

Protective Effects of Phoenix Dactylifera on Monosodium Glutamate Induced Histomorphological Changes in Hepatocytes of Adult Male Albino Rats: A Light Microscopic Study

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ABSTRACT

Objective: To observe the possible protective role of Ajwa date fruit extract on monosodium glutamate induced histomorphological changes in liver of adult male rats.

Study Design: Experimental randomized controlled trial study

Place and Duration of Study: This study was conducted at the Department of Anatomy, Peshawar Medical College, Peshawar from July 2018 to December 2018.

Materials and Methods: This study was carried out on 24 adult male albino rats. The rats were randomly divided into 4 groups, A, B, C1 and C2, containing 06 rats in each group. **Group A** served as control. **Group B** received MSG 2g/kg/day prepared in distilled water; **Group C1** was given MSG 2g/kg and ADFE 1g/kg and **Group C2** was given MSG 2g/kg and ADFE 2g/kg by oral gavage once daily for 21 days. Rats were sacrificed on 22nd day. Liver from each rat was dissected out and examined microscopically.

Results: Histological examinations revealed, statistically highly significant (0.001) decrease in the number and increase in diameter of hepatocytes in toxic MSG group as compared to control group. These changes were protected in group C1 and C2, who received monosodium glutamate plus Ajwa date fruit extract.

Conclusion: Ajwa date fruit extract has protective effects on monosodium glutamate induced liver toxicity.

Key Words: Monosodium glutamate, Ajwa date fruit extract, hepatocytes and histomorphological changes

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INTRODUCTION

Taste and flavor are important to enjoy food; hence food additives are needed worldwide.¹ Food additive has been used for the promotion of, coloring, flavoring and to increase the useful shelf-life of food for decades.² Flavoring systems plays important nutritional role in savory food industry, especially in tasteless foods, by giving the required appeal.³

Monosodium glutamate (MSG) specifically used as flavor enhancer is very well-known food additive throughout the world specially in Chinese foods. It has been used since 1907 as flavor enhancer.⁴

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MATERIALS AND METHODS

This experimental randomized controlled trial study was conducted in the Department of Anatomy, Peshawar Medical College, Peshawar from 1st July 2018 to 31st December 2018. A total of 24 male albino Wistar rats were used. The rats were divided randomly into four groups, A, B, C1 and C2. Group A served as a control group, while group B, C1 and C2 were experimental groups, having six rats in each group. All rats were examined completely before starting the experiment. After labelling the cages the rats were placed in their respective cages, under standard condition, at room temperature (18°C-26°C) for 28 days. The rats were kept in light and dark cycle of twelve hours. All the animals received doses by oral gavage once daily in the morning at 8 am for 21 consecutive days. **Group A:** The animals in this group were given distilled water, **Group B:** The animals in this group received MSG 2g/kg/day prepared in distilled water, **Group C1:** The animals in this group received MSG 2g/kg/day and ADFE 1g/kg/day and **Group C2:** Each animal in this group was given MSG 2g/kg/day and ADFE 2g/kg/day.

The rats were sacrificed by heavy dose of ether anesthesia on 22nd day. After euthanizing, the rat was

placed on the dissecting board. The paws of animal were fixed to dissection board by using thumb pins. The body cavity was opened by using forceps and scissors and incision was made in midline from chin to pubis. Skin flaps were opened and pinned to dissection board. Liver was dissected out from the body and put in a separate properly labelled jar containing 10% neutral buffered formalin (NBF), for further processing. The number and diameter of hepatocytes was noted after staining with hematoxylin and eosin (H and E).

The data was analyzed by analysis of variance (ANOVA) using Statistical Package of Social Sciences (SPSS) version 25. The quantitative variables were described by Mean ± S.D. The p-value <0.05 is considered to be statistically significant.

RESULTS

The number of hepatocytes/HPF was lower in group B may be due to increase in their volume as compared to other groups. Number of hepatocytes /HPF during experiment ranged between 395-420, 328-340, 368-385 and 400-430 in groups A, B, C₁ and C₂ respectively; the mean number of hepatocytes /HPF during the experiment was 410±9.14, 333.16±4.21, 375.16±6.46 and 413.66±11.60 in groups A, B, C₁ and C₂ respectively (Table 1).

Post hoc Tukey test was applied for multiple comparisons, which presented that the number of hepatocytes/HPF in groups B was lower significantly in comparison to remaining groups, while number of hepatocytes/HPF in group C₁ was significantly lower as compare to group A and C₂. However, there was no difference significantly in the number of hepatocytes /HPF in groups A and C₂ (Table 2).

Diameters of hepatocytes during experiment ranged between 20.89-22.11, 26.34-28.32, 21.16-22.69 and 22.89-23.87µm. The mean percentage diameters of hepatocytes (µm) during the experiment were 21.47±0.42, 27.14±0.67, 22.07±0.53 and 23.42±0.32 µm in groups A, B, C₁ and C₂ respectively (Table 3).

To compare the groups, post hoc Tukey test was applied, showed that diameters of hepatocytes (µm) in groups B were significantly larger as compared to other groups, while diameters of hepatocytes (µm) in groups C₂ were significantly lower as compared to groups B and C₁. There was no difference significantly in the diameters of hepatocytes /HPF in groups A and C₂ (Table 4).

Table No. 1: One-way ANOVA showing the mean and standard deviation of number of hepatocytes/HPF among the groups

Variable	Group A (n=6)	Group B (n=6)	Group C ₁ (n=6)	Group C ₂ (n=6)	P value
Number of hepatocytes/HPF	410±9.14	333.16±4.21	375.16±6.46	413.66±11.60	0.001*

*P value 0.001 (Highly significant)

Table No.2: Pair wise comparison of number of hepatocytes /HPF among the groups

(I) Group	(J) Group	Mean Difference (I- J)	Std. Error	Sig.
A	B	76.83333*	4.81144	.000
	C ₁	34.83333*	4.81144	.000
	C ₂	-3.66667	4.81144	.870
B	C ₁	-42.00000*	4.81144	.000
	C ₂	-80.50000*	4.81144	.000
C ₁	C ₂	-38.50000*	4.81144	.000

*P value ≤ 0.001 (Highly significant)

Table No.3: One-way ANOVA showing the mean and standard deviation of diameters of hepatocytes among the groups

Variable	Group A (n=6)	Group B (n=6)	Group C ₁ (n=6)	Group C ₂ (n=6)	P value
Diameter of hepatocytes (µm)	21.47±0.42	27.14±0.67	23.42±0.32	22.07±0.53	0.001*

*P value ≤ 0.001 is highly significant statistically

Table No.4: Pair wise comparison of diameters of hepatocytes among the groups

(I) Group	(J) Group	Mean Difference (I- J)	Std. Error	Sig.
A	B	-5.66500*	.29160	.000
	C ₁	-.59500	.29160	.207
	C ₂	-1.94500*	.29160	.000
B	C ₁	5.07000*	.29160	.000
	C ₂	3.72000*	.29160	.000
C ₁	C ₂	-1.35000*	.29160	.001

*P value ≤ 0.001 is highly significant statistically

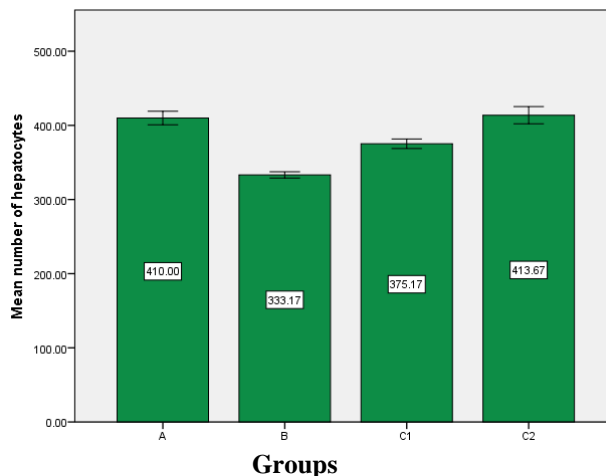


Figure No. 1: Bar chart showing number of hepatocytes / HPF among the groups

DISCUSSION

Monosodium glutamate (MSG) is a widely used food additive and flavor-enhancer that may be present in canned food without appearing on the label. This makes MSG the one of most utilized food flavor additives in

the modern nutrition throughout the world.⁵ Exposure to chemical preservatives and food additives have affected the liver through oxidative stress, which has a key role in xenobiotic induced hepatotoxicity.¹⁹

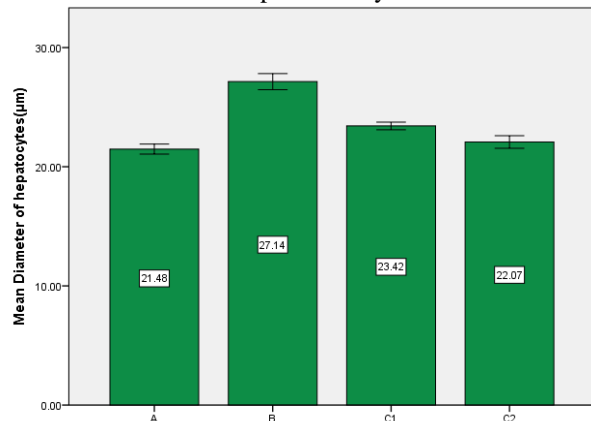


Figure No. 2: Bar chart showing diameters of hepatocytes among the groups

The fruit of Date palm has been used since ancient time as an herbal remedy. Among many varieties of the date fruit, the medicinal properties of the Ajwa dates are unique, which are especially grown in the Al-Madina Al-Munawarah.¹⁹ Studies have shown that Ajwa fruit or pits can significantly improve the liver function by reducing liver marker enzymes and increasing the level of antioxidants.¹⁸ In our study, a significant increase in diameter of hepatocytes in toxic group B was observed after administration of MSG at dose of 2gm/kg body weight in comparison to the control group ($P < 0.001$). The increase in diameter of hepatocytes after administration of MSG might be due to an increase in oxidative stress in liver tissue.¹² However, the groups C1 and C2 treated with MSG as well as ADFE showed normal arrangement of hepatic cords with normal size hepatocytes and reversal of cellular changes that observed in MSG group B. The reversal of these changes is in agreement with study of Ragab et al²⁰, who found hepatoprotective effect of ADFE on lead induced hepatotoxicity in rats.

In the present study we also observed decreased number of hepatocytes/HPF in only MSG treated group as compared to control group, these finding may be due to large size of hepatocytes and congested dilated sinusoids. However, the groups C1 and C2 treated with ADFE plus MSG showed number of hepatocytes close to control group indicate protective effect of Ajwa date fruit extract against MSG induced liver toxicity. This study is in accord with the findings of study by Saafi et al²¹ who found hepatoprotective effects of date palm on dimethoate induced liver toxicity in rats.

CONCLUSION

Ajwa date fruit extract had significant protective effect against monosodium glutamate induced histological

changes in the liver. This effect may be due to the rich vitamins and antioxidants in the extract.

Author's Contribution:

Concept & Design of Study: Rifat Shamim
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 Data Analysis: Seharish Zulfiqar, Nighat Ara, Farooq Khan
 Revisiting Critically: Rifat Shamim, Zafar Iqbal
 Final Approval of version: Rifat Shamim

Conflict of Interest: The study has no conflict of interest to declare by any author.

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