Low Dose Oral Administration of Monosodium Glutamate in Male Albino Rats May be Nephroprotective

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Abstract

The speculation that low dose intake of monosodium glutamate over time may be toxic warranted the present study. The aim was to investigate the effect of the administration of monosodium glutamate at a low concentration on the functional capacity of the kidney. Thus, monosodium glutamate at a dose of 5 mg/kg of body weight was administered to adult male albino rats by oral intubation. Treatment was daily for 28 days. The monosodium glutamate treatment significantly ($p < 0.05$) decreased the serum sodium ion concentration by 11.38 % and the water intake by 9.39 %, but had no apparent change in the serum potassium ion concentration (change, 0.00 %). The treatment increased ($p < 0.05$) the serum urea and creatinine concentration by 12.80 % and 107.81 % respectively. Therefore, treating rats with monosodium glutamate at a low concentration (5 mg/kg of body weight) could be nephroprotective, but with possible significant dehydration. The health implications of the results are highlighted in the discussion.

Keywords: Monosodium glutamate, Urea, Creatinine, Sodium ion, Potassium ion, Water intake

Introduction

Monosodium glutamate (MSG), the sodium salt of the amino acid glutamate, is a food additive, popularly used the world over as "flavour enhancer". It is marketed under such trade names including A-One, Ajinomoto or Vedan and is a popular condiment in West African dishes (Obaseiki-Ebor *et al.*, 2003). Additionally, MSG is abundant in yeast extracts and other food ingredients without otherwise appearing on the label (Healthnotes, 2005). Thus, it could be abused inadvertently.

The possible inadvertent abuse of MSG may have untoward results since the effects of MSG as reviewed in Truth In Labeling Campaign (TILC, 2004 a,b) include brain damaging potentials, stunted skeletal development, behavioral aberration, neuroendocrine disorder, possible learning deficits, seizures (epileptic fits), learned taste aversion and hyperglycemia. Added to these, MSG intake has been implicated in the Chinese restaurant syndrome manifested by migraine, diarrhoea, weakness, vomiting, stomachache and tightness of the chest (Schaumberg *et al,,* 1969; Healthnotes, 2005). Also, MSG intake could induce an increase in the energy intake (Bergen *et al.,* 1998), which could lead to obesity (Mozes *et al.,* 2004) or alter the levels of carbohydrates, lipids and proteins (Diniz *et al.,* 2004) in rats. Furthermore, MSG is an excitotoxin which may damage the brain especially by oral intake without food (Walker and Lupien (2000) yet, it is used mostly as condiment in meats, soups and vegetables without food protection like carbohydrate food.

These reported adverse effects of MSG indicate that the use of this as flavour enhancer over time may be hepatotoxic. Therefore, the present study aimed at determining whether MSG intake in low dose could be toxic on selected functional capacity of the kidney of male albino rats. We based our choice of MSG treatment dose on earlier reports (Olney, 1969; Goldberg, 1994; Onyema *et al.,* 2006).

Materials and Methods

Chemicals: Ajinomoto brand of MSG was purchased from a regular foodstuff market at Nsukka, South East of Nigeria. Other chemicals were of analytical grade.

Animals and treatment: The animals used in this work were adult male albino rats. They were procured from the animal house of the Faculty of Biological Sciences, University of Nigeria, Nsukka.

Eight adult male albino rats with mean body weight (BW) of 93±0.5 g were acclimatized for one week before they were randomly assigned to two groups of four rats each. Group II were fed MSG (5 mg/kg of BW) whereas Group I rats were given distilled water (1 ml/kg of BW). Treatment was by daily oral intubations and lasted for 28 days. The rats were housed in stainless steel cages at room temperature (25^{o}C) and exposed to a normal daylight/dark cycle under humid tropical condition. They were supplied with enough rat feed and drinking (tap) water ad libitum throughout the duration of the experiment. All the animals received humane care in accordance with the guidelines of the National Institute of Health, USA for ethical treatment of laboratory animals.

Blood collection and preparation: The animals were sacrificed 24 hours after the 28 days treatment. Their blood samples were collected individually with sterile capillary tubes into properly labeled polystyrene centrifuge tubes by ocular

Table 1: Effect of monosodium glutamate (MSG) treatment on the serum urea and serum creatinine concentrations of rats

Parameters	Serum creatinine concentration (Mg/100ml)		Serum urea concentration (Mg/100ml)	
	Water (I)	MSG (II)	Water (I)	MSG (II)
Mean ± SEM	1.92±0.05	3.99±0.05*	3.28±0.09	3.70±0.10*
Percentage	100	207.81	100	112.80
% difference		+107.81		+12.80

*The results show mean ± SEM, n = 4, * p<0.05, ANOVA followed by least significant difference (LSD) test, +: increased*

Table 2: Effect of monosodium glutamate (MSG) treatment on the serum sodium ion (Na^+) and serum potassium ion (K^+) concentrations of rats

Parameters	Serum sodium ion (Na^+) concentration (mmol/l)		Serum potassium ion (K^+) concentration (mmol/l)	
	Water (I)	MSG (II)	Water (I)	MSG (II)
Mean ± SEM	1081.00±2.67	958.00±1.41*	28.26±0.16	$28.26±0.16^{NS}$
Percentage	100	88.62	100	100.00
% difference		-11.38%		0.00%

*The results show mean ± SEM, n = 4, * p<0.05, ANOVA followed by least significant difference (LSD) test, -: decreased by, ^{NS} = Not significant.*

puncture technique of Scholm *et al.* (1975). The blood samples thus collected were allowed to clot and then centrifuged at 3000 rotor per minute (rpm) for 10 minutes. The resultant sera were collected individually into stoppered polystyrene tubes, stored in deep freezer for the determination of serum urea, creatinine, sodium ion (Na^+) and potassium ion (K^+) concentrations.

Flame emission photometric estimation of potassium ion (K^+) and sodium ion (Na^+) concentrations: The flame photometer was set in place and the meter reading normalized (brought down to zero) using de-ionized water. Potassium ion (K^+) and sodium ion (Na^+) standards of known concentrations were run through and their readings recorded. The serum samples were successively passed through the nebuliser, and their respective detector readings taken. The potassium ion and sodium ion concentrations were calculated from their respective standard curves.

Calculation of water intake: The water intake was calculated as the total daily water consumed relative to the body weight change as follows; Individual rat water intake = Total water consumed by a group divided by the total body weight change of the group multiplied by the individual rat body weight change.

Assay of serum creatinine concentration: Serum creatinine concentration was determined by the method of Wilding and Kennedy (Wilding and Kennedy, 1977). To 0.1 ml of serum sample in a test tube was added 0.8 ml of acid tungstate. The content was shaken and then centrifuged. Next, 0.6 ml of the resultant supernatant was transferred into another test tube and 0.6 ml of distilled water was added into the reagent blank. But in the standard tube 0.1 ml of serum was mixed with 0.6ml distilled water. Then 0.2 ml of picric acid and 0.1 ml of sodium hydroxide (NaOH) (1.4 mol/l) were added into each tube and the absorption read at 500 nm against the reagent blank.

Assay of serum urea concentration: The serum urea concentration was assayed by the method of Alexander and Griffith, (Alexander and Griffit, 1992) based on the principle that the ammonia from the urease catalyzed hydrolysis of urea (to ammonia and carbon dioxide) is converted to indophenol blue in the presence of sodium nitroferricyanide-phenol and hypochlorite reagents.

To each of the test tubes was added 1 ml of serum sample. Also, 1 ml of the standard was added to each of three different test tubes. Then, 1 ml of water was added to the content of each of the two sets of test tubes. To the blank test tube (with neither the serum sample nor the standard) was added 2 ml of water. Then, 2 ml of urease was added to each of the tubes (including the blank tube) and incubated in a water bath at 37 ºC for 30 minutes before adding 0.05 ml each of phenol reagent and hypochlorite reagent to all the tubes. Then the content of each tube was mixed and incubated at room temperature for 40 minutes. 4 ml of distilled water was added and thoroughly mixed before reading in a spectrophotometer at 625 nm against the blank.

Statistical analysis: The data obtained from the present study were expressed as mean ± SEM. They were analyzed by one-way analysis of variance (ANOVA) based on the modified method of Scheffe (Scheffe, 1952). Multiple comparisons of means were made using the least significant difference (LSD) test with the statistical package for social sciences (SPSS) for Windows version 11.0 package. Differences were considered significant at $p<0.05$ level of significance.

Results

Effect of monosodium glutamate (MSG) treatment on the serum urea and serum creatinine concentrations of rats: Significant ($p<0.05$) increase in the concentration of serum urea (12.80 % or 3.70 ± 0.10 mg/100ml), was observed in the MSG-treated rats compared to the control rats. The serum creatinine concentration was observed to increase (107.81% or 3.99 ± 0.05 mg/100ml representing more than one fold) in the MSG-treated rats compared to the control rats (Table 1).

Effect of monosodium glutamate (MSG) treatment on the serum sodium ion and serum potassium ion concentrations of rats: The decrease in the serum sodium ion (11.38% or 958.00±1.41 mmol/l) observed in group II (MSG group) was significant ($p<0.05$), whereas the no apparent change in the serum potassium ion concentration observed in group D (MSG) (0.00% or 28.26±0.16 mmol/l) was insignificant ($p>0.05$) (Table 2).

Effect of monosodium glutamate (MSG) treatment on the water intake of rats: Furthermore, Table 3 shows that the water intake decreased by 0.08 i.e. 9.39% or 0.67±0.00 liters) ($p<0.05$) in the MSG-treated rats when compared to the control rats (0.75 ± 0.00 liters).

Table 3: Effect of monosodium glutamate (MSG) treatment on the water intake of rats

Parameters	Water intake (Liters)	
	Water (I)	MSG (II)
Mean ± SEM	0.75±0.00	0.67±0.00*
Percentage	100	89.33
% difference	-	-10.67%

*The results show mean ± SEM, n = 4, * p<0.05, ANOVA followed by least significant difference (LSD) test, -: decreased*

Discussion

This study was necessitated by the observed indiscriminate use of monosodium glutamate (MSG) in Nigeria for seasoning/flavour enhancing purposes without minding the possible adverse effects from its use over time at a low concentration.

Water is a key element in the removal of wastes (Batmanghelidj, 2006) and its intake is mainly controlled by thirst (Akio, 2000), antidiuretic hormone (ADH) and retention or excretion of water by the kidneys (Rodwell and Kennelly, 2003). Thus, the decreased water intake in the MSG-fed rats (10.67%) in the present study probably indicates the absence of thirst, perhaps due to decreased excretion (or increased retention) of water or enhanced effect of the antidiuretic hormone. This may be a pointer to the possible anti-diuretic potential of MSG at 5 mg/kg BW that may result to a decrease in the excretion of urea.

The possible anti-diuretic potential of MSG at 5 mg/kg BW may be beneficial in the management of situations (including non-insulin dependent diabetes mellitus, NIDDM) where fluid retention or decreased secretion of urine may be desired. However, the possible anti-diuretic potential of MSG at the tested dose may not be beneficial in the formulation of diuretics hence could not be useful in such cases as poisoning and fluid retention where increased secretion of urine may be desired.

Creatinine is derived from the muscle metabolism, where it arises by spontaneous (nonenzymatic) and irreversible dehydration, loss of phosphate and cyclization of creatine phosphate. The creatinine thus formed can no longer be phosphorylated hence is excreted with urine (Koolman and Roehm, 2005) produced via urea synthesis.

The observed increase in the serum urea concentration by 12.80% and in the serum creatinine concentration by 107.81% in rats that were fed MSG in the present study probably indicates an enhanced renal capacity to excrete urea as suggested by Panda (Panda, 1989). In addition, the results may be indicative of the possible adverse effects of MSG on protein metabolism (deamination) since urea and creatinine are the metabolic waste products of protein catabolism that are excreted in normal urine. MSG-induced elevation of the serum urea and creatinine concentrations indicates possible up-regulation of protein catabolism and the concomitant increase in the excretion of urea and creatinine which may increase thirst thereby leading to increased water intake. These however, could neither explain nor agree with the observation in the present study that water intake decreased in MSG-fed rats, probably due to absence of thirst and this suggests that MSG-induced increase in urea synthesis may not be thirst related hence may lead to dehydration. MSG could easily dissociate into glutamate the deamination of which may produce the toxic ammonium ion (NH_4^+) that must be detoxified and excreted via the reactions of the urea cycle. Thus, the increased serum urea and creatinine concentrations by MSG could therefore be attributed to the possible up regulation of protein catabolism due perhaps to the enhanced production of toxic NH_4^+ which must be excreted via urine produced during the urea cycle so as not to accumulate and become toxic. Furthermore, since urea synthesis converts toxic NH_4^+ to non toxic urea, the possible beneficial effect in the urea cycle as shown by the increased concentrations of urea and creatinine observed in the MSG-fed rats could lead to efficient ammonium ion detoxification. Thus, the use of MSG may be of benefit in cases of renal disorders.

In the present study, the serum K^+ was observed to be unaffected by the MSG treatment in rats, probably indicating absence of either muscular and cardiac disorders (Bush, 1991) or the consequent effects (such as thirst) of diuretic therapy (Johnston, 1999). In the present study, water intake decreased in MSG-fed rats, possibly due to absence of thirst, thus the possibly increased water loss as indicated by the increased serum concentrations of creatinine and urea may not be due to the consequent effects (such as thirst) of diuretic therapy as reported by Johnston (1999). However, the observed effect of MSG on the serum K^+ was not significant ($p>0.05$) hence may be indicating the influence of extraneous factors beyond the control of the present study.

The serum sodium ion concentration was observed to decrease in the MSG-treated rats by 11.38%. This may indicate decreased sodium intake, severe dehydration or diabetes mellitus [24]. The decrease in the serum Na^+ concentration observed in the MSG-fed rats was not as expected since MSG has sodium moiety in its chemical composition which could be easily ionized to release the sodium constituent with a possible resultant increase in the serum Na^+ concentration.

However, severe dehydration is likely going by the report in the present study of an increased serum creatinine and urea concentrations that may enhance the renal capacity to synthesize and excrete more urea leading to increased water loss and consequent dehydration. Furthermore, the observed decrease in the water intake in the MSG-fed rats in this study indicates that the water loss via increased urea synthesis was not replenished which could lead to possible dehydration.

In conclusion, treating male albino rats with MSG at a low concentration (5 mg/kg of BW) could be nephroprotective but with possible significant dehydration. Altogether, the results may have significant underlying clinical and public health implications hence warrant further investigation in humans.

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