Monosodium Glutamate 'Essence of taste' or a 'Health hazard': An Assessment through the degree of aberrations observed in *Urginea indica* Kunth Cytotype I

Richa Sinha
AMITY University Jharkhand
Ranchi, India

Abstract: Food additives are the substances which enhance the flavor, color, texture or other quality of the food. Safety of the food additives is approved by the FDA, but they may not be entirely safe. Such food additives have induced allergic reactions, while others may have carcinogenic effects or birth defects. The safety of food additives has long been a subject of fiery debate, between those who believe food additives are harmless and those who believe that they are silent killer. Monosodium Glutamate is one of the most common food additives which is used to enhance the flavor of the food, especially in Asian cuisines. It is an ingredient used in some packaged foods and restaurant food preparation. MSG commonly called 'Ajinomoto' is a white odorless crystalline powder that is soluble in water and alcohol. It is believed to cause potential reaction in some people, like flushing, sweating, chest pain, weakness, headache, facial pressure, drowsiness, and numbness and tingling in the face, back, and arms. It is also reported to causes a high frequency of clastogenic and carcinogenic effects on plants. But till date the research has not shown the allergic or any such symptoms on MSG consumption. In fact Food Safety and Standards Authority of India has certified Ajinomoto, while US Food and Drug Administration has given MSG as "generally recognized as safe". This clearly shows that neither side had the right information. Thus, the consumption of MSG is safe or not is the matter of massive research.

Therefore, in the present research, the effects of Monosodium Glutamate (MSG) on the root tip cells of Urginea indica Kunth Cytotype I were studied. The principle objective of this research work was to find out the mutagenic effects of MSG on Urginea indica Kunth. Plant system is reported to display numerous genetic and chromosome changes to determine the effects of mutagens. Moreover, plants also play an important role in various aspects of mutagenesis research. The effect of induced mutagens in plants can be estimated by the observation of frequency of chromosomal abnormalities induced in them by the mutagens. Chromosome aberrations are used as a monitoring system to determine the clastogenic nature (capacity to break chromosomes) of the mutagens involved. Observations on the root tip system of plants are considered a rapid and sensitive method for chromosomal monitoring. The degree of cytological aberrations in mitosis is regarded as one of the dependable criteria for estimating the mutagen sensitivity of the species and the effect of the mutagens.

Monosodium Glutamate was reported to induce various types of chromosomal aberrations in Urginea indica Kunth which increased with the increase in concentrations. This revealed the antimitotic and carcinogenic properties of Monosodium Glutamate. The result thus, clearly revealed the harmful effects of MSG on plants and hence on human beings.

Key words: Monosodium Glutamate, *Urginea indica* Kunth, chromosomal aberrations

I. INTRODUCTION

Monosodium glutamate is the sodium salt of amino acids found naturally in many foods and can also be added to food to enhance flavor. It is a white odorless crystalline powder that is soluble in water/alcohol, and is commonly called 'Ajinomoto'. MSG is a food additive widely used as a flavor enhancer of many foods and has long been known for its use in restaurants, frozen meals, packaged snack foods, canned foods and soups, and even seasoning mixes. It is the most stable salt formed from glutamic acid, having distinctive taste known as "umami," a word coined by the Japanese to describe the taste imparted by glutamate. It excites nerve cells in order to relay its signal and hence is referred to as an excitotoxin.

Currently, MSG is widely used in over 130 countries. In India, the market for MSG is 10,500 tons per year and of this 90 per cent is from Chinese companies. Nevertheless, today we are confronted with number of diseases due to the addition of additives and flavor enhancer in our food. Although, MSG has been approved by the FDA, and is considered fit for human consumption, it may not be entirely safe. It has long been popularly linked to various health problems, such as headaches and allergic reactions. The Punjab Food Authority has already banned Ajinomoto, declaring the MSG salt as hazardous to health. The PFA's scientists' panel had declared that the MSG is a harmful chemical which causes health problems which may be lethal sometimes. According to the findings of the scientific panel, MSG can also cause fatigue, palpitations, nausea and vomiting, sweating, flushing and numbness of the face — more so to people who have sensitivity to it. On the other hand Food Safety and Standards Authority of India has certified Ajinomoto while US Food and Drug Administration has given MSG as "generally recognized as safe". This certainly has led to the fiery debate between the scientists who believe in the harmful effects of MSG

and among them who considers MSG as entirely safe flavor enhancer. Even though it has been used extensively for nearly a century, it continues to be examined in light of current scientific knowledge and methods of testing.

Therefore, in the present research, an attempt has been made to find out the effects of MSG on plant which can relay the harmful effects of MSG on human health. For this purpose *Urginea indica* Kunth, a wild poisonous herb belonging to the Liliaceae family, has been selected. *Urginea indica* Kunth is extremely valuable in pharmacology. It is one of the ancient medicinal plants with high therapeutic values and is commonly called Indian Squill or van pyaz.

The selected cytotype of *Urginea indica* Kunth was named as *Urginea indica* Kunth Cytotype I which was treated with different concentrations of Monosodium Glutamate (MSG). Differences in the mitotic index and percentage chromosomal abnormalities at all the stages of mitotic cell divisions after the exposure to different concentrations (0.1, 0.2, 0.3, 0.4 and 0.5) of Monosodium Glutamate (MSG) were statistically analyzed.

II. RESEARCH METHODOLOGY

Bulbs of *Urginea indica* Kunth Cytotype I was collected from Birsa Agriculture University, Ranchi, Jharkhand. Five different concentrations of Monosodium Glutamate (MSG) were prepared. All the concentrations were prepared under aseptic conditions. Fresh and healthy *Urginea indica* Kunth bulbs of uniform size were treated with different concentrations of Monosodium Glutamate (MSG) for six hours. Then the treated bulbs were thoroughly washed in running tap water to remove the residual effect of the chemicals used. The control and the treated bulbs were grown in the experimental plots in the randomized block design to raise M_1 generation.

After each treatment bulbs of *Urginea indica* Kunth Cytotype I were allowed to root. Healthy root tips were excised from the bulbs, and pretreated in the saturated solution of para dichlorobenzene for four hours, fixed in fixative (1:3 acetic alcohols) for twenty four hours and then transferred to 70% alcohol for preservation. The root tips were warmed in 2 per cent acetocarmine and slides were prepared by squash techniques. Photomicrographs of the most frequent chromosomal abnormalities were taken. Approximately 700 to 800 cells were counted for determining mitotic index (number of cells in mitosis/total number of cells counted and expressed as percentage) and the frequency of mitotic phases. Also the number and type of chromosomal abnormalities were recorded. The obtained data were statistically analyzed.

III. RESULTS AND DISCUSSION

The effects of Monosodium Glutamate (MSG) on the root tip cells of the selected cytotype of *Urginea indica* Kunth were studied.

Mitotic index was reported to reduce considerably in dose dependent manner and the percentage of abnormal cells significantly increased at all the concentrations. Various types of chromosomal aberrations were induced by Monosodium Glutamate which increased with the increase in concentrations. Maximum aberrations were induced at metaphase stage followed by anaphase and subsequently at telophase stage. In contrast, chromosomal aberrations were not observed at prophase stage. The chromosomal aberrations observed at metaphase stage were precocious movement of chromosomes, sticky metaphase, ball metaphase, tropokinesis, nucleolus at metaphase, diagonal metaphase, polyploidy, chromosome fragments and translocation ring. While disturbed anaphase, laggards, bridges, multipolar anaphase and unequal separations were observed at anaphase stage. The chromosomal aberrations reported at telophase stage were nuclear lesions, multinucleate cells, micronuclei, telophase bridges and elongated cells.

Percentage of total number of aberrant cells was reported to show linear increase at higher concentrations of Monosodium Glutamate (MSG). Decrease in Relative Division Rate (RDR) was reported as the concentration increased, while Relative Abnormality Rate (RAR) and Mitotic Inhibition (MI) rate increased with the increase in concentrations of Monosodium Glutamate (MSG).

Plant system is reported to display numerous genetic and chromosome changes to determine the effects of mutagens. It is used extensively for mutagen screening (detection and verification of mutagenic activity), mutagenic monitoring and determining mutagen effects and mechanisms of mutagen action of certain mutagens [1]. The effect of induced mutagens in plants can be estimated by the observation of frequency of chromosomal abnormalities induced in them by the mutagens. The principle objective of using chromosome aberrations as a monitoring system is to determine the clastogenic nature (capacity to break chromosomes) of the mutagens involved. The clastogenic nature of mutagen permits the exchange with subsequent cytological or genetic damages [2].

Chromosome aberration studies in plants may be carried out by using actively dividing root tip, stem apex and pollen mother cells. However, observations on the root tip system of plants are considered a rapid and sensitive method for chromosomal monitoring. The degree of cytological aberrations in mitosis is regarded as one of the dependable criteria for estimating the mutagen sensitivity of the species and the effect of the mutagens [3]. Several types of chromosomal aberrations observed after treatment are considered as a reliable indicator of mutational change and are used as reliable evidence for screening of the mutational activity [4].

Therefore, in this investigation the clastogenic and carcinogenic effects of Monosodium Glutamate was measured by studying the degree of aberrations induced by it in the root tip cells of *Urginea indica* Kunth Cytotype I. As the concentrations of Monosodium Glutamate increased, linear decrease in the mitotic index and significant increase in chromosomal aberrations were

observed. Significant decrease in the mitotic index might have been caused by the mitodepressive action of Monosodium Glutamate on the cell division. The mitodepressive action may be the result of inhibition of DNA synthesis caused either due to the blocking of G1 suppressing DNA synthesis [5], or a blocking in G2 preventing cell from entering mitosis [6-8]. Another probable reason for the lowering of mitotic index may be the significant genotoxicity observed at higher concentrations [9]**.** This may also indicate the interference of Monosodium Glutamate with the normal sequence of mitosis, preventing a number of cells entering the prophase stage at interphase [10]. The reduction in mitotic index in *Urginea indica* Kunth with increased concentrations thus showed the ability of Monosodium Glutamate to inhibit DNA synthesis.

Moreover, treatment with different concentrations of Monosodium Glutamate showed a large number of chromosomal aberrations which increased significantly with the higher concentrations. Such physiological anomaly may be due to the stripping of protein covering by the action of Monosodium Glutamate on the chromatin material [11]**.** Chromosome fragments were found frequently which is reported to be the sign of extreme lethal clastogenic effects resulting from chromosome and chromatid breaks [12]. Chromosome fragments are reported to arise from the stretching of chromosomes at metaphase followed by breakage at these fragile sites [13]. Similarly, sticky metaphase was also found abundantly after treatment with Monosodium Glutamate which showed the highly toxic usually irreversible effect, probably leading to cell death [8]. It is reported to be the result of entanglement of interchromosomal chromatin fibers which leads to sub chromatid connection between chromosomes263. Stickiness may also be caused by hetero-chromatinization of a chromosome resulting in denaturation of nucleic acid which makes the chromosome counter to become adhesive. Due to sticky nature of chromosomes further cell cycle do not proceed leading to arrest after metaphase and anaphase which may lead to improper foldings of chromatin[14]**.**

High frequency of nuclear lesions was observed which might be due to disintegration of portion of nuclear material by the action of Monosodium Glutamate [9]. It may also be due to the stripping of chromatin at G1 phase. Prophase erosion might be due to the interaction of the tested chemical at G1 phase, which in turn results in the breakdown of chromosome packaging and subsequent disruption. Binucleate and multinucleate cells were also observed abundantly in both the cytotypes of *U. indica* Kunth which may be the result of inhibition of cytokinesis or cell plate formation [15].

Monosodium Glutamate caused a wide range of chromosomal abnormalities. The result thus showed that Monosodium Glutamate has a potential genotoxic and mutagenic effects.

Table-1: Percentage of Mitotic Abnormalities in each phase after treatment with Monosodium Glutamate (MSG) in *Urginea indica* Kunth Cytotype I in M1 generation

concentration	Total no. of cells observed	Total no. of abnormal cells	Total no. of dividing cells	%TAC	Physiological Aberrations			Clastogenic Aberrations		
					prophase	Metaphase	%PA	Anaphase	telophase	%CA
Control	793	-	571	-	-	-	-	-	-	-
0.1g/l	741	194	485	26.2	-	13.36	13.36	3.8	9.042	12.842
0.2g/l	736	227	463	30.8	-	17.256	17.256	4	9.647	13.647
0.3g/l	713	277	441	38.8	-	19.215	19.215	5.6	14.025	19.625
0.4g/l	758	308	448	40.6	-	19.921	19.921	5.0	15.699	20.699
0.4g/l	725	338	429	46.6	-	22.483	22.483	5.4	18.759	24.159

%TAC = Percentage of Total Number of Aberrant Cells
%PA = Percentage Physiological Aberration
%CA = Percentage Clastogenic Aberration

Table-2: Frequency of mitotic inhibition and abnormalities in root tip cells of *Urginea indica* Kunth Cytotype I after treatment with different concentrations of Monosodium Glutamate (MSG)S

Concentration	Mitotic Index	Relative Division Rate (RDR)	Relative Abnormality Rate (RAR)	Mitotic inhibition
Control	72.005	-	-	-
0.1g/l	65.452	− 23.408	− 163.619	9.101
0.2g/l	62.908	− 32.495	− 147.187	12.634
0.3g/l	61.851	− 36.271	− 118.610	14.102
0.4g/l	59.103	− 46.087	− 112.181	17.918
0.5g/l	59.172	− 46.019	− 90.748	17.822

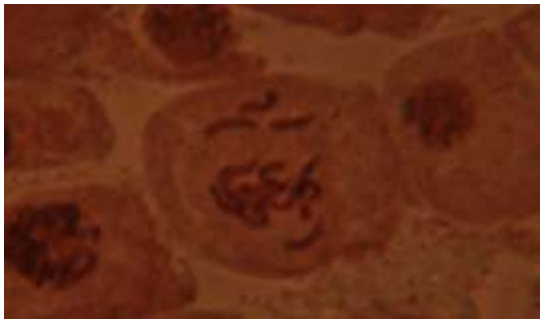


Fig.1. Precocious movement of chromosomes

Fig.2. Metaphase with persistent

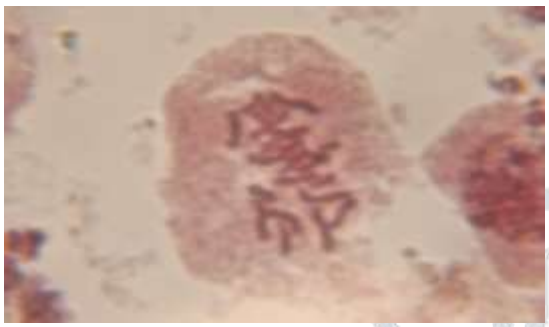


Fig.3. Diagonal Metaphase and Tropokinesis

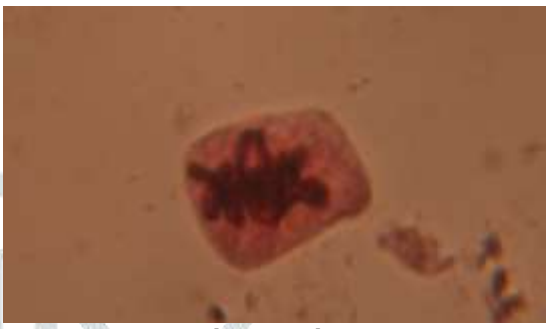


Fig.4. Sticky Metaphase

Fig.5. Polyploidy at Metaphase

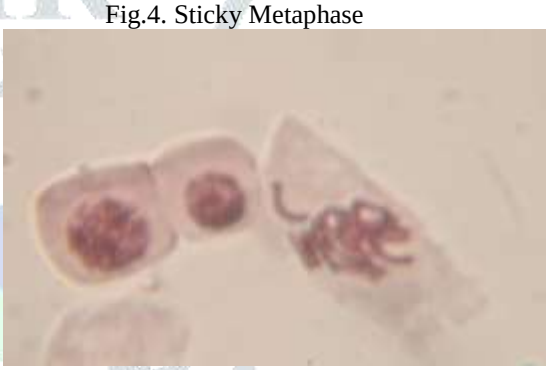
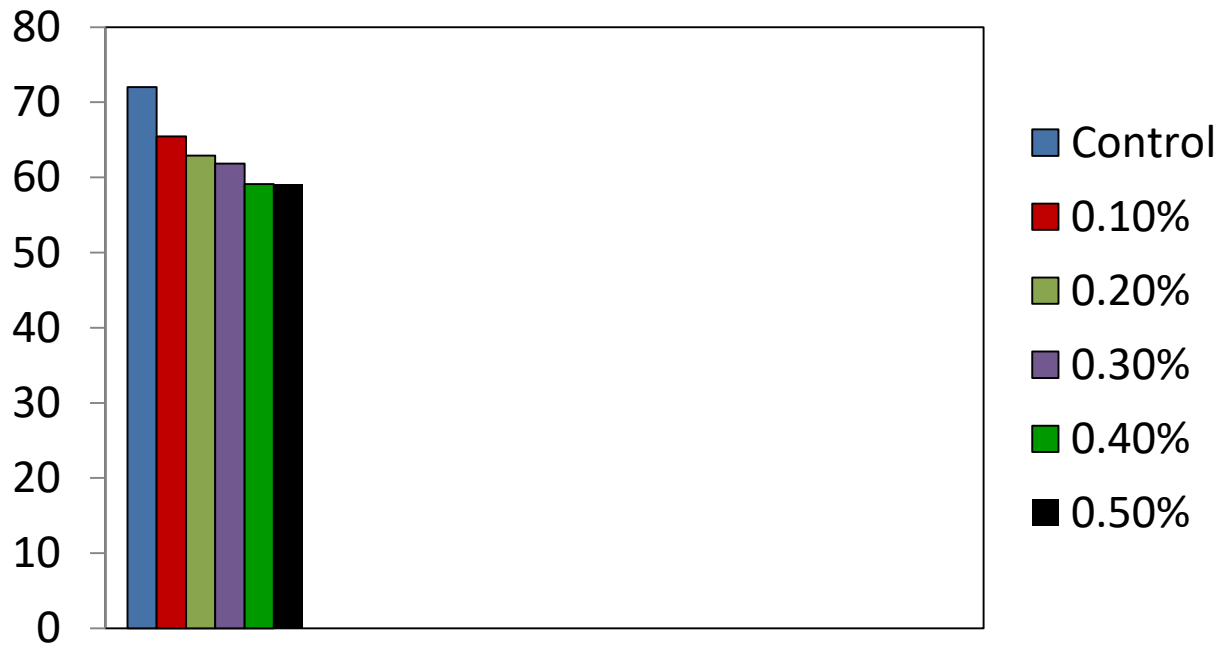


Fig.6. Sticky Metaphase with fragment

Fig.7: Column Graph of Mitotic Index Vs Concentration of Monosodium Glutamate in *Urginea indica* Kunth Cytotype I



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