Effect of Partial Replacement of Monosodium Glutamate with 5´Inosine Monophosphate on Some Organs, Hormonal Balance and Activity of Rats

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ABSTRACT

The effects of oral intake of the most well known flavour enhancer monosodium glutamate (MSG) and/or inosine monophosphate (5´-IMP) on neuronal brain damages, pancreas injury, hypoactivity and imbalance release of noradrenaline (NA) and insulin (Ins) were studied in both adult and infant rats. Different neuronal damages were observed in different brain areas included cerebrum, hypothalamus and hippocampus when MSG was administrated alone at level of 60 mg/kg body weight/day. Synchronous with these damages, hypoactivity in the movement behavior of both adult and infant rats was noticed. Also, imbalance of hormone secretion was demonstrated by recording raised level of NA and reduction in Ins release in blood serum, resulting in raise of blood glucose concentration. Pancreatic damages such as hypertrophy of Langerhan's islets, hemorrhage, and leucocytic cells infiltration were also observed. Partial replacement of MSG with 5´-IMP (1:1 w/w) led to significant ($P<0.05$) reduction in these undesirable changes. The infant rats are more susceptible to the MSG toxic effects than the adult rats. Therefore, the present study is concerned that the use of MSG as a flavour enhancer in the human diet prepared for the adults should be minimized and totally avoided from the infant foods. Although use of 5´-IMP led to minimize MSG-induced hazards, it is not recommended for the free use in infant foods in the early stages of live. Further investigations on safety of the mixture of MSG and 5´-IMP are also required. Sensory evaluation of MSG: 5´-IMP mixture added to beef burger was also achieved. The taste of beef burger contained this mixture was significantly ($P <0.05$) enhanced comparing with that contained MSG alone.

Key words: flavor enhancer- monosodium glutamate- inosine monophosphate- brain- pancreas- hypoactivity- noradrenaline- insulin.

INTRODUCTION

Monosodium glutamate (MSG) is a form of glutamate widely used as a flavour enhancer for a long time. It has been classified by the FDA as a Generally Recognized as Safe (GRAS) ingredient since 1958. This is the safest classification of food ingredient that can be given in the United States, along with pepper, sugar, vinegar and baking powder. The MSG was allocated an "Acceptable Daily Intake (ADI) not specified" by the Joint FAO/WHO Expert Committee of Food Additive (JECFA) and the Scientific Committee for Food of the Commission of the European Committee. The MSG has a characteristic taste called umami "savory deliciousness'', which is considered distinct from the four other basic tastes (sweet, sour, salty, and bitter). The optimal palatability concentration for MSG in foods is between 0.2 – 0.8% with the largest palatable dose for humans being about 60 mg/kg body weight (BW) (Walker & Lupien 2000).

On 1995, report from the Federation of American Societies for Experimental Biology (FASEB, 1995) listed two groups of people that are at risk when ingesting large amounts of MSG. Those people have intolerance to MSG or have severe and poorly controlled asthma. The report also mentioned that 0.5 to 2.5 grams of MSG is needed to produce a response in those individuals. Moreover, Chinese restaurant syndrome, also called monosodium glutamate symptom complex, is a collection of symptoms which may include headache, flushing, sweating, and a sensation of pressure in the mouth or face. It is commonly believed that MSG is the cause, but a short-term scientific study showed no link (Tarasoff & Kelly, 1993).

Several previous studies pointed out axonal and neuronal lesions due to injection of MSG di-

rectly into the brain of either neonatal or adult rats (Nemeroff *et al*., 1981, Park *et al*., 2000, Beas-Zárate, *et al*., 2002; Sanabria *et al*., 2002, Swelim, 2004). Examination of potential neurotoxicity was a major component of the safety evaluation, with reports from several separate studies on mice, rats, hamsters, dogs, rabbits, guinea pigs and duck being considered. Dubovicky *et al*. (1996) suggested the injection with 2 or 4 mg MSG / g BW had negative effect on habituation to a new environment in male rats. This effect was due to an increase in the exploratory activity. Also, MSG treatment of neonatal rats causes neuronal degeneration in various brain areas and leads to several neurochemical, endocrinological and behavioral alterations. However, relatively little is known about the development of neurological reflexes and motor coordination of these animals (Kiss *et al*., 2005). Moreover, elevatation of the erythrocyte glucose content accompanied by increase in lipid peroxidation were demonstrated after subcutaneous administration of MSG to normal adult male mice for 6 days at dose levels of 4 and 8 mg/g body weight (Ahluwalia *et al*., 1994).

Walker, (1999) studied different procedure for administrating MSG to dents and rabbits. He reported that focal necrosis in the hypothalamus was observed reproducibly in dents and rabbits after intravenous or subcutaneous administration of glutamate or after very high bolus doses by oral gavage. To produce these lesions the order of 1000 mg/kg body weight as a bolus dose was required. Moreover, the oral ED50 for production of hypothalamic lesions in the neonatal mouse is about 500 mg/kg body weight by gavage, whereas the largest palatable dose for humans is about 60 mg/kg body weight with higher doses causing nausea (Food Standards Australia Newzeland, 2003). Therefore, the daily oral intake would not exceed this level. Recently, Nakanishi *et al*. (2008) suggested that MSG should have its safety profile re-examined and be potentially withdrawn from the food chain.

The discovery of umami receptors, taste receptors for L-glutamate, is one of the recent highlights of taste research. Following the success of microbial large-scale production of L-glutamate, synergism of 5´-ribonucleotides with MSG stimulated research on nucleotide overproduction greatly (Elhariry *et al*. 2004).The intensity of umami taste is markedly enhanced by mixing glutamate with 5´-ribonucleotides such as 5´-inosine monophosphate (5´-IMP) and 5´-guanisine monophosphate (5´-GMP). Remarkably, single components of a highly familiar odour-taste pair may nevertheless elicit weak rating of the missing substance, possibly due to stimulation of particular neurons with optimum response to their combination by either smell or taste (Delwiche, 2004). A 1:1 mixture of MSG and these 5´-ribonucleotides acts synergistically and gives flavor intensity 30 times stronger than that of monosodium glutamate alone (Maga 1994). Based on this assumption, the aim of the present study was to evaluate the role of the partial replacement of MSG with 5´-IMP for reducing the neuronal hazards induced by the oral intake of MSG. The consequence effects on the secretion balance of some hormones and the exploratory behaviour were also studied.

MATERIALS AND METHODS

Chemicals

Monosodium glutamate (MSG) and 5´inosine monophosphate (5´-IMP) were purchased from Sigma Chemical Company, Louis, U.S.A. The kits for glucose determination was obtained from Stanbio Laboratories, Boerne, Texas, USA; Noradrenaline ELISA (KAPL 10-0200) from Biosource Europe, Nivelles, Belgium and Insuline AccubindTM ELISA Microwells (PR Code = 2425-300A) from Monobind Inc., Lake Forest, California, USA.

Animals and Treatments

Forty eight male Sprague-Dawely strain rats were obtained from Research Institute of Ophthalmology, Giza, Egypt, 24 were adult animals (145±2g and 10 weeks age) and 24 were infant animals (45±2g and 3 weeks age). The animals were housed in separate stainless steel cages raised in a well-ventilated room with 12-h light/dark cycle and free access to food and water throughout the experimental period (90 days). After adaptation period (10 days), rats were divided into two main classes; adult class and infant class. The adult class is one confined to rats aged over 10 weeks (weighed 145±2 g). It was randomly divided into four groups; G1, G2, G3 and G4. The infant class is confined to weaning rats aged 3 weeks (weighed 45±2g). This class was randomly divided into four groups; G5, G6, G7 and G8. Each group contained six rats. All animals were fed on basal diet (10 % casein, 10 % corn oil, 5% cellulose, 1 % vitamin mixture, 4 % salt mixture, 70 % corn starch (Lana Peter & Pearson, 1971) for 90 days. Rats of G1 and G5 were used as controls. The

other six groups were administrated with different concentrations of MSG and/or 5´-IMP by stomach tube. Rats of G2, G3 and G4 were orally administrated with 60 mg MSG/ kg BW, 30 mg MSG +30 mg 5´-IMP (1:1 w/w), and 30 mg 5´-IMP / kg BW, respectively. Rats of G6, G7 and G8 were administrated with 30 mg MSG/ Kg BW, 15 mg MSG + 15 mg 5´-IMP (1:1 w/w) and 15 mg 5´-IMP / kg BW, respectively. The doses of MSG and 5´-IMP were doubled when the rats of G6, G7 and G8 reached to the weight of adult age (7 weeks from the beginning of the experiment).

Exploratory behaviour

Spontaneous exploratory behaviour was evaluated in an open field test as described by Dubovicky *et al*., (1996). All animals were exposed to the open field in an empty cage (36 × 48 cm, wall height 25 cm) for 6 min. The bottom of each cage was divided into six equal squares. Spontaneous motor activity (number of crossed squares) and vertical exploratory activity (number of rears: both forepaws lifted off the floor) were recorded on day 45 and 90.

Body gain weight

Rats were observed daily for the appearance of any symptoms of discomfort that might be related to studied treatments. Body weight (BW) of the rats was recorded daily before administration of studied additives. At the end of experiment period, the percentage of gain weight was expressed [(final weight – beginning weight) / beginning weight] × 100.

Biochemical assay

At the end of experimental period, blood samples were collected from the eye plexuses of animals by a fine capillary glass tubes and placed immediately on ice. Blood serum samples were collected into dry clean centrifuge tubes; the serum was separated after centrifugation for 10 min at 1500 xg and kept at –20 oC until analysis. The activity of noradrenaline (NA) and insulin (Ins) hormones in the blood serum of rats were estimated according to the producer's instructions that based on the methods of Manz *et al*., (1990) and Kahn & Rosenthal (1979), respectively. Glucose concentration in the blood serum of rats was assayed by the method described by Folin & WU (1920).

Brain and pancreas weight

At the end of the experimental period, rats were weighted and killed by diethyl ether. Brain and pancreases were cut off, washed in ice-cooled saline solution 0.15M KCl to remove blood and weighed. The brain and pancreas weight percentage were calculated as a percentage from the body weight on day 90.

Histopathological studies

Different brain sections (included cerebral cortex, hyppocampal and hypothalamic regions) and tissue specimens from rat's pancreases were prepared for histological examination according to the method described by Yang *et al*., (2000) and Bancroft *et al*., (1996), respectively.

Sensory assessment of flavour enhancement effect of MSG/5´-IMP mixture

The MSG was added as flavour enhancer to beef burger at level of 0.5 % according to the Egyptian Standards (E.S. 2005). A 1:1 w/w mixture of MSG and 5´-IMP was applied as a partial replacement of MSG with 5´-IMP (Maga, 1994). Cooked beef burger samples were assessed for their sensory attributes by ten panelists from the staff members of Food Science Department, Faculty of Agriculture, Ain Shams University according to Klein and Bardy (1984). The panelists were asked to score the different samples for appearance, colour, aroma, taste, juiciness, tenderness and overall acceptability as follows: very good 8-9, good 6-7, fair 4-5, poor 2-3, and very poor 1-2.

Statistical analysis

All values are means±SE obtained from six animal groups. Data were analyzed with SAS software (SAS Institute, Cary, N.C.) using SAS analysis of variance (PROC ANOVA). Significant differences between means were determined by Duncan's multiple range test ($P < 0.05$).

RESULTS AND DISCUSSION

The main objective of the present study was to evaluate the role of the partial replacement of MSG with 5´-IMP for reducing the neuronal hazards induced by the oral intake of MSG. Therefore, half amount of MSG in beef burger was replaced with 5´-IMP. Aroma, taste and overall acceptability of beef burger contained the mixture of MSG and 5´-IMP were significantly ($P<0.05$) enhanced compared with samples contained MSG alone (untabulated data). On the other hand, no significant enhancement was recorded for other sensory attributes due to the partial replacement of MSG with 5´-IMP. This find-

ing confirmed that the 1:1 mixture of MSG and 5′ IMP has a synergistic effect and led to improvement of the flavour intensity as compared with those of MSG alone (Maga ,1994 & Delwiche, 2004).

Since (i) the dose of MSG added to foods is between 0.2 – 0.8% depending on the product itself and (ii) the ADI is not specified, the largest palatable dose of MSG (60 mg/kg BW) was applied for the biological evaluation in the present study using both the adult and infant rats. The partial replacement of MSG with 5′-IMP at the level of 1:1 (w/w) was also investigated.

Gain weight and organs weight

Percentage gain body weight and weight of brain and pancreas of investigated adult and infant rat classes were summarized in Table (1). No significant difference ($P<0.05$) was observed among weights of the brain of all studied groups. Remarkably, enlarged pancreases were obtained after MSG treatment in both infant and adult rats (Table 1). On the other hand, gain weight of the rats in infant class (ranged between 82.3 ±0.85 and 85.2 ±0.10%) was higher than those of the adult class (ranged between 55.2 ±0.94 and 60.3±1.20%). Inside the class of adult rats (Table 1) gain weight of the rats received MSG (G2) or combined with 5′-IMP (G3) was significantly ($P<0.05$) higher than those of the other groups (G1 and G4). Response of the infant rats to administration with the studied additives indicated that the infant rats were more sensitive than the adult rats (Table 1). This finding suggested that metabolic behaviour of MSG differed in the infant rats as compared with adult rats. At the same time, MSG induced increment of body weight in infant rats. This could be explained by those mentioned by Martins *et al*. (2001). They suggested that the lesion in hypothalamus causes an impairment of sympathetic transmission in adrenal medulla and less catecholamines accumulation and secretion. They added that this defect might be involved in the onset of MSG obesity in rats.

Brain damages and noradrenaline release due to oral intake of MSG

Neural damage, neuronophagia and multifocal cerebral hemorrhage were observed in the brain of adult rats received 60 mg MSG/kg BW/day (G2, Fig. 1). On the other hand, no histological change was noticed by examining different brain sections of the adult rats in control group (G1), rats administrated with 1:1 mixture of MSG and 5′-IMP (G3) and rats received 5′-IMP only (G4)

In infant-class rats, brain of the control group (G5) revealed normal histological structures (Fig. 2). Various histological alterations were demonstrated in G6 when different brain sections included cerebrum, hypothalamus and hippocampus were examined. One of these alterations was central chromatolysis which was emerged by formation of dark eosinophilic neurons due to dispersion of Nissl bodies around the nucleus (Fig. 2, white closed arrows). The other alterations in brain of G6 were multifocal gliosis (Fig. 2, Black arrows) in addition to neuronal damage and neuronophagia (Fig. 2, white open arrows). The brain of most rats in G7, which received mixture of MSG and 5′-IMP, showed normal histological structures except the brain of only one rat

Table 1: Gain body weight and weight of brain and pancreas of rats after 90 days of oral administration with MSG and/or 5′-IMP

Rat Class	Rat group	Daily intake (mg/kg BW)	Weight of organs (g)		Gain weight (%)
			Brain	Pancreas	
Adult	G1	Control	0.43±0.03^{a}	0.26±0.01^{c}	55.2±0.94^{b}
	G2	60 MSG	0.39±0.03^{a}	0.45±0.03^{a}	60.3±1.20^{a}
	G3	30 MSG + 30 5′-IMP	0.45±0.07^{a}	0.31±0.01^{b}	58.0±0.81^{a}
	G4	30 5′-IMP	0.39±0.09^{a}	0.32±0.04^{b}	56.3±1.01^{bc}
Infant*	G5	Control	0.54±0.01^{a}	0.42±0.03^{b}	82.3±0.85^{b}
	G6	30 MSG	0.43±0.01^{a}	0.51±0.01^{a}	84.7±0.42^{a}
	G7	15 MSG + 15 5′-IMP	0.43±0.04^{a}	0.38±0.05^{c}	85.2±0.10^{a}
	G8	15 5′-IMP	0.45±0.02^{a}	0.38±0.08^{c}	84.5±0.85^{a}

* The doses of MSG and 5′-IMP were doubled when the weight of rat in infant class reached to the weight of adult rats (7 weeks from the beginning of the experiment). In the same rat class, means ± S.E. within the same column with the different superscripted letter are significantly different ($P \leq 0.05$).

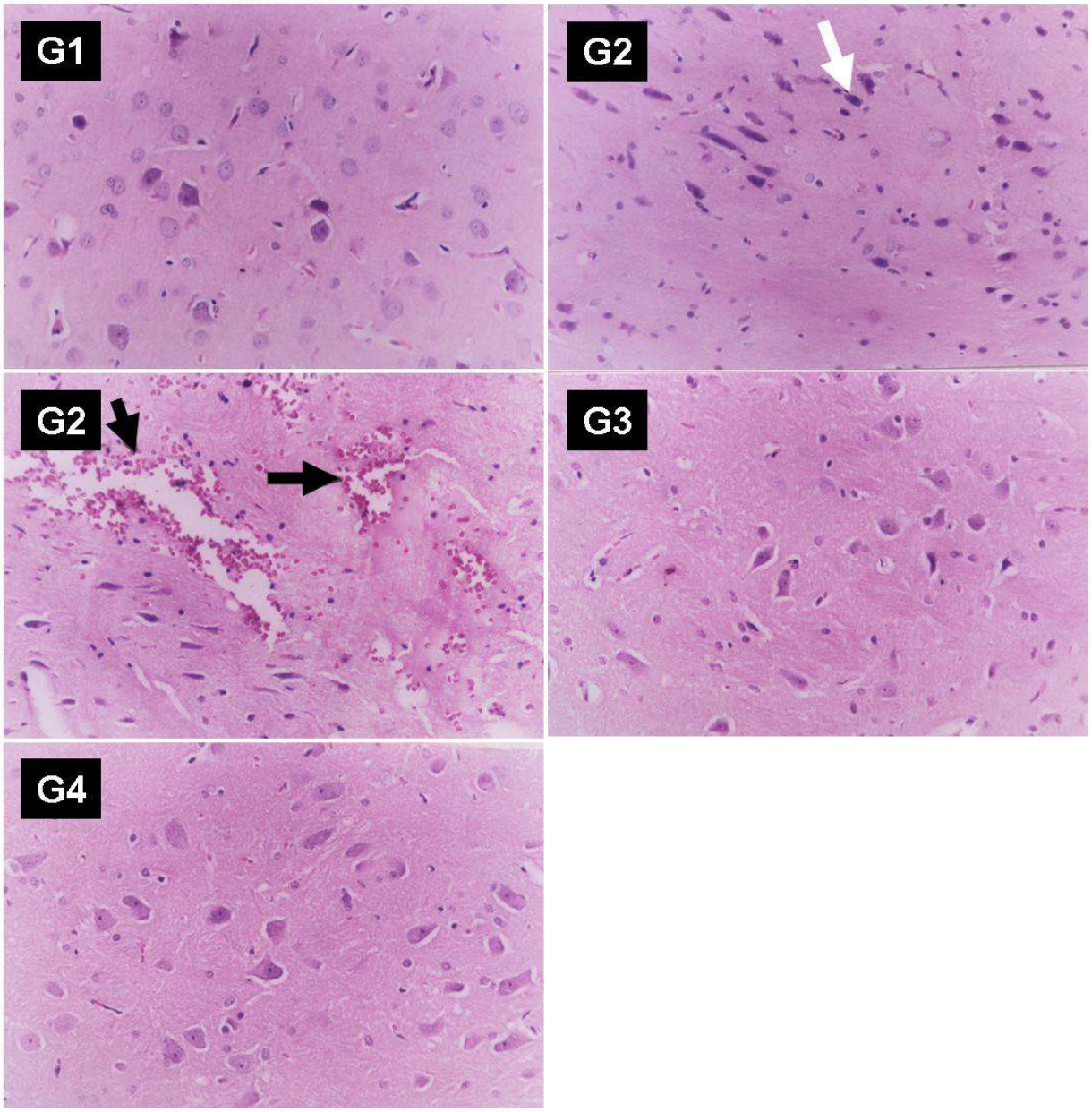

Fig. 1: Histopathological changes in the brain of the adult rats due to daily oral intake of 60 mg MSG (G2), 30 mg MSG + 30 mg 5´-IMP (G3) or 30 mg 5´-IMP (G4) / kg BW. The experimental period was 90 days, G1 presented the control group. White arrow indicates neural damage and neuronophagia, whereas black arrow indicates multifocal cerebral hemorrhage (H&E, X200)

of this group that revealed slight neural damage. On the other hand, rats received 5´-IMP alone (G8) showed no histological changes in brain (Fig. 2).

Administration of 60 mg MSG/ kg BW/ day led to a significant increase ($P<0.05$) in serum noradrenaline (NA) of the adult rats (Table 2), where NA increased from 8.7 ±0.24 to 13.2 ±0.63 ng/ml for adult control group (G1) and MSG group (G2), respectively. Also, slight increase in the level of serum NA was also recorded when 5´-IMP or 5´-IMP combined with MSG was administrated to the adult rats in G3 and G4. In infant rats, remarkable induction in secretion of NA was determined (14.5 ±0.85 ng/ml) in the serum of infant rats received MSG (G6) as compared with that of control rats (10.1±1.04 ng/ml). No significant diference ($P<0.05$) was noticed in serum NA of the infant rats in the control group and G7 or G8 (Table 2).

There are several reports that parental administration of high doses of MSG to adult rats sig-

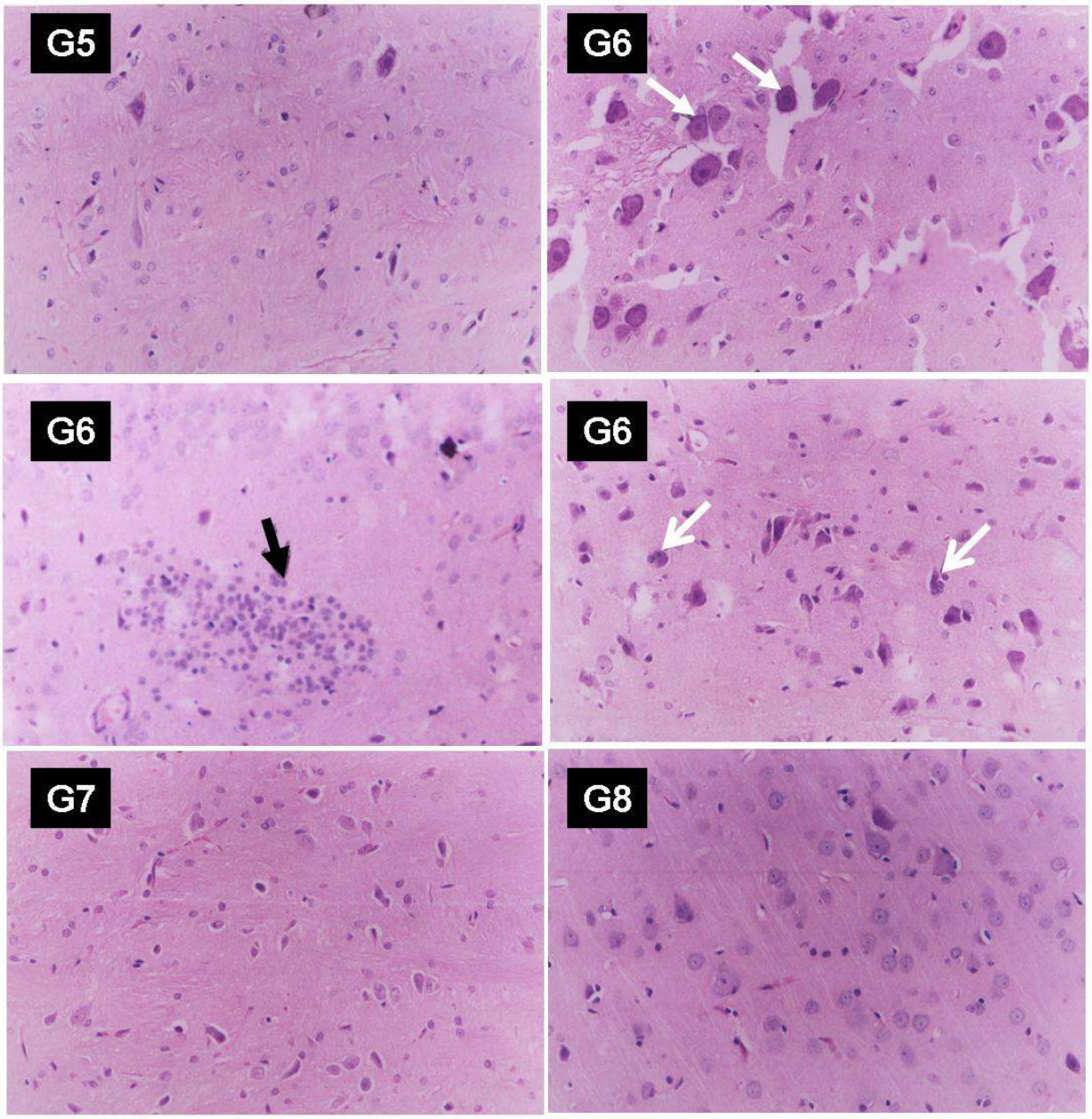

Fig. 2: Histopathological changes in the brain of the infant rats due to daily oral intake of 30 mg MSG (G6), 15 mg MSG+15 mg 5´-IMP (G7) and 15 mg 5´-IMP (G8) / kg BW. The doses of MSG and 5´-IMP were doubled when the weight of rat in infant class (G6, G7 and G8) reached to the weight of adult rats (7 weeks from the beginning of the experiment). The experimental period was 90 days, G5 presented the control. White closed arrow indicates central chromatolysis, Black arrow indicates multifocal gliosis, White open arrow indicates neuronal damage and neuronophagia (H&E, X200)

nificantly raised the levels of the acidic amino acids in the brain resulting in brain lesions (Park *et al*., 2000). The histological changes in the present study indicate occurrence of damages in the brain of the adult and infant rats due to receiving MSG. These brain lesions may lead to induce secretion of stress hormones such as noradrenaline (also known as "norepinephrine") which releases from terminals of the sympathetic nerve system. This finding is in harmony with those stated by Mautes *et al*., (2001). They found that NA content was elevated in the serum and cerebrospinal fluid in most patients with acute cerebral blood deficiency. Therefore NA was believed to participate in the pathophysiology of injury to the blood-brain barrier following brain damage. Also, Zhiqiang *et al*., (2005) studied the

Table 2: Noradrenaline (NA), Insulin (Ins) and blood glucose concentration in blood serum of rats after 90 days of oral administration with MSG and/or 5′-IMP

Rat Class	Rat group	Daily intake (mg/kg BW)	NA ng/ ml	Ins μIU/ ml	Glucose mg/dl
Adult	G1	Control	8.7±0.24^{b}	10.4±0.33^{a}	88±1.11^{c}
	G2	60 MSG	13.2±0.63^{a}	6.4±1.85^{b}	153±5.43^{a}
	G3	30 MSG + 30 5′-IMP	10.7±1.68^{ab}	8.4±1.2^{ab}	131±2.35^{b}
	G4	30 5′-IMP	10.7±0.23^{ab}	9.3±0.88^{a}	132±1.26^{b}
Infant*	G5	Control	10.1±1.04^{b}	10.6±0.88^{ab}	74±2.28^{c}
	G6	30 MSG	14.5±0.85^{a}	9.0±0.58^{b}	125±7.43^{a}
	G7	15 MSG + 15 5′-IMP	10.9±0.76^{b}	12.1±1.0^{ab}	88±1.49^{b}
	G8	15 5′-IMP	9.4±0.79^{b}	13.8±1.2^{a}	86±2.85^{b}

* The doses of MSG and 5′-IMP were doubled when the weight of rat in infant class reached to the weight of adult rats (7 weeks from the beginning of the experiment). In the same rat class, means ± S.E. in the same column with the different superscripted letter are significantly different ($P \leq 0.05$).

changes of noradrenaline in brain homogenate of rats with brain damage secondary to intracerebral hemorrhage. They mentioned that noradrenaline is involved in the pathogenesis of secondary damage of the brain during intracerebral hemorrhage.

Changes of pancreas histology, insulin secretion and blood glucose level

Pancreas of the control adult-class rats (G1) did not reveal any histological changes (Fig. 3). Meanwhile, pancreas of the rats in G2 showed hemorrhage in Langerhan's islets and leucocytic cells infiltration (Fig. 3). Moreover, pancreas of rats in G3 showed hyperplasia of epithelial liming pancreatic duct associated with few leucocytic cells infiltration. However, all investigated pancreatic specimens of rats that received an oral administration of 5′-IMP (G4) did not show any histological changes (Fig. 3).

In infant-class rats, number of multiple sections of pancreatic specimens judged a normal histological pancreatic architecture in G5 (Fig. 4). Conversely, hypertrophy of Langerhan's islets and vacuolation of their cells were visualized in pancreas of all rats of G6 (Fig. 4). Also, in the rat of G7, vacuolation of some cells of Langerhan's islets was also noticed. On the other hand, no histological changes were observed in the pancreas of adult rats that administrated with 5′-IMP alone (G8, Fig 4).

The concentrations of insulin (Ins) in the blood serum of adult and infant control rats were 10.4 ±0.33 and 10.6 ±0.88, respectively (Table 1). Significant reduction ($P < 0.05$) in the level of Ins was recorded when rats were administrated with studied doses of MSG (G2 and G6). In adult rats, slight reduction in Ins concentration was noticed in the rats of G3, which received the mixture of MSG and 5′-IMP (1:1 w/w). On the other hand, no significant difference ($P < 0.05$) was obtained in the level of Ins of adult rats received 5′-IMP alone (G4) compared with that of the control (G1). In infant rats, the highest level of Ins was measured in the blood serum of rats received 5′-IMP alone (G8), whereas the lowest Ins concentration was recorded for the rats administrated with MSG alone (G6). Due to the significant reduction of insulin secretion, blood glucose concentrations of G2 and G6 were considerably increased comparing to the control groups (G1 and G5). In general, blood glucose concentrations of adult rats were higher than those recorded for the rats in infant class.

Along adrenaline, noradrenaline also underlies the fight-or-flight response, directly increasing heart rate, triggering the release of glucose from energy stores and increasing the blood flow into other organs (Okada *et al*., 2008). This may explain hypertrophy of Langerhan's islets and vacuolation of their cells in the pancreas of rats in G2 and G6. Moreover, the obtained results ensure that release of insulin is strongly inhibited by noradrenaline, which leads to increased blood glucose levels during stress. The simultaneous increases of noradrenaline and glucose in addition to decrease of insulin recorded after receiving MSG indicates that the normal crosstalk between α cells and β cells of the pancreas is disrupted. With respect to this, it is well known that glucose excites β cells (which secrete insulin) and inhibits α cells (which secrete glucagons). The aforementioned factors depend on

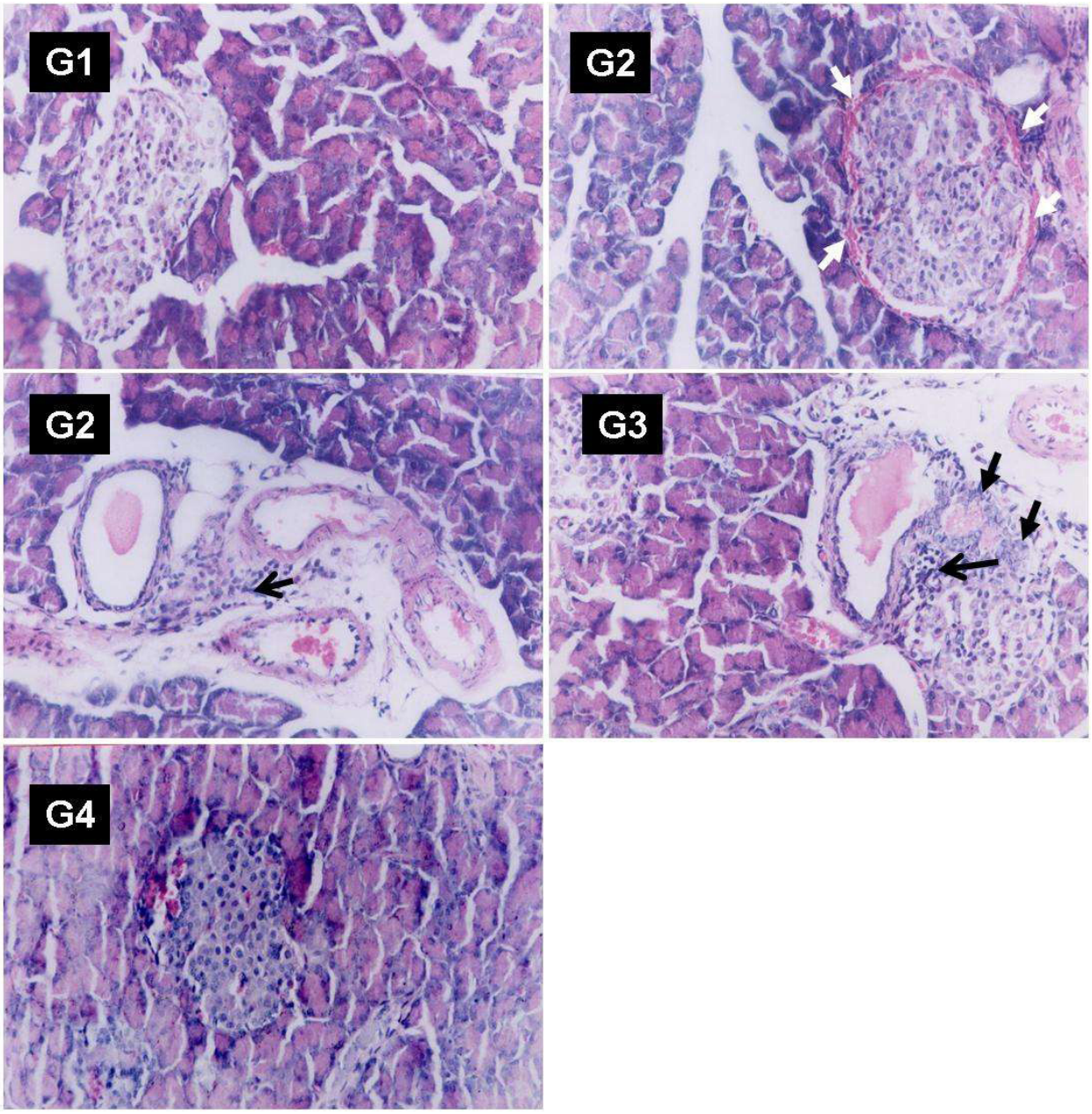

Fig. 3: Histopathological changes in the pancreases of the adult rats due to daily oral intake of 60 mg MSG (G2), 30 mg MSG + 30 mg 5´-IMP (G3) or 30 mg 5´-IMP (G4) / kg BW. The experimental period was 90 days, G1 presented the control. White arrow indicates haemorrhage in the cells of Langerhan's islets; black open arrow indicates leucocytic cells infiltration; black closed arrow indicates hyperplasia of epithelial lining pancreatic duct associated with few leucocytic cells infiltration (H&E, X200)

autonomic nervous system mediation. For instance, noradrenaline released from sympathetic nerves inhibit β cells which are crowded with β2-receptors (Lechin & van der Dijs, 2006). The hypertrophy of Langerhan's islets in the infant animals demonstrated the sensitivity of young rats to MSG toxic effect comparing with the adult rats.

Changes of exploratory behaviour due to oral intake of MSG

Exploratory behaviour has two components: Horizontal and vertical. Characteristic horizontal activity (motor activity) are walking, running and sniffing. Vertical activity of exploratory behaviour is manifested by attempts to turn the head up, before

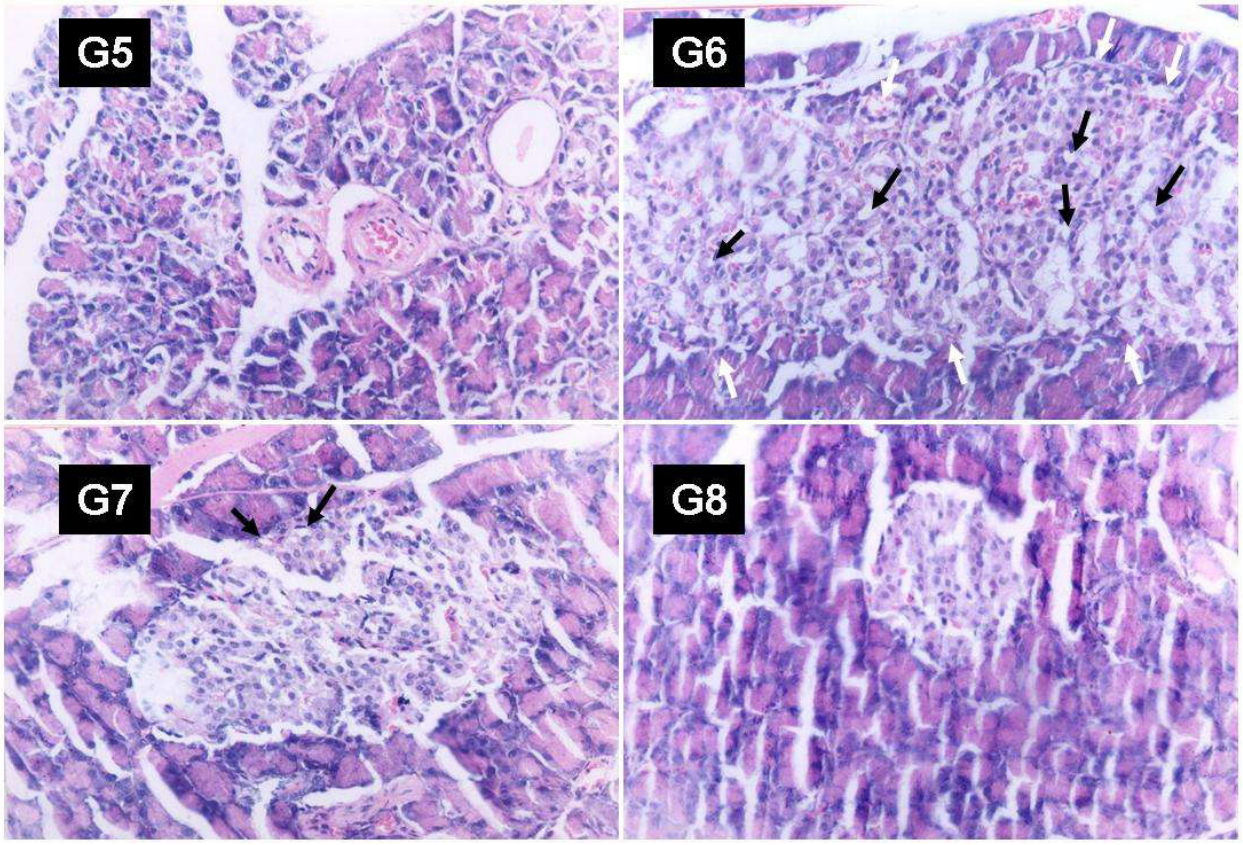

Fig. 4: Histopathological changes in the pancreases of the infant rats due to daily oral intake of 30 mg MSG (G6), 15 mg MSG + 15 mg 5´-IMP (G7) and 15 mg 5´-IMP (G8) / kg BW. The doses of MSG and 5´-IMP were when the weight of rats in infant class (G6, G7 and G8) reached to the weight of adult rats (7 weeks from the beginning of the experiment). The experimental period was 90 days, G5 presented the control (H&E, X200)

the animal is able to stand on its hind legs. All investigated rats displayed both types of behaviour. On day 45 (empty columns, Fig. 5), the motor activity, which expressed as number of squares traversed by rats in G2 and G3, was elevated as compared with control group (G1). However, insignificant difference ($P<0.05$) was noticed in the motor activity between rats in G1 and G4. Remarkable increase in motor activity was recorded by infant rats received MSG (G6) compared with other groups in infant class (Fig. 5). After 90 days (filled columns, Fig. 5), motor activity increased significantly ($P<0.05$) in G1 and G4 compared with its values on day 45. However, the adult rats received 60 mg MSG/ kg BW/ day showed remarkable decline in their motor activity. Also, the motor activities recorded by the infant rats in G6 and G7 on day 90 were found to decrease significantly compared with values recorded on day 45.

Differences in the vertical activity recorded on day 45 by the rats in G3 and G4 were less evident than those of G1 and G2 (empty columns, Fig. 5). On the other hand, no significant difference ($P<0.05$) in the vertical activity was obtained between infant rats on day 45. After 90 days, significant ($P < 0.05$) decrease in the vertical activity was noticed when the adult rats were fed on MSG alone (G2) or the mixture of MSG and 5´-IMP (G3). In infant rats, MSG had a negative effect on the vertical activity on day 90 compared with those obtained on day 45 (Fig.5).

The obtained data suggested that MSG induced different levels of neuronal lesions in the brain of both adult and infant rats. The low level of these changes was recorded when rats received the suggested mixture of MSG and 5´-IMP. The severe damages were visualized when MSG was applied alone. Moreover, these lesions were in different parts of the brain including the hyppocampus, hypothalamus and cerebral cortex. These areas of the brain in addition to NA are involved in locomotor activity (Gladfelter & Brobeck, 1962; Sławinska & Kasicki, 1998, Kramer *et al*. 2003). The obtained results suggested that the motor and vertical activity

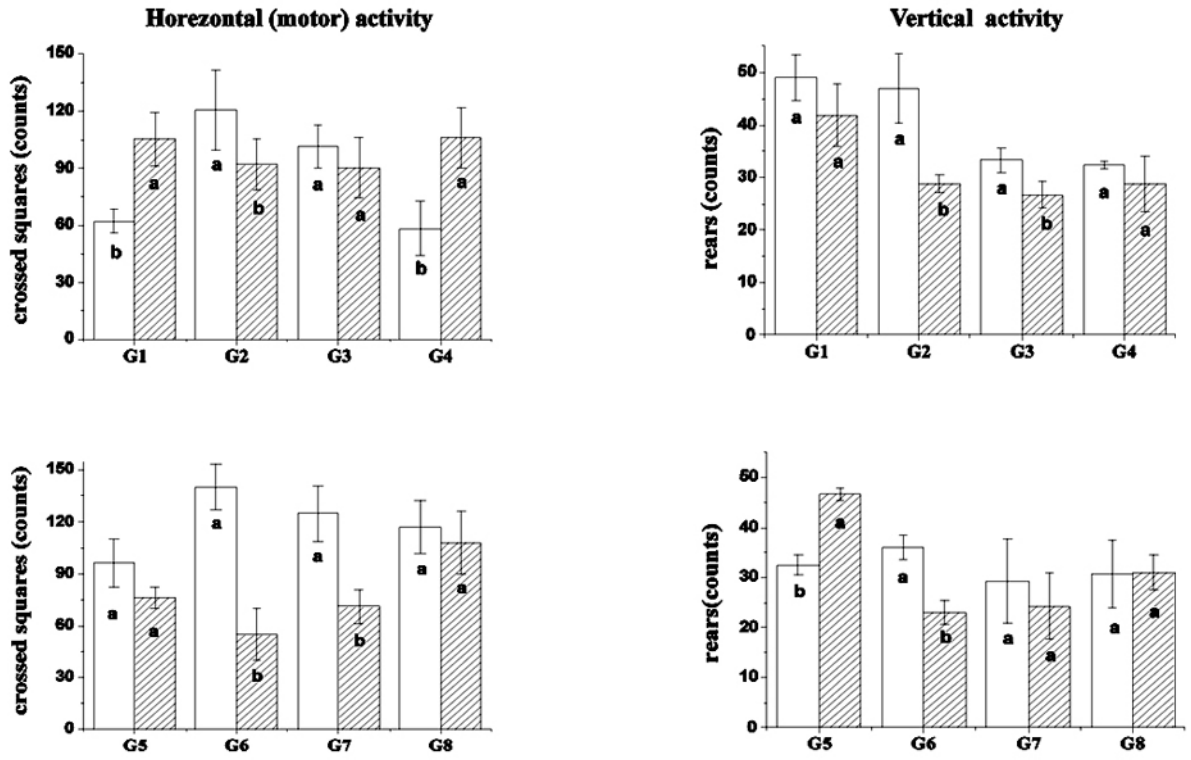

Fig.5: Exploratory behavior of adult (G1–G4) and infant (G5–G8) rats on day 45 □ and 90 ▨ after oral administration with MSG and/or 5´-IMP. The doses of MSG and 5´-IMP were doubled when the weight of rats in infant class (G6, G7 and G8) reached to the weight of adult rats (7 weeks from the beginning of the experiment). Columns with the different letter in the same group stand for significant difference (P ≤ 0.05) between the behaviour on day 45 and 90.

were reduced on day 90 compared with that recorded on day 45 due to treatment with MSG. Moreover, the infant rats appeared to be more vulnerable to MSG than the adult rats. These findings were in agreement with those described by Lorden & Caudle (1986) and Fisher *et al*. (1991). They observed hypoactivity in mouse and rats after MSG treatment. It is possible that structural and functional changes of brain resulted in low exploration because of poor locomotor coordination. Recently, Kiss *et al*. (2007) stated that neonatal MSG treatment leads to early temporary changes in the locomotor activity followed by hypoactivity at 2 months of age. On contrary, some other authors have described increased exploratory behaviour in the rats injected with MSG directly in the brain (Grimm & Frieder, 1985).

Generally, it can be demonstrated that the infant animals are more susceptible than the adults to the MSG toxic effect. This may possible because the amino acid intake mechanisms are not yet completely functional or because the blood-brain barrier has not been completely established, or both reasons combined in a newborn animal (Olney, 1980).

In summary, MSG induced hazards in adult and infant rats were reduced by the partial replacement with 5´-IPM. However, the effect of this ribonucleotide, as synergistic alternative flavour enhancers, on other internal organs such as kidney and liver should be also studied. Although use of 5´-IMP led to minimize MSG-induced hazards, it is not recommended to the free use in infant foods in the early stages of live.

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تأثير الإحلال الجزئي لجلوتامات أحادى الصوديوم بالإينوزين أحادى الفوسفات علي بعض الأعضاء والتوازن الهرموني ونشاط الفئران

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تم دراسة تأثيرات تناول جلوتامات أحادي الصوديوم (MSG) كأحد أشهر المواد المحسنة لنكهة الأغذية منفرداً او فى مخلوط مع الإينوزين أحادي الفوسفات (5′-IMP) على مدي إحداث اضرار بالمخ والبنكرياس و انخفاض النشاط الحركى و خلل إفراز هرمونات النورأدرينالين (NA) و الأنسولين (Ins) و ذلك على مجموعتين من فئران التجارب البالغة (عمر ١٠ أسابيع) و الفئران الصغيرة بعد مرحلة الفطام مباشرة (عمر ٣ أسابيع). وقد اتضح من خلال هذه الدراسة العديد من الأضرار فى مناطق مختلفة من المخ ضمت القشرة الخارجية و الهيبوثلاموس والفص ألامامى (الهيبوكامبوس) وذلك عند تناول جرعة مقدارها ٦٠ مجم من MSG لكل كجم من وزن الجسم يومياً. وقد تزامن مع هذه الأضرار حدوث إنخفاض فى نشاط السلوك الحركى فى كل من الفئران البالغة و فئران مرحلة ما بعد الفطام. و قد لوحظ أيضا حدوث خلل فى إفراز الهرمونات و الذى تمثل فى زيادة مستوى هرمون نورأدرينالين وانخفاض مستوى هرمون الأنسولين فى سيرم الدم و الذى أدى بدوره الى ارتفاع تركيز جلوكوز الدم. و قد أظهرت الدراسة التشريحية للبنكرياس حدوث تضخم فى جزر لانجرهانس ، نزيف دموى و ارتشاح خلايا الكريات البيضاء. و قد ادى استبدال نصف كمية MSG بـ 5′-IMP (مخلوط بنسبة ١:١ و/و) الى خفض معنوى ($P<0.05$) لهذه التغيرات غير المرغوبة السابق ذكرها. وبصفة عامة كانت الفئران الصغيرة أكثر حساسية لتأثيرات السمية المستحثة نتيجة تناول MSG. وتوصي الدراسة بتقليل كميات MSG المضافة الى أغذية البالغين بغرض تحسين النكهة و عدم استخدامها فى أغذية الأطفال. و بالرغم من ان الاستبدال الجزئى للـ MSG بـواسطة -5′ IMP أدى إلى تقليل مخاطر الـ MSG إلا أنه لا ينصح باستخدامه بلا حدود خاصة فى أغذية الأطفال كما ان هناك حاجة ضرورية إلى إجراء بعض دراسات مستقبلية لتأكيد أمان استخدام مخلوط الـ MSG و 5′-IMP المقترح فى هذه الدراسة و الذي ثبت من خلال التقييم الحسى لبرجر اللحم الذي أضيف له هذا المخلوط بنسبة ١:١ انه يؤدي إلى تحسين معنوى فى نكهة برجر اللحم مقارنة بالبرجر المضاف له الـ MSG فقط.