EFFECT OF MONOSODIUM GLUTAMATE ON THE EPITHELIAL HEIGHT OF FALLOPIAN TUBE OF RAT AND ITS PREVENTION WITH DILTIAZEM

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ABSTRACT

Objective: To study the effect of monosodium glutamate on the fallopian tube of adult Sprague Dawley rat and prevention of this effect with diltiazem.

Study Design: Laboratory based randomized controlled trial.

Place and Duration of the Study: Anatomy Department, Army Medical College, Rawalpindi in collaboration with National Institute of Health (NIH), Islamabad from 9th April – 23rd April 2012.

Material and Methods: In this study 30 adult female Sprague Dawley rats of average weight of 500 g were randomly assigned into three groups A (control group), B and C (experimental groups) with 10 rats in each group. The experimental group B was given 0.08 mg/kg of monosodium glutamate (MSG) orally and experimental group C was given 0.08 mg/kg of MSG and 10 mg/kg of diltiazem in distilled water orally for 14 days. The control group (A) received only laboratory diet prepared at NIH Islamabad with liberal water intake. The rats were sacrificed on the 15th day of the experiment. The fallopian tubes (F.T) were carefully dissected out and fixed in 10% formalin for routine histological examination.

Result: The histological findings in the experimental group B showed evidence of an increase in epithelial height whereas no such finding was observed in groups A and there was a minor increase in epithelial height in group C.

Conclusion: MSG causes an increase in epithelial height of fallopian tubes in adult female rats but treatment with diltiazem prevents this effect.

Keywords: Diltiazem, Epithelial height, Fallopian Tubes, Monosodium Glutamate.

INTRODUCTION

Monosodium glutamate is the sodium salt of the non-essential amino acid glutamic acid which is one of the most abundant amino acids found in nature and exists both as free glutamate and bound to other amino acids in protein. Animal proteins contain 11 to 22% by weight of glutamic acid and the plant proteins have as much as 40% glutamate. Glutamate is found in a wide variety of foods where it has a flavor enhancing effect. It is also found in relatively high concentration in foods such as tomatoes, mushrooms, peas and cheese. As a result of its flavor enhancing effects glutamate is often deliberately added to foods usually as the purified monosodium salt. MSG is sold in the open market in Pakistan manufactured by National Foods and Shangrilla Foods. MSG stimulates the appetite centre and improves the taste of food[1-3]. There are certain reports indicating the toxicity of MSG to experimental animals and human beings[4]. In testicular tissue ascorbic acid content is reduced by MSG[5]. The degenerative and atrophic changes in the fallopian tubes are induced by MSG when administered in higher dosage and for prolonged period[6]. It induces the uterine fibroid in the rats by increasing the levels of total protein, cholesterol and estradiol (estrogen)[7].

The fallopian tubes are two muscular tubes which extend from uterus to the ovaries. The wall of the fallopain tubes consists of folded mucosa, a thick muscularis with circular and longitudinal layers and a thin serosa. The mucosa is composed of simple columnar epithelium on a lamina propria of loose connective tissue. The epithelium

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contains two types of cells: ciliated cells and secretory cells, or peg cells.

Diltiazem is a member of the class of drugs known as calcium channel blockers, used in the treatment of hypertension, angina pectoris, and some types of arrhythmias. It is also an effective preventive medication for migraine. Diltiazem prevents the toxic effects of MSG by reducing the intracellular calcium overload. Diltiazem blocks the calcium channels in the cell membrane, so reducing permeability of cell membrane for calcium ions. Previous studies have shown that pretreatment with diltiazem prevents the effects of MSG on hypothalamus of rats[8].

The effects of MSG on the fallopian tube have been studied however the role of diltiazem in the prevention of these effects has not been investigated.

MATERIAL AND METHODS

These laboratory based randomized controlled trials were carried out at Anatomy Department, Army Medical College, Rawalpindi in collaboration with National Institute of Health (NIH), Islamabad from 9th April -23rd April 2012. Thirty adult female Sprague Dawley rats weighing 490 to 510 grams were taken from the animal house of NIH, Islamabad. The rats were kept in cages at standard room temperature maintained on 12 hour light/dark cycle. The rats were randomly divided into three groups A (control group), B and C (experimental groups) with 10 rats in each group. The group A was fed on a laboratory diet for 14 days. The group B was fed on normal lab diet and was given 0.5 ml of distilled water by oral gavage tube containing monosodium glutamate 0.08 mg/kg body weight/day at 10 am daily for 14 days. The group C was fed on laboratory diet and was given 1 ml of distilled water containing diltiazem 10 mg/kg body weight/day and 0.5 ml distilled water containing monosodium glutamate 0.08 mg/kg body weight/day by oral gavage tube at 10 am daily for 14 days. Blood was drawn for hormonal analysis through cardiac puncture.The animals were euthanized with an overdose of ether anesthesia and sacrificed on the 15th day. The uterine horns, fallopian tubes and ovaries were identified and right sided fallopian tube of each rat (group A, B, C,) were dissected out and put into 10% formalin for fixation. The tissue was further processed for paraffin embedding. The staining of sections was done with Haemtoxylin and Eosin (H & E). Motic image plus microscope Model DMB3: 223 was used for histological study.

For photomicrography, the Motic image plus microscope Model: DMB3: 223 using the Motic Images plus 2.0 ML software on the computer was used. The height of the epithelium was taken from the basement membrane up to the upper limit of the cell facing the lumen from three regions and their mean was calculated as the reading for that animal to avoid the biasing and to minimize the error.

Table-1: Comparision of epithelial height (µm) between control and experimental groups.

Stat	Control (A)	Experimental (B)	Experimental (C)
Mean ± SD	13.24 ± 1.492	24.14 ± 3.615	16.45 ± 3.034
p- Value	A&B = 0.000	A&C = 0.047	B&C = 0.000

Table-2: Comparison of serum estrogen levels (pmol/l) between the control and experimental groups.

Stat	Control (A)	Experimental (B)	Experimental (C)
Mean ± SD	83.53 ± 9.877	136.97 ± 12.509	64.79 ± 17.853
p- Value	A&B = 0.000	A&C = 0.014	B&C = 0.000

The data was entered for analysis in the computer software SPSS version 18. The

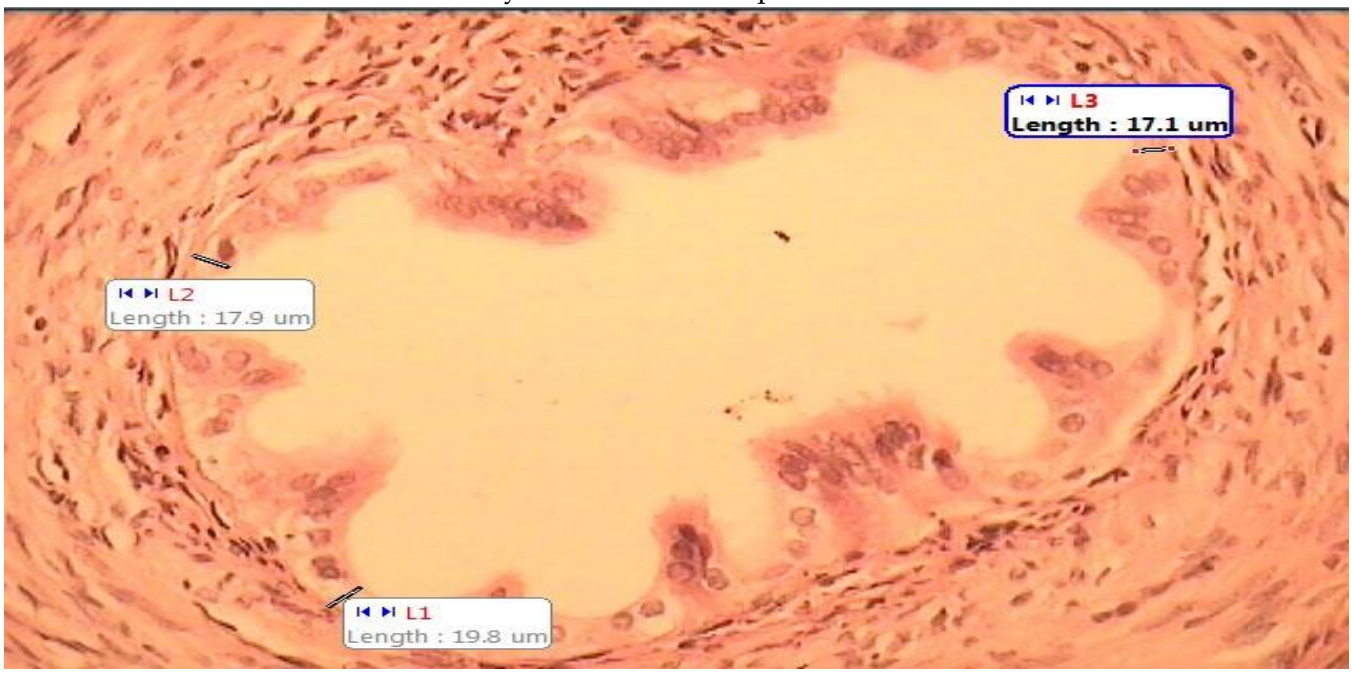

Figure-1: Photomicrograph of a cross section from fallopian tube of control group A showing epithelial height. H&E stain. 40X

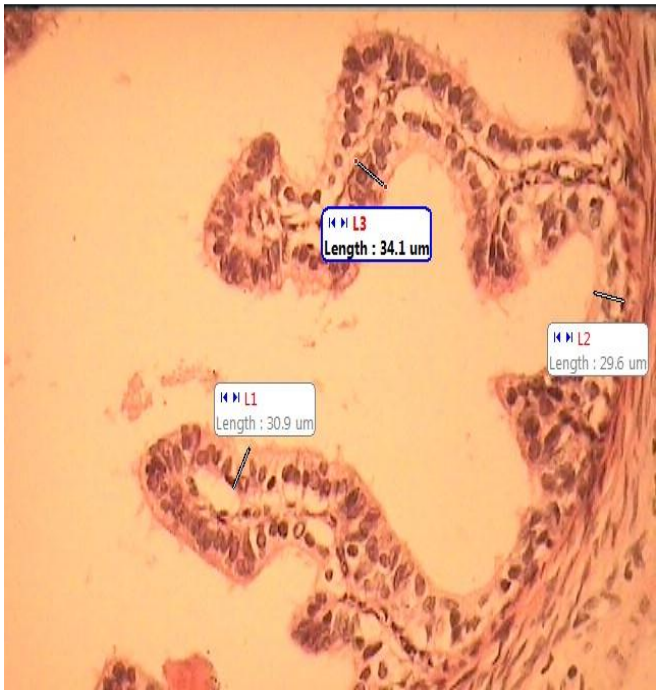

Figure-2: Photomicrograph of a cross section from fallopian tube of experimental group B showing epithelial height. H&E stain. 40X

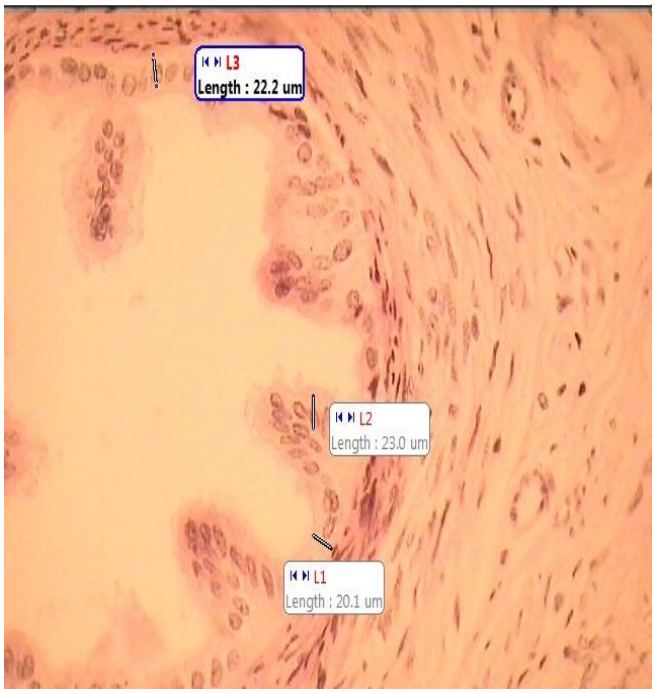

Figure-3: Photomicrograph of a cross section of fallopian tube of experimental group C showing epithelial height. H&E stain. 40X

quantitative variables includes height of epithelium of fallopain tubes and serum estrogen levels of control and experimental groups. The means were compared for significance using the ANOVA followed by post-hoc Tukey test for inter-group comparison at a confidence limit of 95 percent. p value < 0.05 was considered as significant.

RESULTS

This study included thirty animals divided into 3 groups with 10 in each group. The fallopian tubes of the control group A showed normal histological features, a well defined tubal wall consisted of three layers: the internal mucosa (endosalpinx), the intermediate muscular layer (myosalpinx), and the outer serosa. The mean epithelial height of fallopian tube of control group A was calculated as 13.24 ± 1.49 µm (Fig-1). The mean epithelial height of fallopian tube of group B was calculated as 24.14 ± 3.62 µm (Fig-2) and for group C it was 16.45 ± 3.03*u*m (Fig 3). The difference in the mean of epithelial height between three groups was signigicant (p=<0.001).

The mean serum estrogen level of control group A was 83.53 ± 9.87 pmol/l. The difference in the mean of serum estrogen levels between three groups was significant (p<0.001) (Table-2).

DISCUSSION

MSG is the sodium salt of the non-essential amino acid glutamic acid and has long been used due to its flavor enhancing properties as it improves the taste of food by stimulating the appetite centre as well as debated for its safety and harmful effects. The epithelial height of fallopian tube of control group A remained within normal limits whereas in experimental group B (on MSG) it was markedly increased and in experimental group C (on MSG and diltiazem) it was slightly increased. In group B the epithelial height of the fallopian tube was found significantly increased as compared with control group A. Other studies have also found not only similar effects on fallopian tubes but also confirmed that MSG causes cellular hypertrophy of many other body organs. In one of the studies increase in cellular height of the epithelium and atrophic and degenerative changes were found in fallopian tube of MSG treated group[6]. Earlier separately published studies on different body parts of the rat by the same researchers confirmed that MSG affects the epithelium of other body organs as well. Among the MSG treated study animals they found an increase in cellular height of stomach epithelium[9], small intestine[10], medial geniculate body[11]. The increase in epithelial height may be attributed to increase food intake and increase in the serum estrogen level in group B. The MSG increases the serum estrogen levels by activating the enzyme (aromatase),which catalyze the coversion of androgens to estrogen. As the fallopian tube epithelium is the most common site for the fertilization of the ova. This increase in epithelial height and degenerative changes may lead to changes in the functional aspects of the fallopian tube becoming a leading cause of infertility in females. The histological finding of group C showed a minor increase in height in the epithelium of fallopian tube as compared to group B. We found significant differences between the epithelial height of experimental group B (MSG) and experimental group C (MSG and diltiazem). This shows that diltiazem has protected the epithelium against the damaging effects induced by MSG on the fallopian tubes. MSG might have exerted these effects through increasing the permeability of cell membrane for calcium ions. Diltiazem reduces the permeability of calcium ions through the cell membrane and decreasing the intracellular load of calcium and thus preventing the cellular changes induced by the MSG[8].

Although preventive effects of diltiazem against MSG induced effects on various body parts have been demonstrated however this has not been tested on fallopian tubes. The diltiazem prevents the effects of MSG on the ovary of adult rat[12]. During the neonatal period diltiazem administration prevented the toxic effects of MSG

on the hypothalamus[8] and on the variability in rat[13].

Our findings determined the significance of diltiazem in prevention the toxicity induced by MSG on fallopian tube.

CONCLUSION

The findings obtained from our study after administration of MSG showed increase in height of columnar epithelium of fallopian tube while treatment with diltiazem prevents these effects.

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