The addictive behaviour induced by food monosodium glutamate. Experimental study

Anca BUZESCU, Aurelia Nicoleta CRISTEA, Luminița AVRAM, Cornel CHIRIȚĂ

Department of Pharmacology and Clinical Pharmacy, Faculty of Pharmacy,
„Carol Davila" University of Medicine and Pharmacy, Bucharest

◆

ABSTRACT

Introduction. *Glutamic acid under the form of monosodium salt (monosodium glutamate) is regulated in the EU as a food additive (E 621), a flavoring agent, to give the so-called „umami" taste. Endogen glutamic acid is the major excitatory neurotransmitter of the central nervous system. Cortico-striatal glutamatergic transmission has been implicated in both the initiation and expression of addiction-related behaviors. In this paper, we have proposed to examine the extent to which food addictive behavior manifests itself in experimental animals, after repeated consumption of monosodium glutamate (MSG).*

Materials and methods. *We have experimented on NMRI adult mice. Monosodium glutamate (MSG) was given daily, dissolved in water in the drinking bottles, for three weeks. Three doses of MSG were tested. Each lot of mice had two identical drinking bottles, one with MSG solution, another with plain water. The consumption of MSG solution was measured daily, together with the consumption of water, for all working groups.*

Results. *The mice consumed preferentially the solution of monosodium glutamate (MSG), not the plain water. The greatest increase in MSG solution consumption, 71,02% (p<0.0001), was noticed in the group receiveing the highest dose (=1/10 DL50/mice).*

Conclusions. *We experimentally demonstrated that monosodium glutamate (MSG) may induce an addictive alimentary behavior.*

Key words: monosodium salt of glutamic acid (MSG, E 621), addictive behaviour, NMRI mice

INTRODUCTION

Glutamic acid (Glu) is regulated in the EU as a food additive, flavor enhancer, because it gives the so-called „umami" taste (etim. jap. = delicious). It can be used by itself (E 620), or as salts: monosodium glutamate (MSG - E621), monopotassium glutamate (E 622), calcium diglutamate (E 623), ammonium glutamate (E 624), magnesium diglutamate (E 625) (1). These additives are extensively used in Chinese and Japanese cuisine and in fast food. The glutamic acid's most frequently used salt is MSG (E 621). Romanian legislation on food safety, harmonized with EU legislation, including EC Regulation 178/2002 on Food Additives For Use In Foods For Human Consumption No 438/295 of July 2002, published in the Official Gazette, Part 1 No. 722, of October 3 2002, allowes the addition of no more than 10 g/kg food (= 1 g%) of MSG (E621). In spices, MSG can be added quantum satis (in the amount which is needed). The use of MSG

(E621) is not legally permitted in foods for infants and young children.

Since there are no official reports of severe adverse effects, glutamic acid and its' salts are considered safe additives by the Joint Committee of WHO experts and F.A.Q. However, there have been people who have accused a number of symptoms associated with MSG consumption such as: dizziness, headache, weakness, numbness, tremors, palpitations (known as „Chinese restaurant syndrome"), or nausea, respiratory problems, excessive sweating, hyperactivity, sudden mood changes, panic attacks (1,2). But, to date, a clear *relationship between MSG ingestion and the development of these conditions hasn't been established (3). Yet a* double-blind, placebo-controlled, crossover study with the purpose to examine the effect of a single intake and repeated monosodium glutamate (MSG) intake revealed that a single intake of MSG may cause headache and increased muscle sensitivity and repeated intake of MSG induced mechanical sensitization in masseter muscle and adverse effects such as headache and short-lasting blood pressure elevation for which tolerance did not develop over 5 days of MSG intake (4).

In animal experiments, administration of monoglutamate sodium in rat pups resulted in neurotoxicity affecting certain brain areas, such as arcuate nucleus, together with endocrine and behavioural dysfunctions (5,6) and also in retinotoxicity (7,8).

Given these informations, the consequences of chronic consumption of glutamic acid salts due to their frequent use as food additive, should be further investigated. The fact that glutamate, associated with food, may lead to a „consumer loyalty" due to the „umami" taste, generating an addictive alimentary behavior, should be considered.

Furthermore, in a diet with a high number of products containing MSG as additive (meat, poultry, processed food, sauces, soups, marinades, juice, refreshing drinks, etc.), it is possible for an individual to reach and even surpass a MSG dose of maximum 150 mg/kg/day. In this situation, of unintentional abuse, the major problem arises from the fact that glutamic acid (Glu) is the major neurotransmitter of the glutamatergic transmission, the main central excitatory aminoacidergic transmission (9-11). It has a large number of physiological roles such as: trophic role during ontogenesis of CNS, involvement in memory and learning; but also the role in stimulating the release of dopamine in the striatum, with implication in addiction (9,12).

The excitatory role of the glutamic acid is essential from the early development of the brain, but an excessive activation of this transmission can lead to neural degeneration and death. The overstimulation of glutamatergic receptors determines pathological neurotoxicity, probably through a high increase of calcium influx into the neurons and an excesive intraneuronal calcium build-up (13,14) and formation of a large number of free radicals. Death through ischemia arises, phenomenon which is considered to be the underlying cause of neurodegenerative ilnesses such as Alzheimer disease, Parkinson's disease, Huntington chorea, Lateral Amiotrophic Sclerosis (9,12).

Glutamic acid homeostasis imbalance generates neuroplasticity changes, especially in the dopaminergic neurons. This affects communication between the prefrontal cortex and nucleus accumbens (15,16). Neuroplasticity in corticostriatal circuits is associated with dependence, and it is an important phenomenon for addictive behavior (17,18).

Given the above-mentioned data on the involvement of glutamatergic transmission in addiction and the enhanced use of glutamic acid and its' salts as food additives (E620-E625), the aim of this research is focused on investigating the addictive potential behaviour of MSG. MSG was dissolved into the drinining water, at experimental animals.

MATERIALS AND METHODS

Reagents
Monosodium glutamate (MSG = E 621) was purchased from Sigma-Aldrich.

Experimental animals
48 NMRI mice, adults weighing 20-25 g from UMF biobase, Bucharest, were used. The mice were housed in plastic cages, in an air-conditioned animal room, with free access to water and granulated food. The temperature was between 20°C and 22°C and the relative humidity was maintained at 35-45%.

All procedures were carried out in accordance with the Directive 86/609/EEC of 24th November 1986 and The Romanian Government Ordinance 37/30.01.2002, regarding the protection of animals used for experimental and other scientific purposes.

Experimental protocol
The animals were divided in four groups of 12 mice, weighting 32 ± 5 g/group. During the 7

days accomodation period, the average water consumption was evaluted for each group as 150 ml/day. During the experiment, for 25 days, the mice from every cage received daily 2 bottles of liquid A and B, each bottle containing 200 ml (to ensure a possible increase in liquid consumption over 150 ml), as it follows:

- Group I: two bottles, A and B, each containing 200 ml water;
- Groups II, III, IV: two bottles, one A containing 200 ml of MSG solution and one B, containing 200 ml water.

Taking into account of the DL50 for MSG in mice ($DL_{50} = 15,000$ mg/kg = 15 g/kg), the concentration of MSG solution from the bottles „A" was calculated to ensure the administration of MSG daily doses of maximum:

- *Group II*: $D_1 = 1/10$ DL_{50}/animal;
- *Group III*: $D_2 = 1/20$ DL_{50}/animal;
- *Group IV*: $D_3 = 1/40$ DL_{50}/animal.

The liquid consumption was determined every day, at an exact interval of 23 hours and 30 minutes, using precise measuring instruments.

The bottles, type A and B, were kept the same for each group and they always had the same position (left or right) and the same type of content (water or glutamate solution), in order to mantain the specific odour of the group, to ensure freedom of rapid acces, without fear and for creating a conditioned reflex regarding the location of the bottle with glutamate solution.

Statistic analysis was made using the soft GraphPad Prism vers. 5 (Graphpad Software, San Diego California USA).

RESULTS

The average liquid consumption/group for 25 days, from each bottle, is presented in Table 1.

The percentage differences in the liquid consumption between bottle A and B are ilustrated in Figures 1 and 2.

Percentual change in average liquid consumption from both bottles (A + B), compared to group I is presented in Table 2.
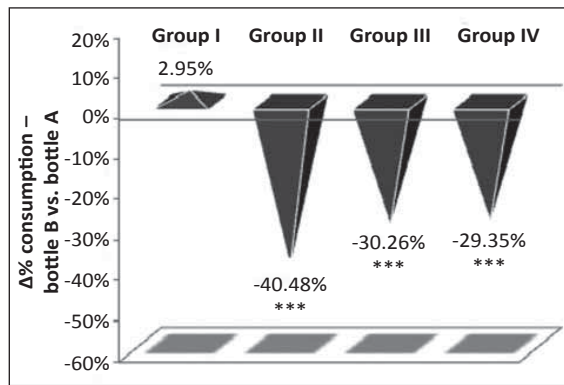

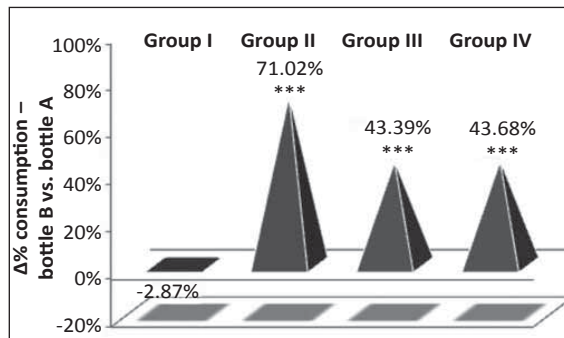FIGURE 1. *Percentual change of the liquid consumption – bottle B compared with bottle A*

FIGURE 2. *Percentual change of the liquid consumption – bottle A compared with bottle B*

The percentual change in average liquid consumption from each of the bottles A and B compared to group I (control), is illustrated in Figure 3.
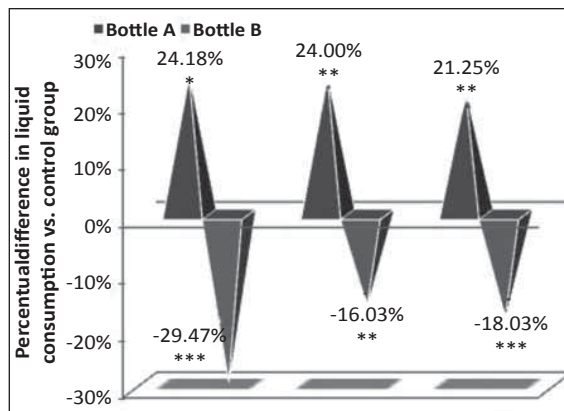

FIGURE 3. *Percentual change of the liquid consumption from bottles A and B, compared with group I*

TABLE 1. *Medium liquid consumption/bottle/group in 25 days of exposure*

	Liquid consumption (average volume/bottle/group)							
	Group I (control)		Group II		Group III		Group IV	
	Bottle A (ml)	Bottle B (ml)	Bottle A (ml)	Bottle B (ml)	Bottle A (ml)	Bottle B (ml)	Bottle A (ml)	Bottle B (ml)
Average±DS	74.11 ± 31.46	76.30 ± 21.79	92.03 ± 21.84	53.81 ± 26.52	91.89 ± 18.93	64.08 ± 22.99	89.86 ± 12.37	62.54 ± 23.89
SEM	5.15	3.58	3.26	4.36	3.16	3.78	1.75	3.93
Dn D'Agostino Pearson	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
DS – standard deviation; Dn – normal distribution; SE – standard medium error								

TABLE 2. *Total average liquid consumption (bottle A+B) during 25 days of study*

	Liquid consumption (average volume/bottles A+B/group)			
	Group I	Group II	Group III	Group IV
Average±DS	150.4 ± 40.79	145.8 ± 30.52	157.4 ± 28.10	152.4 ± 27.18
SEM	6,71	5.02	4.62	4.47
Dn D'Agostino Pearson test	Yes	Yes	Yes	Yes
Δ% – vs. control group		- 3.03%	4.65%	1.32%
p		0.3049 **ns**	0.1103 **ns**	0.6728 **ns**

DS – standard deviation; SEM – standard medium error; Dn – normal distribution Δ – change; * – statistically significant, ** – with high statistical significance, *** – with extremely high statistical significance, ns – with no statistical significance

DISCUSSIONS

The consumption of water form bottle B decreased *statistically significant (p < 0.0001), while the consumption of monosodium glutamate (MSG) solution from bottle A increased statistically significant (p < 0.0001), for all three test groups (II, III, IV), as seen in Figures 1 and 2. The intensity of the effect is in a direct relation with the concentration of the solution of MSG given (the greatest increase in MSG consumption, 71.02%, is noticed in the group receiveing the highest MSG dose (corresponding at 1/10 DL$_{50}$/mice). For the control group, group I, the difference of consumption between the two bottles is minimum, having no statistical significance (p > 0.05).* On what concerns the total average liquid consumption, the variations between all four groups have no statistical difference (as showed in Table 2). In comparison with the control group, the consumption of water form bottle B decreased *statistically significant, while the consumption of MSG solution from bottle A increased statistically significant for all three test groups (II, III, IV), illustrated in Figure 3. Again, a direct relation between the intensity of the effect and the concentration of the given MSG solution can be observed (the greatest variations, a 24,18% increase of MSG solution consumption and a –29.47% decrease in water consumption, are noticed between control group I and group II, the group receiveing the most concentrate solution of glutamate equal to 1/10 DL$_{50}$/mice).*

*These results indicate that monosodium glutamat (*MSG), used as food additive (E 621), may influence eating behavior, inducing a «loyalty» for glutamate enriched-food and an addictive behavior. One of the underlying causes of this phenomenon may be the fact that MSG activates chorda tympani (CT) neurons, by stimulating separate receptors for Na$^+$, sugars and glutamate, in taste bud cells (2). The specific savoury taste („umami") is elicited by both components of MSG: glutamate anion and Na$^+$ cation (17). It must be remembered also that glutamatergic plasticity in the nucleus accumbens is critical for the expression of the behaviors related to addiction (18,19). Desensibilization of D2 receptors and sensibilization of glutamatergic pathways are linked at cellular level and are considered to be molecular mechanisms of conditioning and drug addiction (20-22). Primary rewards like food and water, when presented in an unexpected manner, are among the most effective stimuli for dopamine neurons in the ventral tegmental area (VTA) (23) which receive a major excitatory response from the prefrontal cortex, and also other brain regions (24), the major neurotransmission involved being the glutamatergic transmission (25). The evidence supports the hypothesis that VTA neural cells are involved in reward based learning, like the one involved in drug-addiction (25).

CONCLUSIONS

The purpose of this experimental research was to investigate the hypothesis that sodium monoglutamate (MSG), used as food additive (E621), may influence eating behavior, inducing a «loyalty» for glutamate enriched-food and an addictive behavior.

Statistically significant increase in the consumption of aqueous solution of MSG from bottles A, accompanied by a significant decrease in water consumption from bottles B, for the three groups exposed to MSG, but not for the control group, and also a similar daily total fluid intake for all 4 groups, verified the working hypothesis of an addictive effect of MSG used as additive in food.

This conclusion resulting from this animal experimental research, together with the re-

search presented in the introduction and in discussions of the paper, highlighting the involvement of endogenous glutamatergic system in the phenomenon of addiction, raises the question of obvious negative report financial-benefit/adverse effects cumulative time for MSG as a food additive.

REFERENCES

1. **Ciurea A.V., Edu F.V.** Probleme de nocivitate în alimentele uzuale. *Edit. Galaxia Gutenberg*, Târgu-Lăpuş, 2011: 140-141.

2. **Bush R.K., Taylor S.L.** Adverse reactions to food and drug additives. In: Atkinson NF Jr, ed. *Middleton's Allergy: Principles and Practice*. 7th ed. Philadelphia, Pa: Mosby Elsevier; 2008:866-894.

3. **Williams A.N., Woessner K.M.** Monosodium glutamate „allergy": menace or myth? *Exp. Allergy*. 2009; 39 (5):640-6.

4. **Shimada A., Cairns B.E., Vad N., Ulriksen K., Lynge Pedersen A.M., Svensson P., Baad-Hansen L.** Headache and mechanical sensitization of human pericranial muscles after repeated intake of monosodium glutamate (MSG). *The Journal of Headache and Pain* 2013; 14:2.

5. **Aravich P., Sladek C., Clough R.** Monosodium glutamate neurotoxicity: A sex-specific impairment of blood pressure but not vasopressin in developing rats. *Brain Res. Bulletin.* 1986; 17(1): 51–58.

6. **Meister B., Ceccatelli S., Hokfelt T.** Neurotransmitters, neuropeptides and binding sites in the rat mediobasal hypothalamus: effects of monosodium glutamate (MSG) lesions. *Exp. Brain Res.* 1989; 76:343-368.

7. **Ohguro H., Katsushima H., Maruyama I.** A high dietary intake of sodium glutamate as flavoring (Ajinomoto) causes gross changes in retinal morphology and function. *Exp. Eye Res.* 2002; 75(3):307-15.

8. **Szabadfi K., Atlasz T., Horvath G.** Early postnatal enriched environment decreases retinal degeneration induced by monosodium glutamate treatment in rats. *Brain Res.* 2009; 1259:107-12.

9. **Cristea A.N.,** Farmacologie generală. Edit. Didactică şi Pedagogică, Bucureşti, Ediția a II-a, 2009. Subcap. „Transmisia glutamatergică" (Chiriță C.): 424-428.

10. **Cotman C.W., Kahle J.S., Miller S.E.** Excitatory amino acid neurotransmission. In Psychopharmacology: The Fourth Generation of Progress (Bloom F.E. and Kupfer D.J., eds.), Raven Press, New York, 1995: 75-95.

11. **Mahler M., Rafiki A., Represa A.** Acides amines excitateurs: les voies glutamatergiques et leurs principaux types de récepteurs dans le système nerveux central. In „Neuropeptides et neuromédiateurs", 2ᵉ ed., Ed. Sandoz Rueil-Malmaison, 1995: 27-45.

12. **Nakanishi S.** Molecular diversity of glutamate receptors and implications for brain function. *Science*. 1992; 258: 597-603.

13. **Porter A.G., Dhakshinamoorthy S.** Apoptosis initiated by dependence receptors: a new paradigm for cell death? *Bioessays*. 2004; 26: 656-664.

14. **Danysz W., Parsons C.G., Mobius H.J.** Neuroprotective and symptomatological action of memantine relevant for Alzheimer's disease--a unified glutamatergic hypothesis on the mechanism of action. *Neurotox Res.* 2000; 2(2-3):85-97.

15. **Miyamoto Y., Yamada K., Noda Y.** Hyperfunction of Dopaminergic and Serotonergic Neuronal Systems in Mice Lacking the NMDA Receptor E1 Subunit. *The Journ. of Neuroscience*. 2001; 21(2): 750-757.

16. **Paz H.R.** Modelos fisiopatológicos de la esquizofrenia; de dopamina a glutamato, de glutamato a GABA. *Rev Chil Neuro-Psiquiat.* 2005, 43 (4): 314-328.

17. **Heyer B.R., Taylor-Burds C.C., Tran L.H., Delay E.R.** Monosodium glutamate and sweet taste: generalization of conditioned taste aversion. *Chem Senses.* 2003; 28(7):631-41.

18. **Guo Y., Wang H.L., Xiang X.H., Zhao Y.** The role of glutamate and its receptors in mesocorticolimbic dopaminergic regions in opioid addiction, *Neurosc. and Biobehav. Rev.* 2009; (33): 864-873.

19. **Pshenichkin S., Dolińska M., Klauzińska M.** Dual neurotoxic and neuroprotective role of metabotropic glutamate receptor 1, in conditions of trophic deprivation – possible role as a dependence receptor. *Neuropharmacol.* 2008; 55(4): 500-508.

20. **Self D.W., Nestler E.J.** Molecular Mechanisms of Drug Reinforcement and Addiction, *Annu Rev of Neuroscience.* 1995; 18: 463-495.

21. **Wise R.A.** Addictive Drugs and Brain Stimulation Reward, *Annu Rev of Neurosci.* 1996; 19: 319-340.

22. **Di Chiara G.** The role of dopamine in drug abuse viewed from the perspective of its role in motivation. *Drug Alcohol Depend.* 1995; 38: 95-137.

23. **Mirenowicz J., Schultz W.** Preferential activation of midbrain dopamine neurons by appetitive rather than aversive stimuli. *Nature. 1996; 379.*449-451.

24. **White F.J.** Synaptic regulation of mesocorticolimbic dopamine neurons. *Annu Rev Neurosci.* 1996; 19: 405-436.

25. **Gonon F., Sundström L.** Excitatory effects of dopamine released by impulse flow in the rat nucleus accumbens in vivo. *Neurosci.* 1996; 75:13-18.