

Effect of Adrenalectomy on the Activity of Small Intestine Enzymes in Monosodium Glutamate Obese Rats

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Summary

It is well known that adrenalectomy (ADX) reverses the eating and energy balance disturbances in a variety of models of obesity associated with elevated food intake. We have previously demonstrated enhanced functional activity in the small intestine of neonatally monosodium glutamate-treated (MSG) obese rats despite the absence of overeating and we concluded that these changes might also contribute to the development of MSG obesity. The objective of the present experiments was to investigate whether ADX would affect the small intestinal functions and whether their changes would counteract attenuation or prevention of obesity development in MSG rats. Therefore the investigation was carried out in MSG-obese Wistar male rats and untreated intact rats adrenalectomized on day 40, as well as in lean littermates of MSG rats and intact rats subjected to Sham-ADX surgery. All animals had free access to a standard pellet diet after weaning. At the age of 80 days, body mass, body fat content and food consumption as well as changes of the brush-border-bound duodenal and jejunal alkaline phosphatase (AP), the dipeptidyl(amino)peptidase IV (DPP IV) and maltase activity were measured. During the postoperative period, ADX resulted in a significant decrease of mass gain in both MSG and control rats ($P < 0.05$). ADX fully prevented the development of obesity in MSG rats (significantly decreased epididymal+retroperitoneal fat pad mass, $P < 0.05$) and increased mean daily food intake ($P < 0.001$). These effects were only minimal in the ADX controls suggesting that enhanced adrenal secretion is involved in the expression of MSG obesity and its complications. The AP activity in obese MSG rats was increased by about 21 % ($P < 0.01$) in both intestinal segments when compared to the lean controls, whereas no parallel variations in the activities of DPP IV and maltase were observed in the intestinal parts mentioned. In MSG rats, ADX significantly reduced the AP activity in the duodenum and jejunum ($P < 0.01$). A similar tendency was also seen in the DPP IV activity of adrenalectomized MSG rats as well as in lean control rats. Nevertheless, no significant effect of adrenal withdrawal on maltase activity was found. These results indicate that the decrease of enzyme activities in the small intestine and the different effectiveness of nutrient absorption might be a significant factor preventing the development of excess adiposity in glutamate-treated rats. This information contributes to a better understanding of the importance of small intestinal function for the development of obesity and its maintenance in later life.

Key words

Neonatal MSG treatment • Adrenalectomy • Small intestine enzymes • Obesity

Introduction

It has been well established that glucocorticoids affect development and maintenance of obesity by control of the feeding behavior, body fat and body mass. It has been found that several forms of obesity are closely related to hypercorticism (Cunningham *et al.* 1986, Guillaume-Gentil *et al.* 1990, Mantha *et al.* 1999, Macho *et al.* 1999a,b, Guimaraes *et al.* 2002). This role of corticoids was also supported by experiments using either adrenalectomized (ADX) obese rats in which adrenal withdrawal normalized their previous hyperphagia and led to a reduction of higher body fat content and overweight or by experiments in which changes of these variables in ADX animals were reversed after glucocorticoid replacement (Castonguay *et al.* 1986, Freedman *et al.* 1986, Romsos *et al.* 1987, Mantha *et al.* 1999). As compared to obese rats, a similar effect of corticoids on body fat has also been observed in non-obese animals; hypercorticism (implanted corticosterone pellets) resulted in enhanced adiposity (McIntosh *et al.* 1999), while adrenal insufficiency decreased body fat stores (Trocki *et al.* 1995, Edens *et al.* 1999, Mantha and Deshaies 2000, Bhatnagar *et al.* 2000). In addition, in lean rats ADX decreased the caloric efficiency and food intake (Edens *et al.* 1999). These findings suggest that glucocorticoid levels may exert a modulational effect on the digestive and absorptive function of the small intestine. In agreement with this assumption, ADX in non-obese rats resulted in significant morphological disorganization and in decreased levels of some brush-border-bound small intestinal digestive enzymes (Majumdar 1981, Foligne *et al.* 2001), whereas a subsequent corticoid replacement abolished the effect of ADX and restored enzymes activity (Majumdar 1981).

In contrast to other forms of obesity which are associated with hyperphagia, the obesity model of neonatally monosodium glutamate (MSG)-treated rodents is of a special interest since increased plasma levels of corticosterone (Tokuyama and Himms-Hagen 1989, Macho *et al.* 1999a,b, Guimaraes *et al.* 2002) as well as increased lipogenesis and reduced lipolysis in adipose tissues (Dolnikoff *et al.* 2001, Macho *et al.* 2000) occurred despite their normophagia or even hypophagia (Tokuyama and Himms-Hagen 1989, Zhang *et al.* 1994, Morris *et al.* 1998, Mistlberger and Antle 1999, Martinková *et al.* 2000). Moreover, adrenalectomy prevented the development of obesity in MSG-treated mice without anorexia (Tokuyama and Himms-Hagen

1989). As compared to lean controls, mature MSG rats displayed an enhanced capacity to spare body fat stores during fasting, more marked adaptational increase of food intake and more rapid body fat restoration after refeeding (Raček *et al.* 2001). In contrast to the non-obese controls a permanently higher alkaline phosphatase activity has also been demonstrated in the brush-border of the duodenal enterocytes of MSG-rats during *ad libitum* feeding (Možeš *et al.* 2000, Martinková *et al.* 2000), after food restriction and fasting, as well as after refeeding (Raček *et al.* 2001). From this point of view, the enzymatic changes in the small intestine seem to be related to the components of regulatory mechanisms maintaining their obesity at critical values. Another question is whether these permanent alterations might also be attributed to the increased adrenal secretion and whether ADX might replace the function of this tissue. Such data would be useful to gain further insight into the role of the intestinal epithelium on the development of MSG-induced obesity.

The activity of brush-border-bound alkaline phosphatase, dipeptidyl(amino)peptidase IV, and maltase was therefore studied in duodenal and jejunal enterocytes of 80-day-old rats neonatally treated with MSG or saline (controls) and subjected to ADX or sham-surgery on day 40. The measured parameters also included food intake, body mass and epididymal + retroperitoneal fat pad mass as indicators of MSG-obesity.

Methods

Subjects

Wistar rat mothers and their offsprings (eight pups per nest) were housed from birth to weaning in Plexiglass cages in a temperature-controlled environment of 22 ± 2 °C and at 12L:12D regime (light on 06:00-18:00 h) with free access to a standard laboratory diet (Velaz/Altromin 1520 DOS 2b, Velaz, Prague) and tap water.

Half of the pups in each nest containing males and females received a s. c. injection of monosodium salt of L-glutamic acid (Sigma, St. Louis, MO) 2 mg/g b.m. (MSG groups) or an equivalent volume of 1.25 % saline solutions (control groups) daily for 4 days after birth. MSG was dissolved in distilled water and the drug concentration adjusted so that each pup received 50 µl of solution/g b.m. After weaning (on day 30) pairs of male rats were housed in Plexiglass cages under the same conditions (diet, water, temperature, light-dark regime) as

before weaning.

Surgery

On day 40 the rats were subjected to bilateral ADX or sham-surgery (a whole surgical procedure except for removal of the adrenal gland) through dorsal incisions under ketamine/xylazine anesthesia (100/20 mg/kg). After the surgery, ADX rats were maintained by means of an 0.9 % NaCl solution, the sham-operated controls had access to tap water and both groups of animals were fed the same laboratory diet *ad libitum* as before the operation.

Tissue sampling

On day 80, the animals were killed by decapitation after an overnight fast between 08:00 and 09:00 h. Small (0.5 cm) segments of the proximal duodenum and middle part of the jejunum were immediately removed, the lumen rinsed with distilled water, and frozen in hexane (−70 °C). The segments of the frozen tissue were cut (8 µm) in a cryostat at −25 °C and the tissue slices were transferred to glass slides and air-dried.

Enzyme assay

Demonstration of alkaline phosphatase activity was performed by using the modified simultaneous azo-coupling method (Lojda *et al.* 1979) with naphthol AS-BI phosphate (Sigma, Deisenhofen, Germany) and Fast Blue B (Sigma), dipeptidyl(amino)peptidase IV by using the simultaneous azo-coupling method (Lojda 1977) with Gly-pro-4-methoxy-β-naphthylamide (Sigma) and Fast Blue B (Sigma); maltase was demonstrated by the azo-coupling method (Gossrau 1976) with 2-naphthyl-α-glucopyranoside (Sigma) and hexazonium-p-rosaniline (Sigma). Enzyme activity was cytophotometrically analyzed with a Vickers M85a microdensitometer. The measurements were performed by using an x40 objective, an effective scanning area of 28.3 µm² and a scanning spot of 0.5 µm. The integrated absorbance was measured at a wavelength of 480 nm for alkaline phosphatase and 540 nm for DPP IV and maltase. The mask was set over in at least 30 brush border areas along the villus length in five sections of the duodenum and the jejunum. AP activity was calculated as the absorbance values recorded by the instrument /min/µm³ brush border ± S.E.M. and these mean values were referred to one animal.

Table 1. Body mass, body fat and food intake parameters of control and neonatally MSG-treated sham-operated or adrenalectomized (ADX) rats

	Control		MSG	
	Sham	ADX	Sham	ADX
Body mass (g)	420.0±10.6	381.6±7.5	369.1±10.5 ^a	333.1±14.7 ^a
Weight gain (g) from day 40-80	261.1±11.4	221.8±11.1*	222.1±6.9 ^a	182.0±9.8* ^a
Fat pads mass (g)	8.9±1.0	7.6±1.1	10.9±0.9	7.0±0.7
Fat pad mass (g/100g b.w.)	2.1±0.2	2.0±0.3	3.1±0.3 ^a	2.1±0.2*
Food intake (g/day)	29.6±1.1	26.7±1.0	23.9±1.2 ^b	28.9±1.1*
Food intake (g/100g b.w.)	7.06±0.12	7.00±0.16	6.45±0.24 ^a	8.65±0.17*** ^c

Values are means ± S.E.M. of 8 rats in each group. Data were obtained on day 80 (40 days after surgery). Fat represents epididymal plus retroperitoneal pads. The 24-h food intake presents means of five measurements (day 74-79) for each rat. Significantly different from sham group: * P<0.05, ***P<0.001. Significant differences between similarly treated MSG and control groups: ^a P<0.05, ^b P<0.01, ^c P<0.001 (comparisons are based upon one way ANOVA with Tukey's test).

From day 74-79 the MSG-treated and control rats were individually housed and food intake was measured at 24-h intervals as the difference between the amount offered and that remaining in the cups. Epididymal and retroperitoneal fat was removed after killing the animals and the wet mass of the whole pad was obtained.

Statistical analysis

Statistical evaluation of the results was carried out by one-way analysis of variance (ANOVA) followed by Tukey's test to compare the differences between the groups. The two-way ANOVA was used to test the effect of MSG treatment, surgery and their interaction.

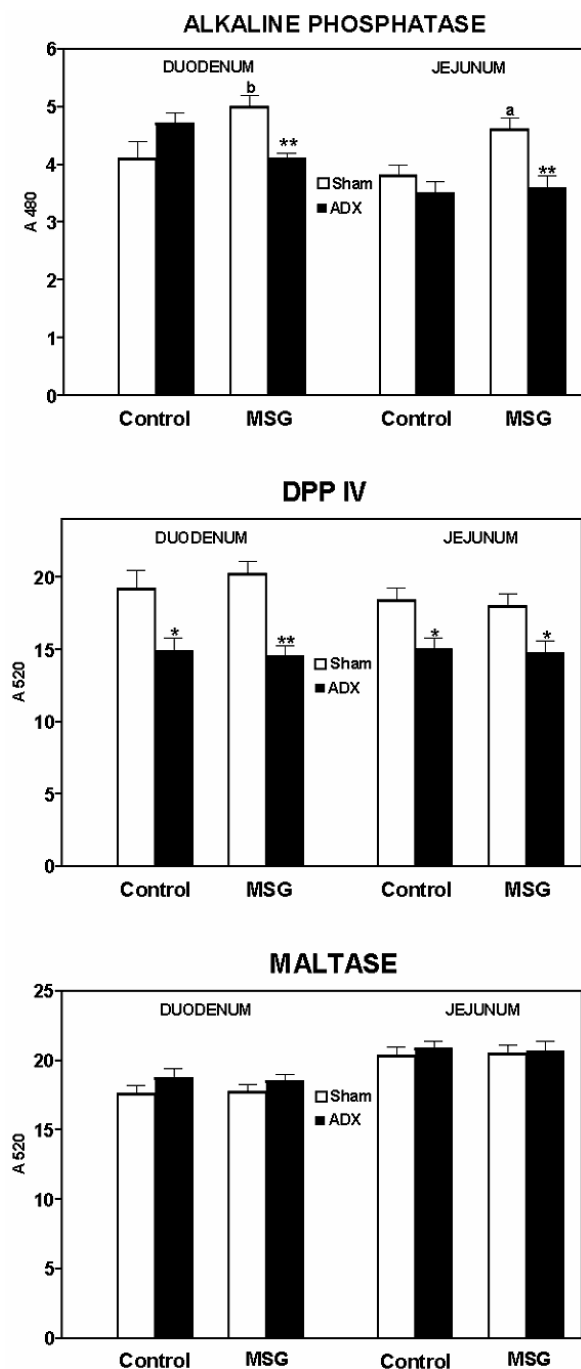


Fig. 1. Duodenal and jejunal activity of alkaline phosphatase, DPP IV (dipeptidyl-amino-peptidase IV) and maltase in control and neonatally MSG-treated rats subjected to sham-operation or adrenalectomy (ADX). Columns denote the mean activity \pm S.E.M. calculated as the absorbance values recorded by the instrument /min/ μm^3 brush border. Significantly different from the sham group: * $P < 0.05$, ** $P < 0.01$. Significant differences between similarly treated MSG and control groups: a $P < 0.05$, b $P < 0.01$.

Results

The mean body mass at the beginning of the experiment was 149.2 ± 7.0 (n=16) for neonatally MSG

treated rats and 159.2 ± 6.0 for saline-treated littermates (n=16). From day 40 until day 80 of age the MSG rats became significantly lighter and displayed lower mass gain ($p < 0.05$) than the controls. Adrenalectomy (ADX) resulted in a pronounced decrease of body mass in both MSG and control rats their mass gain being lower by about 15 % and 18 % ($p < 0.05$), respectively, in contrast to the sham-treated animals (Table 1).

Epididymal+retroperitoneal fat pad mass was significantly higher in sham-operated MSG rats, as related to sham-operated controls (saline-treated as neonates). ADX resulted in a significantly decreased mass of the fat tissue in MSG rats, i.e. their fat tissues expressed in terms of body mass was the same as in postnatally saline-treated sham-operated animals on day 80. On the other hand, mass of adipose tissues in ADX controls remained unaltered (Table 1). Two-way ANOVA revealed that there were specific changes in fat mass (interaction $F=5.019$, $P < 0.05$ for fat mass in grams and interaction $F=6.562$, $P < 0.02$ for the fat mass in g/b.m.). In this way a significant effect of MSG treatment ($F=10.72$, $P < 0.03$) and an effect of ADX ($F=7.614$, $P < 0.01$) on the fat pad mass (% of b.m.) was observed.

Animals receiving MSG as neonates consumed about 20 % less food ($P < 0.01$) than their saline-treated littermates. Whereas the mean daily food intake was relatively slightly decreased after ADX in the controls, food consumption was significantly elevated (about 20 %; $P < 0.05$) in ADX-MSG rats as related to that observed in the sham-MSG controls (Table 1). Two-way ANOVA showed specific changes of food intake expressed either in g/day or in g/100 g b.m. (interaction $F=13.41$, $P < 0.001$ and $F=49.43$, $P < 0.0001$, respectively). Furthermore, a significant effect of MSG-treatment ($F=9.48$, $P < 0.05$) and effect of ADX ($F=41.25$, $P < 0.0001$) on food intake expressed in % of body mass was also found.

Brush-border-bound alkaline phosphatase (AP), dipeptidyl(amino)peptidase IV (DPP IV) and maltase activity of duodenal and jejunal enterocytes in 80-day-old rats are shown in Figure 1. Associated with postnatal MSG-treatment the obese rats displayed a significantly increased AP activity in both intestinal segments as compared to the lean controls, whereas no parallel variations in the activities of DPP IV and maltase have been observed in the intestinal parts mentioned. The differences of AP activity between MSG and control rats were about 21 % ($P < 0.01$) and 22 % ($P < 0.05$) in the duodenum and in the jejunum, respectively.

In ADX-MSG rats a significantly reduced AP activity was found (the decrease was 18 % in the duodenum and 22 % in the jejunum, $P<0.01$). A similar tendency was also observed in the DPP IV activity of the ADX-MSG as well as of lean control rats. As compared to the sham-operated groups the decrease of enzyme activity was about 28 % and 23 % ($P<0.05$) in the duodenum and 18 % and 18 % ($P<0.05$) in the jejunum of ADX-MSG and ADX-control rats, respectively. In contrast, no significant effect of adrenal withdrawal on maltase activity has been found. Two-way ANOVA revealed specific changes of AP activity in both structures of the small intestine (interaction $F=10.78$, $P<0.05$ in the duodenum and interaction $F=4.63$, $P<0.05$ in the jejunum). In this way a significant effect of MSG-treatment ($F=6.916$, $P<0.05$) and of ADX ($F=10.530$, $P<0.05$) on the jejunal AP activity could be observed. The same analysis did not disclose a significant interaction of DPP IV and maltase.

Discussion

Obesity associated with significantly increased body fat, decreased body mass, and food intake in sham-operated MSG rats as well as mitigation of obesity in ADX-MSG rats observed in our experiment are in agreement with earlier studies (Tokuyama and Himms-Hagen 1989, Morris *et al.* 1998, Mistlberger and Antle 1999, Martinková *et al.* 2000). Our results also confirmed a significantly enhanced alkaline phosphatase activity in the small intestine (Mozeš *et al.* 2000, Martinková *et al.* 2000). This enzyme is involved in nutrient (fat) absorption and transport of long chain fatty acids in the intestinal mucosa (Takase and Goda 1990, Bernard *et al.* 1992) in response to postnatal glutamate-treatment, suggesting its possible fundamental role for the maintenance of an elevated body fat mass once obesity had been established. There also exists a parallelism between another form of obesity and the activities of small intestinal disaccharidases which influence the digestion of carbohydrates. However, due to the different experimental approaches and models used, these investigations have yielded different results. Thus, increased activity of jejunal sucrase and maltase was observed in obese Zucker rats in contrast to the unchanged maltase activity in 25-week-old VMH lesioned rats maintained on standard laboratory diets as compared to their lean littermates (Matsuo *et al.* 1992). On the other hand, in Otsuka Long-Evans Tokushima

fatty rats, significantly increased sucrase and isomaltase activities and increased mRNA encoding the sucrase isomaltase complex in the jejunum were observed only at 48 weeks of age but not in younger animals (Adachi *et al.* 1999). In addition, it was also found that genetically obese mice and MSG-treated rats in comparison to their lean littermates exhibited a significantly higher biochemically estimated intestinal sucrase activity at day 21 and higher histochemically estimated alkaline phosphatase activity at day 30 (Flores *et al.* 1990, Mozeš *et al.* 2000), which surprisingly precedes the development of excessive obesity and feeding perturbations observed in later periods of life. From this point of view studies have shown that the induction of enzymatic changes during the postweaning period might primarily be independent of the obesity status and that other factors are important in mediation of the increased enzyme "set point" at that time.

Our present results provide first experimental evidence that the adrenal cortex plays an important role in mediating the induction of enzymatic changes in the enterocytes of MSG rats. Moreover, considering the assumption that these functional changes were proportional to the previously enhanced adrenal secretion and their deficit after adrenalectomy (Tokuyama and Himms-Hagen 1989, Kiss *et al.* 1999, Guimaraes *et al.* 2002), some differences in the activity of small intestinal digestive enzymes after ADX surgery have been reported between MSG and control rats. While adrenal withdrawal selectively decreased both duodenal and jejunal alkaline phosphatase activity in MSG obese rats, no specific interaction of ADX with DPP IV and maltase activity has been observed in the above mentioned groups of rats. The present data are also in agreement with the observation regarding participation of intestinal enzymes in the digestion and absorption of individual food macronutrients (Leichter *et al.* 1984, Takase and Goda 1990, Matsumoto *et al.* 1995) and with the effect of ADX which significantly reduced food efficiency, i.e. decreased fat mass gain without corresponding reduction in food intake (Edens *et al.* 1999). In fact, it has been found that ADX in MSG rats induced more pronounced enzymatic disturbances and a decreased body mass gain despite the almost similar food intake as compared with lean rats. Thus both energy utilization and increased AP activity in obese MSG-sham rats appears to be mediated *via* elevated corticoids levels. Our results obtained in ADX control rats are, with the exception of maltase, not in agreement with previous data in the literature on

decreased AP and leucine aminopeptidase activity in ADX non-obese rats (Majumdar 1981, Foligne *et al.* 2001). This low responsiveness can be partially explained by the different age and perhaps the enzymatic status of the animals prior to adrenalectomy (20-day-old vs. 3-month-old rats) or might also be attributed to the duration of the experiments that differently affects the adaptability of the small intestine to ADX. For example, partial atrophy and disorganization of the intestinal epithelium (Foligne *et al.* 2001) was found 10 days after ADX, while no impairment of the intestinal mucosa morphology was observed four weeks after ADX (Gloria *et al.* 1997). Conflicting data have been found in the literature on the effect of ADX upon corticosterone levels in the circulation of non-obese animals. Corticosterone concentrations in the serum were still reported 40 days after bilateral adrenalectomy (Tokuyama and Himms-Hagen 1989), although shortly after adrenal withdrawal (day 12) the corticosterone levels were not detectable (Mantha and Deshaies 2000).

An important result of our study concerns the fact that the decreased function of the small intestine contributes to the ADX-induced attenuation of MSG obesity. It is not clear whether such a mechanism is also operative in other forms of obesity (associated with hyperphagia). In dietary obese and Zucker fatty rats, a pharmacological blockade of glucocorticoid receptors (Langley and York 1990, Okada *et al.* 1992) or suppression of intestinal disaccharidase activities by administration of its inhibitor (Kobatake *et al.* 1989, Matsuo *et al.* 1992) caused a substantial reduction of body fat depots and food intake. This suggests that both nutrition and enzyme activities in the small intestine may

be stimulated when the corticoids are elevated. However, obesity developed in Zucker rats even after food restriction (Cleary *et al.* 1980) which indicated that similarly as in MSG obesity, enhanced enzyme activity in the small intestine and the different effectiveness of nutrient absorption might also be significant in the maintenance of their higher adiposity. Such a possibility seems to be supported by the evidence emerging from studies demonstrating a) increased AP activity without hyperphagia in MSG rats (Mozeš *et al.* 2000), b) decreased AP activity and attenuation of obesity despite enhanced food intake in ADX-MSG rats (present experiment), c) lower satiating effect of fats in the small intestine of obesity-prone rats than that in obesity-resistant rats (Greenberg *et al.* 1999), d) more effective absorption of food in the upper part of the intestine of obese subjects (Wright *et al.* 1983, Wisen and Johansson 1992). From this point of view our results brought additional evidence for the idea that enzyme activity in the small intestine might be an important factor in the modulation of altered energy balance in MSG-induced obesity. This provides a model for investigations on the interaction between the function of the small intestine and inadequate food intake control as well as for energy metabolism in other forms of obesity and allows better understanding of the mechanisms that may limit or prevent the risk of obesity in later life.

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References

- ADACHI T, TAKENOSHITA M, KATSURA H, YASUDA K, TSUDA K, SEINO Y, ENOMOTO T, YAMAJI R, MIYATAKE K, INUI H, NAKANO Y: Disordered expression of the sucrase-isomaltase complex in the small intestine in Otsuka Long-Evans Tokushima fatty rats, a model of non-insulin-dependent diabetes mellitus with insulin resistance. *Biochim Biophys Acta* **1426**: 126-132, 1999.
- BERNARD A, CASELLI C, BLOND JP, CARLIER H: Diet fatty acid composition, age and rat jejunal microvillus enzyme activities. *Comp Biochem Physiol A* **101**: 607-612, 1992.
- BHATNAGAR S, BELL ME, LIANG J, SORIANO L, NAGY TR, DALLMAN MF: Corticosterone facilitates saccharin intake in adrenalectomized rats: does corticosterone increase stimulus salience? *J Neuroendocrinol* **12**: 453-460, 2000.
- CASTONGUAY TW, DALLMAN MF, STERN JS: Some metabolic and behavioral effects of adrenalectomy on obese Zucker rats. *Am J Physiol* **251**: R923-R933, 1986.
- CLEARY MP, VASSELLI JR, GREENWOOD MRC: Development of obesity in Zucker obese (fa/fa) rat in absence of hyperphagia. *Am J Physiol* **238**: E284-E292, 1980.

- CUNNINGHAM JJ, CALLES-ESCADON J, GARRIDO F, CARR DB, BODE HH: Hypercorticosteronuria and diminished pituitary responsiveness to corticotropin-releasing factor in obese Zucker rats. *Endocrinology* **118**: 98-101, 1986.
- DOLNIKOFF M, MARTIN-HIDALGO A, MACHADO UF, LIMA FB, HERRERA E: Decreased lipolysis and enhanced glycerol and glucose utilization by adipose tissue prior to development of obesity in monosodium glutamate (MSG) treated-rats. *Int J Obes* **25**: 426-433, 2001.
- EDENS NK, MOSHIRFAR A, POTTER GM, FRIED SK, CASTONGUAY TW: Adrenalectomy reduces adiposity by decreasing feed efficiency, not direct effect on white adipose tissue. *Obes Res* **7**: 395-401, 1999.
- FLORES CA, BEZERRA J, BUSTAMANTE SA, GODA T, MACDONALD MP, KAPLAN ML, KOLDOVSKY O: Age-related changes in sucrase and lactase activity in the small intestine of 3- and 10-week-old obese mice (C57BL/6J obob). *J Am Coll Nutr* **9**: 255-260, 1990.
- FOLIGNE B, AISSAOUI S, SENEGAS-BALAS F, CAYUELA C, BERNARD P, ANTOINE J-M, BALAS D: Changes in cell proliferation and differentiation of adult rat small intestine epithelium after adrenalectomy: kinetic, biochemical, and morphological studies. *Dig Dis Sci* **46**: 1236-1246, 2001.
- FREEDMAN MR, HORWITZ BA, STERN JS: Effect of adrenalectomy and glucocorticoid replacement on development of obesity. *Am J Physiol* **250**: R595-R607, 1986.
- GLÓRIA H, PORTELA-GOMES GM, GRIMELIUS L, AHLMAN H, FERRA MA: Effect of adrenalectomy on serotonin-, somatostatin-, and gastrin-immunoreactive cells in rat gastrointestinal tract. *Dig Dis Sci* **42**: 1216-1220, 1997.
- GREENBERG D, MCCAFFERY J, POTACK JZ, BRAY GA, YORK DA: Differential satiating effect of fats in the small intestine of obesity-resistant and obesity-prone rats. *Physiol Behav* **66**: 621-626, (1999)
- GOSSRAU R: Histochemische und biochemische Untersuchung der α -Glucosidasen mit 2-Naphthyl- α -D-glucosid. *Histochemistry* **49**: 193-211, 1976.
- GUILLAUME-GENTIL C, ROHNER-JEANRENAUD F, ABRAMO F, BESTETTI GE, ROSSI GL, JEANRENAUD B: Abnormal regulation of the hypothalamo-pituitary-adrenal axis in the genetically obese fa/fa rat. *Endocrinology* **126**: 1873-1879, 1990.
- GUIMARAES RB, TELLES MM, COELHO VBO, MORI RCT, NASCIMENTO CMO, RIBEIRO EB: Adrenalectomy abolishes the food-induced hypothalamic serotonin release in both normal and monosodium glutamate-obese rats. *Brain Res Bull* **58**: 363-369, 2002.
- KISS A, ŠKULTETYOVÁ I, JEŽOVÁ D: Corticotropin-releasing hormone synthesizing neurons in the hypothalamic paraventricular nucleus of rats neonatally treated with monosodium glutamate can respond to different stress paradigms. *Neurol Res* **8**: 775-780, 1999.
- KOBATAKE T, MATSUZAWA Y, TOKUNAGA K, FUIOKA S, KAWAMOTO T, KENO Y, INUI Y, OKADA H, MATSUO T, TARUI S: Metabolic improvements associated with a reduction of abdominal visceral fat caused by a new α -glucosidase inhibitor, AO-128, in Zucker fatty rats. *Int J Obes* **13**: 147-154, 1989.
- LANGLEY SC, YORK DA: Effect of antiglucocorticoid RU 486 on the development of obesity in obese fa/fa Zucker rats. *Am J Physiol* **259**: R539-R544, 1990.
- LEICHTER J, GODA T, BHANDARI S, BUSTAMANTE S, KOLDOVSKÝ O: Relation between dietary induced increase of intestinal lactase activity and lactose digestion and absorption in adult rats. *Am J Physiol* **247**: G729-G735, 1984.
- LOJDA Z: New data on dipeptidylaminopeptidase IV (DAP IV). *Sb Cs spol histochem cytochem* **6**: 59-69, 1977.
- LOJDA Z, GROSSRAU R, SCHIBLER TH: *Enzyme Histochemistry*. Springer, Berlin, 1979, pp 59-70.
- MACHO L, JEŽOVÁ D, ZÓRAD Š, FICKOVÁ M: Effect of postnatal monosodium glutamate treatment on adrenal cortex function. *Physiol Res* **48** (Suppl 1): S50, 1999a.
- MACHO L, JEŽOVÁ D, ZÓRAD Š, FICKOVÁ M: Postnatal monosodium glutamate treatment results in attenuation of corticosterone metabolic rate in adult rats. *Endocr Regul* **33**: 61-67, 1999b.
- MACHO L, FICKOVÁ M, JEŽOVÁ D, ZÓRAD Š: Late effects of postnatal administration of monosodium glutamate on insulin action in adult rats. *Physiol Res* **49** (Suppl 1): S79-S85, 2000.

- MAJUMDAR APN: Bilateral adrenalectomy. Effect of hydrocortisone and pentagastrin on the activity of intestinal disaccharidases and alkaline phosphatase in weanling rats. *Scand J Gastroenterol* **16**: 177-182, 1981.
- MANTHA L, DESHAIES Y: Energy intake-independent modulation of triglyceride metabolism by glucocorticoids in the rat. *Am J Physiol* **278**: R1424-R1432, 2000.
- MANTHA L, PALACIOS E, DESHAIES Y: Modulation of triglyceride metabolism by glucocorticoids in diet induced obesity. *Am J Physiol* **277**: R455-R464, 1999.
- MARTINKOVÁ A, MOŽEŠ Š, LENHARDT L: Effect of neonatal MSG treatment on day-night alkaline phosphatase activity in the rat duodenum. *Physiol Res* **49**: 339-345, 2000.
- MATSUMOTO H, ERICKSON RH, KIM YS: Localization and characterization of rat small intestinal aminopeptidase P and its role in prolyl peptide digestion. *J Nutr Biochem* **6**: 104-110, 1995.
- MATSUO T, ODAKA H, IKEDA H: Effect of intestinal disaccharidase inhibitor(AO-128) on obesity and diabetes. *Am J Clin Nutr* **55**: 314S-317S, 1992.
- McINTOSH M, BAO H, LEE C: Opposing actions of dehydroepiandrosterone and corticosterone in rats. *Proc Soc Exp Biol Med* **221**: 198-206, 1999.
- MISTLBERGER RE, ANTLE MC: Neonatal monosodium glutamate alters circadian organization of feeding, food anticipatory activity and photic masking in the rat. *Brain Res* **842**: 73-83, 1999.
- MORRIS MJ, TORTELLI CF, FILIPPIS A, PROIETTO J: Reduced BAT function as a mechanism for obesity in the hypophagic, neuropeptide deficient monosodium glutamate-treated rat. *Regul Pept* **75-76**: 441-447, 1998.
- MOŽEŠ Š, LENHARDT L, MARTINKOVÁ A: Alkaline phosphatase activity of duodenal enterocytes after neonatal administration of monosodium glutamate to rats. *Physiol Res* **49**: 269-277, 2000.
- OKADA S, YORK DA, BRAY GA: Mifepristone (RU 486), a blocker of type II glucocorticoid and progestin receptors, reverses a dietary form of obesity. *Am J Physiol* **262**: R1106-R1110, 1992.
- RAČEK L, LENHARDT L, MOŽEŠ Š: Effect of fasting and refeeding on duodenal alkaline phosphatase activity in monosodium glutamate obese rats. *Physiol Res* **50**: 363-372, 2001.
- ROMSOS DR, VANDER TUIG JG, KERNER J, GROGAN CK: Energy balance in rats with obesity-producing hypothalamic knife cuts: effects of adrenalectomy. *J Nutr* **117**: 1121-1128, 1987.
- TAKASE S, GODA T: Effects of medium chain triglycerides on brush border membrane-bound enzyme activity in rat small intestine. *J Nutr* **120**: 969-976, 1990.
- TOKUYAMA K, HIMMS-HAGEN J: Adrenalectomy prevents obesity in glutamate-treated mice. *Am J Physiol* **237**: E139-E144, 1989.
- TROCKI O, BAER DJ, CASTONGUAY TW: Comparison of effects of adrenalectomy and RU-486 in rats given a choice of maintenance diet and fat supplement. *Am J Physiol* **269**: R708-R719, 1995.
- WISER O, JOHANSSON C: Gastrointestinal function in obesity: motility secretion, and absorption following a liquid test meal. *Metabolism* **41**: 390-395, 1992.
- WRIGHT RA, KRINSKY S, FLEEMAN C, TRUJILLO J, TEAGUE E: Gastric emptying and obesity. *Gastroenterology* **84**: 747-751, 1983.
- ZHANG WM, KUCHAR S, MOŽEŠ Š: Body fat and RNA content of the VMH cells in rats neonatally treated with monosodium glutamate. *Brain Res Bull* **35**: 383-385, 1994.

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