3}$ M, $n = 7$ from control and $5.0 \pm 1.1 \times 10^{-3}$ M, $n = 7$ from monosodium-glutamate-treated rats) and epididymal ($6.9 \pm 1.2 \times 10^{-3}$ M, $n = 6$ and $6.7 \pm 0.6 \times 10^{-3}$ M, $n = 6$, respectively) preparations. Bradykinin also did not show marked changes in responses of vas deferens preparations from monosodium-glutamate-treated rats as compared to controls. Actually, the EC_{50} values of this agonist in monosodium glutamate vas deferens prostatic ($136 \pm 29 \times 10^{-9}$ M, $n = 7$) and epididymal ($100 \pm 16 \times 10^{-9}$ M, $n = 6$) portions were approximately equal to the controls (prostatic — $97 \pm 15 \times 10^{-9}$ M, $n = 6$; epididymal — $113 \pm 16 \times 10^{-9}$ M, $n = 7$).

4. Discussion

Neonatal monosodium glutamate treatment has been reported to cause hypothalamic lesions (Legradi et al., 1998), which affect food intake and alter the feeding patterns (Wong et al., 1997; Stricker-Krongrad et al., 1998) in rodents. The disturbances in the rat intestinal tract may be mediated by specific alterations in hormone secretion including the gastroenteropancreatic system (De Paolo and Steger, 1985) and/or the changes in intestinal muscle reactivity.

Food intake is diminished in monosodium-glutamate-treated animals as found by Stricker-Krongrad et al. (1998). For the first time, we now show that feces excretion was reduced significantly in the monosodium-glutamate-treated rats when compared to the controls. The hypothalamic arcuate nucleus destruction promoted by the neonatal monosodium glutamate treatment decreases the hypothalamic neuropeptide Y content (Nemeroff et al., 1978) that stimulates food intake (Taylor et al., 1990). Thus, the reduced food intake and feces excretion can be explained by the lack of neuropeptide Y stimulation. However, the relationship between the amount of feces excreted and the amount of food intake cannot be ruled out.

Plasma glucose and cholesterol concentrations in monosodium-glutamate-treated rats were similar to that of controls; however, plasma concentrations of nonesterified fatty acids, insulin and triglycerides increased. It was shown that high insulin in vitro can modify both the motility responses of rat intestine and the neuropeptide release from these segments (Allescher et al., 1991). Therefore, present data suggest that hyperinsulinemia in monosodium-glutamate-treated rats could be a possible mechanism which mediates the specific intestinal disturbances.

To study other pathways for the effect of monosodium glutamate treatment on intestinal muscles, we tested the ileum and colon responsiveness to ATP and acetylcholine which are transmitter substances in these tissues (Burnstock et al., 1970) and also evaluated muscle responses to the main gastrointestinal myotropic peptides — bradykinin and angiotensin II.

We found that monosodium glutamate treatment sensitizes ileum and colon preparations to ATP by a 2.4–3.0-fold

significant decrease in EC_{50} ($p < 0.05$) and 1.2– 1.6-fold significant increase in E_{max} ($p < 0.05$), respectively, and colon preparations to acetylcholine as well. However, no effects were observed with ATP stimulation in vas deferens preparations from monosodium-glutamate-treated rats as compared to controls. Similar to our data, Wong et al. (1988), using other nonintestinal muscle preparations, have shown no alterations in response to acetylcholine.

Monosodium-glutamate-treatment-induced facilitation in response to agonists can be mediated by appropriate changes in receptor number and/or binding affinity of muscle cells to agonist molecules. Taking into account the participation of these substances in neuromuscular transmission, we reasoned that membrane alterations are playing a compensating, secondary role in response to hormonal disturbances in the intestine and, probably, in the decrease of the neurotransmitters released from the intramural nervous system. A significant decrease in choline acetyltransferase activity and an increase in the number of muscarinic receptors was observed in the brain of monosodium-glutamate-treated rats also as a result of neuronal loss in the arcuate nucleus (Beas-Zarate et al., 1994; Ortuno-Sahagun et al., 1997). Therefore, it is quite reasonable to conclude that the neonatal monosodium glutamate treatment induces changes in the sensitivity of the ileum and colon smooth muscles to several neurotransmitters and hormones. Since we found no alterations in reactivity of vas deferens to agonists, it is suggested that the mentioned compensating events have a specific intestinal localization.

It is important to note that this treatment sensitizes the colon preparations to ATP and acetylcholine more when compared with the effect on the ileum. These data may be explained in terms of the difference in cholinergic and purinergic innervation of the rat ileum and colon (Driel and Drukker, 1973; Boeckxstaens et al., 1990) and/or by the difference in origin of the sympathetic nerves supplying this region (Belai et al., 1991).

The monosodium glutamate treatment of rats did not alter the characteristics of intestinal responses to bradykinin and induced the shift to the right of the dose–response curve for angiotensin II in the ileum. Bradykinin and angiotensin II evoked responses in the rat intestinal smooth muscle via intracellular pathways (Zagorodnyuk et al., 1998) mediated by different receptor types (Schinke et al., 1991; Calixto, 1995). Therefore, our data may be due to some changes in cell membrane properties (such as decrease in the number and/or affinity of angiotensin II receptors) rather than to alterations in the contractile apparatus of these muscles. No analysis of this machinery was done in the present study, although the gross structure of the monosodium-glutamate-treated colon and ileum was similar to that of the control. Since it was shown that the high plasma insulin level attenuates angiotensin-II-stimulated vascular contraction (Saito et al., 1993), present data also suggest a direct effect of high insulin on the responsiveness of intestinal smooth muscles to angiotensin II.

5. Conclusion

Neonatal monosodium glutamate treatment provokes hormonal alterations and specific intestinal changes in smooth muscle reactivity to agonists. We suggest that gastrointestinal disturbances are mediated by the changes in the intramural nervous system (neuronal loss or decrease in neurotransmitter release) and/or specific alterations in smooth muscle receptor level.

Acknowledgements

This study was supported by a grant from the ''Fundação de Amparo a Pesquisa do Estado de São Paulo, Brasil, Proc. N 1997/13183-1''. We thank Dr. Irina A. Vladimirova (Department of Pharmacology, Escola Paulista de Medicina) for the helpful discussion of the present data and Dr. Hanna Rothschild for critical comments on the manuscript.

References

Allain, C.C., Poon, L.S., Chan, C.S., Richmond, W., Fu, P.C., 1974. Enzymatic determination of total serum cholesterol. Clin. Chem. 20, 470–475.

Allescher, H.D., Willis, S., Schusdziarra, V., Classen, M., 1991. Modulatory effect of insulin on rat small intestinal motility and peptide release in vitro. Digestion 48, 192–201.

Beas-Zarate, C., Schliebs, R., Ortuno-Sahagun, D., Morales-Villagran, Feria-Velasco, A., 1994. Effect of systemic monosodium L-glutamate on muscarinic cholinergic receptors in selected rat brain regions during development. Arch. Med. Res. 25, 341–346.

Belai, A., Lincoln, D., Milner, P., Burnstock, G., 1991. Differential effect of streptozotocin-induced diabetes on the innervation of the ileum and distal colon. Gastroenterology 100, 1024–1032.

Boeckxstaens, G.E., Pelckmans, P.A., Rampart, M., Verbeuren, T.J., Herman, A.G., Van Maercke, Y.M., 1990. Nonadrenergic noncholinergic mechanisms in the ileocolonic junction. Arch. Int. Pharmacodyn. Ther. 303, 270–281.

Bucolo, G., David, H., 1973. Quantitative determination of serum triglycerides by the use of enzymes. Clin. Chem. 19, 476–482.

Burnstock, G., Campbell, G., Satchell, D., Smythe, A., 1970. Evidence that adenosine tri-phosphate or a related nucleotide is the transmitter substance released by non-adrenergic inhibitory nerves in the gut. Br. J. Pharmacol. 40, 668–688.

Calixto, J.B., 1995. Multiple mechanisms of bradykinin-induced contraction in rat and guinea pig smooth muscles in vitro. Eur. J. Pharmacol. 281, 279–288.

De Paolo, L.V., Steger, R.W., 1985. Alterations in immunoreactive somatostatin levels in hypothalamic and gastroenteropancreatic tissue as a consequence of neonatal treatment with monosodium glutamate. Proc. Soc. Exp. Biol. Med. 178, 419–425.

Desjardins, G.C., Brawer, J.R., Beaudet, A., 1992. Monosodium glutamate-induced reductions in hypothalamic beta-endorphin content result in mu-opioid receptor upregulation in the medial preoptic area. Neuroendocrinology 56, 378–384.

Dolnikoff, M.S., Kater, C.E., Egami, M., de Andrade, I.S., Marmo, M.R., 1988. Neonatal treatment with monosodium glutamate increases plasma corticosterone in the rat. Neuroendocrinology 48, 645–649.

Driel, C., Drukker, J., 1973. A contribution to the study of the architec-

ture of the autonomic nervous system of the digestive tract of the rat. J. Neural Transm. 34, 301–320.

Flemming, W.W., Westfall, D.P., de Lande, I.S., Jellet, L.B., 1972. Log-normal distribution of equieffective doses of norepinephrine and acetylcholine in several tissues. J. Pharmacol. Exp. Ther. 181, 339–345.

Legradi, G., Emerson, C.H., Ahima, R.S., Rand, W.M., Flier, J.S., Lechan, R.M., 1998. Arcuate nucleus ablation prevents fasting-induced suppression of ProTRH mRNA in the hypothalamic paraventricular nucleus. Neuroendocrinology 68, 89–97.

Miles, J., Glasscock, R., Aikens, J., Gerich, J., Haymond, A., 1983. Microfluorometric method for the determination of free fatty acids in plasma. J. Lipid Res. 24, 96–99.

Nemeroff, C.B., Lipton, M.A., Kizer, J.S., 1978. Models of neuroendocrine regulation: use of monosodium glutamate as an investigational tool. Dev. Neurosci. 1, 102–109.

Olney, J.W., 1969. Brain lesions, obesity, and other disturbances in mice treated with monosodium glutamate. Science 164, 719–721.

Ortuno-Sahagun, D., Beas-Zarate, C., Adame-Gonzalez, G., Feria-Velasco, A., 1997. Effect of L-glutamate on cholinergic neurotransmission in various brain regions and during the development of rats, when administered perinatally. Neurochem. Int. 31, 683–692.

Paiva, T.B., Juliano, L., Nouailhetas, V.L., Paiva, A.C., 1974. The effect of pH on tachyphylaxis to angiotensin peptides in the isolated guinea pig ileum and rat uterus. Eur. J. Pharmacol. 25, 191–196.

Remke, H., Wilsdorf, A., Muller, F., 1988. Development of hypothalamic obesity in growing rats. Exp. Pathol. 33, 223–232.

Rogulja, I., Harding, J.W., Ritter, S., 1987. Reduction of ^{125}I-angiotensin II binding sites in rat brain following monosodium glutamate treatment. Brain Res. 419, 333–335.

Saito, F., Hori, M.T., Fittingoff, M., Hino, T., Tuck, M.L., 1993. Insulin attenuates agonist-mediated calcium mobilization in cultured rat vascular smooth muscle cells. J. Clin. Invest. 92, 1161–1167.

Schinke, M., Doods, H.N., Ganten, D., Wienen, W., Entzeroth, M., 1991. Characterization of rat intestinal angiotensin II receptors. Eur. J. Pharmacol. 204, 165–170.

Stricker-Krongrad, A., Burlet, C., Beck, B., 1998. Behavioral deficits in monosodium glutamate rats: specific changes in the structure of feeding behavior. Life Sci. 62, 2127–2132.

Taylor, I.L., Mannon, P.J., Heintz, G.G., Kaiser, L.M., Nguyen, T.D., 1990. Comparison of the neuropeptide Y receptor in the rat brain and intestine. Ann. N.Y. Acad. Sci. 611, 48–57.

Wong, C.C., Chu, J.Y., Chiu, K.W., 1988. Effect of neonatal monosodium-L-glutamate treatment on the response of isolated uterine preparation to various uterine contraction stimulators. Gen. Pharmacol. 19, 673–677.

Wong, P.T., Neo, L.H., Teo, W.L., Feng, H., Xue, Y.D., Loke, W.H., 1997. Deficits in water escape performance and alterations in hippocampal cholinergic mechanisms associated with neonatal monosodium glutamate treatment in mice. Pharmacol. Biochem. Behav. 57, 383–388.

Zagorodnyuk, V., Santicioli, P., Maggi, C.A., 1998. Evidence for the involvement of multiple mechanisms in the excitatory action of bradykinin in the circular muscle of guinea-pig colon. Naunyn-Schmiedeberg's Arch. Pharmacol. 357, 197–204.