Biological and Biomedical Journal

Journal homepage: https://bbj.journals.ekb.eg

Protective effect of silymarin on monosodium glutamate-induced liver toxicity in rats

Nwakaego O. Ebong[1], Elias Adikwu[2]*, Emmanuel Julius[3]

[1]Department of Clinical Pharmacy and Management, Faculty of Pharmaceutical Sciences, University of Port Harcourt, Rivers State, Nigeria.
[2]Department of Pharmacology/Toxicology, Faculty of Pharmacy, Niger Delta University, Bayelsa State, Nigeria
[3]Department of Pharmacology/Toxicology, Faculty of Pharmacy, Madonna University, River State, Nigeria

ARTICLE INFO

Received: 29/06/2024
Accepted: 16/12/2024

***Corresponding author:**
Elias Adikwu
E-mail: adikwuelias@gmail.com
Mobile: (+234)9036073094

P-ISSN: 2974-4334
E-ISSN: 2974-4324
DOI:
10.21608/bbj.2024.300220.1031

ABSTRACT

Monosodium glutamate (MSG) is a food additive with potential hepatotoxic effect. Silymarin (SIL) has promising liver protective activity. This study assessed the protective effect of SIL on MSG-induced liver toxicity in rats. Thirty adult Wistar rats (180 -200 g) were grouped into 6 of 5 rats/group. The rats were orally treated for 14 days as follows: Group 1 (Control: Distilled water), group II (MSG; 600 mg/kg/day) and group III (SIL; 200 mg/kg/day). Groups IV-VI were supplemented with SIL (50, 100 and 200mg/kg/day) before treatment with MSG (600 mg/kg/day). On day 15, blood samples were collected for liver function marker investigations. Liver samples were weighed and analysed for oxidative stress markers and histology. MSG significantly ($p< 0.01$) increased body and liver weights ($p< 0.01$) serum gamma-glutamyl transferase, alkaline phosphatase, amino transferases, lactate dehydrogenase, bilirubin and liver malondialdehyde levels when compared to the control. Glutathione, superoxide dismutase, catalase, and glutathione peroxidase levels were significantly ($p< 0.001$) decreased by MSG when compared to control. MSG caused hepatocyte necrosis. However, SIL supplementation restored body and liver weights at 50 mg/kg ($p< 0.05$), 100 mg/kg ($p< 0.01$) and 200 mg/kg ($p< 0.01$) when compared to MSG. SIL supplementation restored the aforementioned biomarkers at 50 mg/kg ($p< 0.05$), 100 mg/kg ($p< 0.01$) and 200 mg/kg ($p< 0.001$) when compared to MSG. Liver structure was restored by SIL supplementation. It was concluded that SIL protects against MSG-induced liver toxicity in a dose-related fashion.

Keywords: Monosodium glutamate, Liver, Protection, Silymarin, Toxicity

1. Introduction

Liver is an essential metabolic organ that is involved in the metabolism of molecules. Aside from its metabolic role, it plays an important role in the detoxification and excretion of endogenous and exogenous chemical substances, thus protecting the body from toxic substances. This function, predisposes the liver to injury due to prolonged exposure to chemical substances such as drugs and their metabolites (Shakya, 2020). Liver injury due to chemical substances can cause a multiplicative effect where previous damage can feed-forward causing impaired drug metabolism and further toxicity (Mudd and Guddati, 2012). Severe liver injury can result from damage to structures such as liver sinusoids, hepatocytes, vasculature, and bile ducts. Also, changes including elevated serum liver function markers, hepatitis, steatosis, and chronic outcomes such as fibrosis and liver failure are features of liver damage caused by chemical substances (Shakya, 2020).

Monosodium glutamate (MSG) is a flavour enhancer sourced from L-glutamic acid, which is a natural occurring amino acid in most food products. In addition to its flavour-enhancing property, it is used as a food additive in the form of hydrolyzed protein or purified monosodium salt. MSG was described as the fifth basic taste in addition to salty, sweet, sour, and bitter tastes (Bayram et al., 2013). In recent times, MSG usage as a food additive has become more popular, with an estimated daily human consumption of about 0.3–1.0 g in European countries (Zanfirescu et al., 2019). MSG has been labelled as safe by food safety regulators, but some preclinical and clinical studies have raised safety concern, especially following chronic exposure. Preclinical studies have associated MSG with nephrotoxicity, cardiotoxicity, neurotoxicity, inflammation, metabolic syndromes, behavioural changes, tumorigenesis and genotoxicity (Zanfirescu et al., 2019). Also, preclinical studies suggest that MSG usage may cause hepatotoxicity marked by alterations in liver biochemical markers, perturbation in liver architecture and the induction of oxidative stress in the liver (El-Morsi et al., 2019; Shinde et al., 2022).

Silymarin (SIL) is extracted from the seeds and fruits of *Silybum marianum* (Milk thistle) (Cheemerla and Balakrishnan, 2021). It contains the flavonolignan isomers; silichristin, and silibinin as its primary constituents. The silibinin isomer is the most physiologically active and prevalent constituent. It forms between 50-60% of the complex, whereas flavonolignan isomers such as silichristin make up 35% (Gillessen and Schmidt, 2020; Marceddu et al., 2022). Silibinin, the primary active constituent of SIL, has a renowned and potent antioxidant effect, which scavenges free radicals and inhibits oxidative stress pathways (Kadoglou et al., 2022). It has anti-inflammatory property that prevents the production of inflammatory mediators and inflammatory metabolites. SIL shows antifibrogenic activity in animal model of hepatic fibrosis (Liu et al., 2023) with promising protective effects on cirrhosis and alcohol-related liver disease (Koushki et al., 2020), non-alcoholic fatty liver disease (Li et al., 2020), amatoxin-induced liver failure (Fu et al., 2022) and antituberculosis drug-induced liver damage (Tao et al., 2019). Considering the aforementioned information, the current study novelty assessed whether SIL supplementation can protect against MSG-induced hepatotoxicity in rats.

2. Materials and methods

Chemicals and animals

Biochemical reagent kit for the evaluation of biochemical parameters were purchased from Transasia Bio-medicals Ltd. (India) while MSG and ELISA kit were obtained from Sigma Aldrich USA. All other chemicals used were procured from Merck and Himedia Pvt. Ltd. (India).

Thirty adult Wistar rats of both sexes (8 weeks, weighing 180-200g) were purchased from the Animal House of the Faculty of Pharmacy, Madonna University, Nigeria where the study was performed. The rats were randomly grouped into 6 groups of n=5/group and housed in soft wooden cages, under standard temperature (25-30ºC) with 12-h light/dark cycle. The rats were allowed to adapt to climatic conditions with free access to food and water and were handle according to the Guide for the Care and Use of Laboratory Animals. 8th edition, 2011

Administration of drug and chemicals.

The dose of MSG used was obtained from a pilot study whereas the doses of SIL used were based on previous studies (Gao et al., 2017). MSG (Atef et al., 2019) and SIL (Oda and El-Ashmawy, 2012) were dissolved in normal saline.

Group I (Control group): The rats were orally administered with distilled water (0.2mL/day) for 14 days. Group II: The rats were orally administered with SIL (200 mg/kg/day) for 14 days. Group III: The rats were orally administered with MSG (600 mg/kg/day) for 14 days. Groups IV-VI: The rats were orally supplemented with SIL (50, 100 and 200 mg/kg/day) for 30 minutes before the administration of MSG (600 mg/kg/day) for 14 days.

Animal sacrifice and sample collection

On day 15, the rats were anesthetized using thiopental sodium (40mg/kg), and sacrificed by decapitation. Blood samples were collected by

cardiac puncture in heparinized tubes for gamma-glutamyl transferase (GGT), alkaline phosphatase (ALP), alanine amino transferase (ALT), conjugated bilirubin (CB), lactate dehydrogenase (LDH), total bilirubin (TB) and aspartate amino transferase (AST) evaluations. Liver samples were collected and processed to make 10% (w/v) homogenate in ice-cold 20 mM tris (hydroxymethyl) aminomethane buffer (pH 7.4). The homogenates were centrifuged (3000 X g for 30min at 4ºC). The supernatants were collected and assayed for superoxide dismutase (SOD), malondialdehyde (MDA), glutathione (GSH), catalase (CAT) and glutathione peroxidase (GPX). Liver (left lobe) samples were removed, rinsed in physiological saline (0.9% NaCl) and stored in neutral buffered formalin (10%) for histological analysis.

Determination of biochemical markers

Serum AST, ALT, LDH ALP, GGT, CB and TB were analysed with an auto chemistry analyzer.

Determination of oxidative stress markers

SOD and CAT activities were investigated as reported by Sun and Zigman, 1978 and Aebi, 1974, respectively. MDA and GSH activities were analysed using the processes described by Buege and Aust, 1978 and Sedlak and Lindsay, 1968, respectively. GPX was evaluated using the method reported by Rotruck et al. (1973).

Histological analysis

Liver (left lobe) samples were rinsed in physiological saline (0.9% NaCl) and fixed in neutral buffered formalin (10%). After dehydration in series of ethanol solutions (50%, 70%, 95%, 100%), the liver tissues were cleared in xylene and fixed in paraffin. Five-micrometer-thick tissue sections were stained using hematoxylin and eosin and stained sections were examined and photographed using a light microscope.

Data analysis

The estimated results are the mean of five replicates. This study used SPSS (Statistical Package for the Social Sciences; SPSS Inc., Chicago, IL, USA) for Windows Version 22 for data analysis. Data were evaluated by two-way analysis of variance (ANOVA) and Duncan's Multiple Range Test. Results were presented as mean and standard error of mean (SEM). Significance was set at $p< 0.05$, $p< 0.01$ and $p< 0.001$.

3. Results

Effect of silymarin on body and liver weights of monosodium glutamate-administered rats

Administered SIL (200 mg/kg) had no significant ($p>0.05$) effects on the body and liver weights but administered MSG significantly ($p<0.01$) increased the aforementioned parameters when compared to the control (Table 1). On the other hand, SIL supplementation restored body and liver weights at 50 mg/kg ($p<0.05$), 100 mg/kg ($p<0.01$), and 200 mg/kg ($p<0.01$) when compared to MSG (Table 1).

Effect of silymarin on serum liver biochemical markers of monosodium glutamate administered rats

SIL (200 mg/kg) had no significant ($p>0.05$) effects serum ALT, ALP, AST, GGT, LDH, CB and TB levels, but MSG significantly ($p<0.001$) increased the serum levels of the aforementioned biochemical markers when compared to the control (Table 2). Interestingly, SIL supplementation restored the serum levels of the aforementioned biochemical markers in a dose-related fashion at 50 mg/kg ($p< 0.05$), 100 mg/kg ($p<0.01$), and 200 mg/kg ($p< 0.001$) when compared to MSG (Table 2).

Effect of silymarin on liver oxidative stress markers of monosodium glutamate administered rats

Liver oxidative stress markers remained unchanged ($p>0.05$) following the administration of SIL (200 mg/kg) when compared to the control (Table 3). In contrast, liver SOD, CAT, GSH and GPX levels decreased significantly ($p<0.001$) whereas MDA level increased significantly ($p< 0.001$) following the administration of MSG when compared to the control (Table 3). However, liver SOD, CAT and GSH, GPX and MDA levels were restored by SIL supplementation in a dose-related fashion at 50 mg/kg ($p<0.05$), 100 mg/kg ($p<0.01$), and 200 mg/kg ($p<0.001$) when compared to MSG (Table 3).

Effect of silymarin on liver histology of monosodium glutamate administered rats

The liver of rats used as control (Figure A) and the liver of SIL (200 mg/kg/day) administered

rats (Figures B) showed normal histology. On the other hand, the liver of MSG administered rats showed hepatocytes necrosis (Figure C). Furthermore, the liver of rats supplemented with SIL (50 mg/kg) (Figure D), SIL (100 mg/kg) (Figure E) and SIL (200 mg/kg) (Figure F) showed normal histology.

Tables 1. Effect of silymarin on the body and liver weights of monosodium glutamate-administered rat

Groups	Dose (mg/kg)	FBW (g)	ALW(g)	RLW (%)
Group I	DW	200.6 ± 16.9	5.20 ± 0.76	2.59 ± 0.05
Group II	SIL 200	208.2 ± 18.4	5.34 ± 0.11	2.57 ± 0.08
Group III	MSG 600	303.8 ±15.6$^{\#}$	12.72 ± 0.32$^{\#}$	4.19 ± 0.78$^{\#}$
Group IV	SIL 50 + MSG 600	251.3 ±17.1^{a}	8.80 ± 0.71^{a}	3.50 ± 0.16^{a}
Group V	SIL 100 + MSG 600	202.6 ± 16.8^{b}	5.21 ± 0.43^{b}	2.57 ± 0.33^{b}
Group VI	SIL 200 + MSG 600	200.5 ± 14.3^{b}	5.10 ± 0.55^{b}	2.54 ± 0.41^{b}

Values are mean ± SEM, n = 5. DW: Distilled water, MSG: Monosodium glutamate, SIL: Silymarin, FBW: Final body weight, ALW: Absolute liver weight, RLW: Relative liver weight, # p<0.01 Significant when compared to control, a: p<0.05; and b: p<0.01 Significant when compared to MSG (ANOVA).

Table 2. Effect of silymarin on serum liver biochemical markers of monosodium glutamate-administered rats

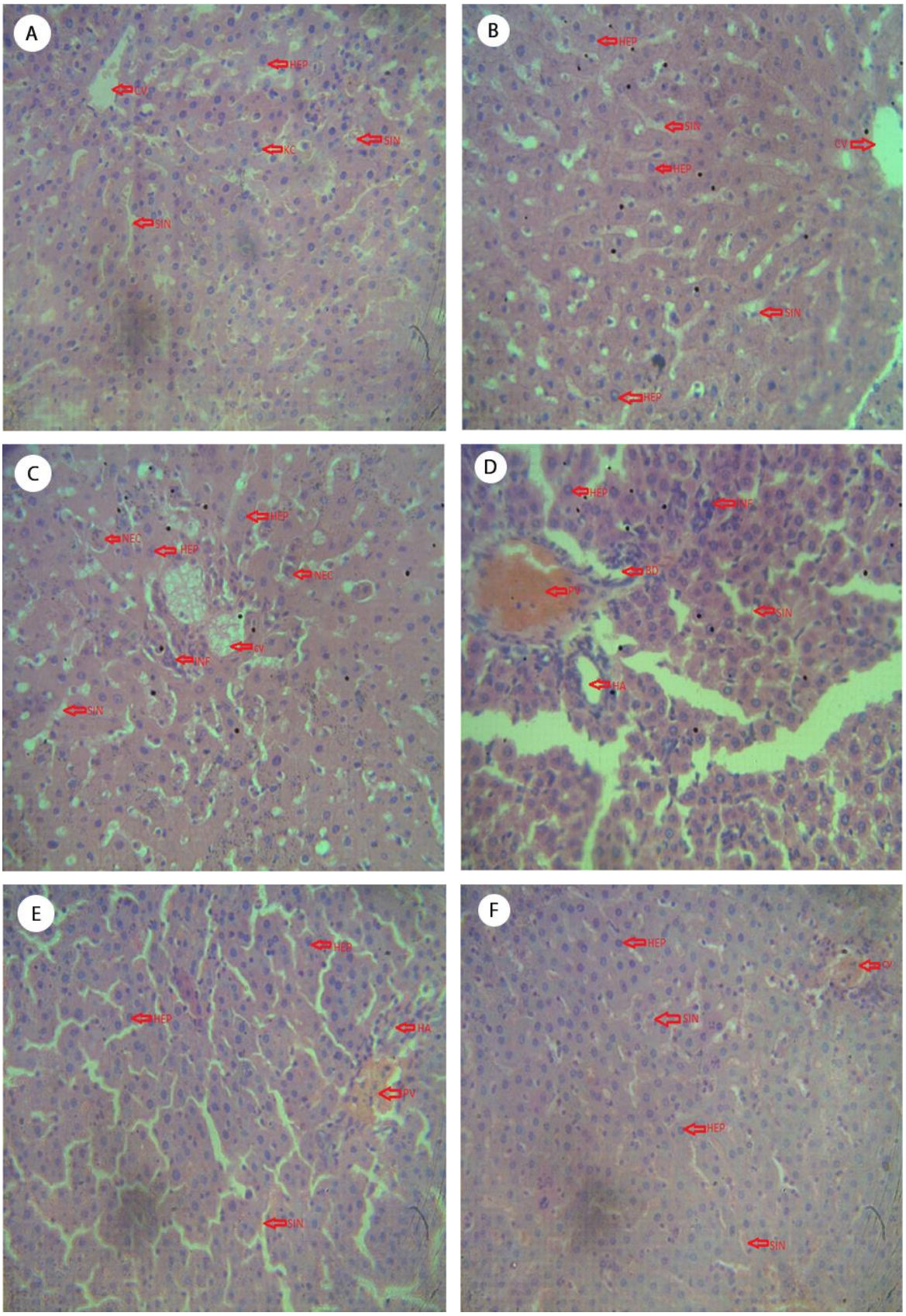
Groups	Dose (mg/kg)	AST (U/L)	ALT(U/L)	ALP(U/L)	TB(g/dL)	CB(g/dL)	LDH(U/L)	GGT(U/L)
Group I	DW	39.12 ± 3.65	38.03±3.67	31.63 ± 3.90	6.85 ± 0.40	4.89 ± 0.34	24.71 ± 3.98	0.20 ± 0.01
Group II	SIL 200	37.61 ± 2.79	37.91±3.39	31.07 ± 4.72	6.67±0.23	4.63±0.54	24.52 ± 4.70	0.28 ± 0.06
Group III	MSG 600	158.03 ± 15$^{\#}$	148.73±14.8$^{\#}$	98.54 ± 10.9$^{\#}$	18.96 ± 2.55$^{\#}$	15.74 ± 1.66$^{\#}$	108.71±17$^{\#}$	1.21 ± 0.04$^{\#}$
Group IV	SIL 50 + MSG 600	98.25 ±12.8^{a}	98.92 ± 13.00^{a}	77.71 ± 5.27^{a}	14.73 ± 1.40^{a}	12.31 ± 1.00^{a}	69.53 ± 6.76^{a}	0.96 ± 0.09^{a}
Group V	SIL100 + MSG600	69.16 ± 5.8^{b}	62.63 ± 6.75^{b}	56.83 ± 4.54^{b}	8.93 ± 0.36^{b}	8.03 ± 0.53^{b}	43.94 ± 4.61^{b}	0.48 ± 0.05^{b}
Group VI	SIL200 + MSG600	44.72 ± 3.63^{c}	42.71 ± 4.47^{c}	34.21 ± 3.61^{c}	6.73 ± 0.28^{c}	5.00 ± 0.45^{c}	28.72 ± 3.54^{c}	0.24 ± 0.08^{c}

Values are mean ± SEM, n = 5. DW: Distilled water, MSG: Monosodium glutamate, SIL: Silymarin AST: Aspartate aminotransferase, GGT: Gamma glutamyl transferse, ALT: Alanine aminotransferase, LDH: Lactate dehydrogenase, TB: Total bilirubin, ALP: Alkaline phosphatise, CB: Conjugated bilirubin, $^{\#}$ p<0.001 Significant when compared to control. a: p<0.05; b: p<0.01 and c: p<0.001 Significant when compared to MSG. (ANOVA).

Table 3. Effect of silymarin on liver oxidative stress markers of monosodium glutamate - administered rats

Group	Dose (mg/kg)	SOD (u/mg protein)	CAT (u/mg protein)	GSH (µg/mg protein)	GPX (u/mg protein)	MDA (nmol/mg protein)
Group I	DW	39.86 ± 3.43	42.85± 5.00	21.44 ± 2.11	25.17 ± 2.71	0.16 ± 0.05
Group II	SIL 200	41.14 ± 3.67	43.16 ± 4.51	20.57 ± 2.32	27.22 ± 2.15	0.12 ± 0.03
Group III	MSG 600	14.85 ± 1.68$^{\#}$	16.43 ± 1.67$^{\#}$	6.34 ± 0.11$^{\#}$	8.48 ± 0.37$^{\#}$	0.68 ± 0.01$^{\#}$
Group IV	SIL 50+MSG 600	19.62 ± 2.64^{a}	21.75 ± 2.00^{a}	10.52 ± 0.12^{a}	11.57 ± 0.67^{a}	0.49 ± 0.07^{a}
Group V	SIL 100+MSG 600	26.47 ± 3.66^{b}	28.47 ± 4.63^{b}	13.61 ± 1.56^{b}	16.63 ± 1.74^{b}	0.27 ± 0.04^{b}
Group VI	SIL 200+MSG 600	35.28± 3.57^{c}	39.97± 5.71^{c}	20.03 ± 2.60^{c}	23.92 ± 2.47^{c}	0.14 ± 0.06^{c}

Values are mean ± SEM, n = 5. DW: Distilled water, MSG: Monosodium glutamate, SIL: Silymarin, SOD: Superoxide dismutase, CAT: Catalase, GSH: Glutathione, MDA: Malondialdehyde, GPX: Glutathione peroxidase, Data as mean± SEM, n=5, $^{\#}$ p< 0.001 Significant when compared to control. a: p<0.05; b: p<0.01; and c: p<0.001 Significant when compared to MSG. (ANOVA).



Figures A-F Liver of rats administered with SIL and MSG. Figure A: Control. Figure B: SIL (200mg/kg/day). Figure C: MSG (600 mg/kg/day), Figure D: Supplemented with SIL (50mg/kg/day), Figure E: Supplemented with SIL (100mg/kg/day), Figure F: Supplemented with SIL (200mg/kg/day). HEP: Normal hepatocytes, CV: Central vein, PV: Portal vein, NEC: Necrosis, SIN; Sinusoids, HP: Hepatic artery; INF: Inflammatory cells.

4. Discussion

The use of food enhancers and food additives may cause deleterious effects on humans. MSG, a widely and frequently used food additive has been associated with a number of toxicities in animal studies with suggestions that excess consumption may impair the liver (Hajihasani et al., 2020). This study assessed whether, SIL can offer benefit as a protective agent against MSG-induced hepatotoxicity in rats. In this study, SIL caused no conspicuous changes in any of the evaluated parameters. On the other hand, MSG caused vivid increases in body and liver weights in rats. Similarly, Bernaje and others orally administered MSG (200-600 mg/kg body) for 28 days in rats and documented increased body weight (Banerjee et al., 2012). Also, MSG (100 mg- 4 g /kg/day) for 60 days was shown to increase body weight in rats (Sreejesh and Sreekumaran, 2018). Ezeokeke and Ezekwe (2017) documented increased liver weight in MSG (0.6 mg/g body weight) administered rats which is consistent our findings. The increased in body and liver weights caused by MSG may be the consequences of increased body mass and inflammation, respectively. However, in this study, body and liver weights were restored in SIL supplemented rats.

In addition, this study assessed the impact of MSG on selected and specific serum liver biochemical markers (ALT, AST, ALP, GGT, LDH CB and TB) and liver structure. MSG notably impaired serum liver biochemical markers marked by the elevated levels of serum ALT, AST, ALP, GGT, LDH CB and TB. It perturbed liver structure by causing hepatocellular necrosis. The elevated levels of the aforementioned markers in MSG-administered rats shows a dysfunctional hepatocellular metabolism as a consequence of compromised liver activity. The hepatocellular necrosis caused by MSG attests to impaired structural integrity of the liver. The findings in MSG-administered rats' correlate with the elevated levels of the aforementioned biochemical markers in Sprague Dawley adult male rats administered with MSG (35 mg/kg/day) for 14 days reported by Pal and others (Pal et al., 2020). Also, MSG (2, 4 and 8 g/kg) conspicuously elevated serum levels of the aforementioned markers as documented by Omogbiya *et al.* (2020). The observed hepatocelluar necrosis caused by MSG is in resonance with similar reports (Eid et al., 2019). It is fascinating to know that SIL supplementation restored serum biochemical markers and the structural integrity of the liver in a dose-related fashion.

It has been suggested that MSG causes tissue injuries through increased cellular oxidative perturbation (Hazzaa et al., 2020), therefore, oxidative stress markers were evaluated in this study. It was observed that MSG visibly increased MDA and depleted antioxidants (GSH, SOD, GPX and CAT) activities in the liver of rats. The observed increased MDA level is a sign of lipid peroxidation which connotes the breakdown of poly unsaturated fatty acids whereas decreased antioxidants obviously attests to oxidative stress in the liver of MSG-administered rats. When polyunsaturated fatty acids in cells are assaulted by reactive oxygen species, by-products of aldehyde such as MDA are produced. These by-products can expand from their origins to intracellular and extracellular targets, amplifying oxidative stress impact, thus facilitating more damage (Banerjee et al., 2021). Remarkably, in this work, SIL supplementation restored liver MDA and antioxidant levels in a dose-related fashion. Studies showed that MSG can disrupt the body's physiological state via the up-regulation of intracellular free radicals and electrophiles, causing oxidative stress and pro-inflammatory response mediated cellular injury. The production of reactive oxygen species can stimulate and enhance lipid peroxidation, disrupting the intracellular antioxidant/free radical balance (Banerjee et al., 2021). Many studies have attributed MSG-induced hepatocellular injuries such as fibrosis, hepatitis, steatosis, and necrosis to the formation of oxidative moieties (Omogbiya et al., 2020).

SIL is an antioxidant that has vital and functional chemical compositions such as polyphenols, flavonolignans, and flavonoids. It scavenges free radicals, up-regulates liver antioxidants, proteins and phospholipids syntheses and down-regulates lipid peroxidation in hepatocytes (Aghemo et al., 2022). The aforementioned actions can prevent

cellular damage, stabilize cell membranes, and reduce membrane permeability. The phenolic content of SIL which is the basis of its liver protective and antioxidant activity produces stable compounds with free radicals, thereby curtailing their deleterious effects (Aghemo et al., 2022). SIL has anti-inflammatory property that limits the production of inflammatory mediators and inflammatory metabolites which can prevent hepatic fibrosis and cirrhosis closely linked to inflammation (Basu et al., 2023). It was concluded that, SIL supplementation exhibits protective effect against MSG-induced hepatotoxicity in rats.

Acknowledgements

The authors appreciate the staff of the animal house of the Department of Pharmacology and Toxicology, Faculty of Pharmacy, Madonna University, Nigeria

Conflict of interest statement

The authors declare no conflict of interest.

Funding sources

There are no sources of funding to declare

5. Reference

Aebi H, 1974. Catalase. In Methods of enzymatic analysis. Academic Press.;673- 84

Aghemo A, Alekseeva OP, Angelico F, Bakulin IG, Bakulina NV et al., 2022 Role of silymarin as antioxidant in clinical manangement of chronic liver disease: a narrative review. Annals Med 54 (1) 1548-1560.

Atef H, EL-Morsi DA, EL-Shafey M, EL-Sherbiny M, EL-Kattawy, HA, Fahmy EK, and Saeed AA, 2019. Monosodium Glutamate Induced Hepatotoxicity and Oxidative Stress: Pathophysiological, Biochemical and Electron Microscopic Study. Med. J. Cairo Univ., 2019; 87 (1) 397-406.

Banerjee A, Mukherjee S, Maji BK, 2021. Monosodium glutamate causes hepato-cardiac derangement in male rats. Hum Exp Toxicol. 40(12):S359-S369.

Basu A, Namporn T, Ruenraroengsak P, 2023. Critical review in designing plant-based anticancer nanoparticles against hepatocellular carcinoma. Pharm, 15(6), 1611.

Bayram HM, Akgoz HF, Kizildemir O, Ozturkcan A, 2013. Monosodium Glutamate: Review on Preclinical and Clinical Reports, Bio Res App. Chem, (13), 2; 149; 1-27.

Buege JA, Aust SD, 1978. Microsomal lipid peroxidation. In: Methods in Enzymology Vol. 52 (C). S. Fleischer, L. Parker (eds), Academic Press, New York, pp. 302 – 310.

Cheemerla S, Balakrishnan M, 2021. Global epidemiology of chronic liver disease. Clin Liver Dis, 17(5), 365–370.

Eid RA, Al-Shraim M, Zaki MS, Kamar SS, Abdel Latif NS *et al.,* 2019. Vitamin E protects against monosodium glutamate-induced acute liver injury and hepatocyte ultrastructural alterations in rats. Ultrastruct Pathol.;43(4-5):199-208.

El-Morsi HA, El-Sherbiny ME, Fahmy HA, Saeed AA, 2019. Monosodium Glutamate Induced Hepatotoxicity and Oxidative Stress: Pathophysiological, Biochemical and Electron Microscopic Study. *The Med J Cairo Uni.* 87 (1) 397-406

Ezeokeke CT, Ezekwe OC, 2017. Effect of monosodium glutamate on the liver and kidney function of adult albino rats and the protective potentials of vitamin E. J of Diet Ass Nig. 8; 34-43.

Fu Y, Zhou Y, Shen L, Li X, Zhang *et al.,* 2022. Diagnostic and therapeutic strategies for non-alcoholic fatty liver disease. Frontiers Pharmacol, 13, 973366.

Gao X, Xiao ZH, Liu M, Zhang N, Khalil MM, Gu, CQ, Qi DS, and Sun L, 2018. Dietary Silymarin Supplementation Alleviates Zearalenone-Induced Hepatotoxicity and Reproductive Toxicity in Rats Xin J Nutr;148:1209–1216

Gillessen A, Schmidt HHJ, 2020. Silymarin as supportive treatment in liver diseases: A narrative review. Adv in Ther, 37(4), 1279–1301.

Guide for the Care and Use of Laboratory Animals. 2011.8th edition.,

Hajihasani MM, Soheili V, Zirak MR, Sahebkar A, Shakeri A. 2020. Natural products as safeguards against monosodium glutamate-

induced toxicity. Iran J Basic Med Sci.; 23(4):416-430

Hazzaa SM, El-Roghy ES, Abd Eldaim MA *et al.*2020. Monosodium glutamate induces cardiac toxicity via oxidative stress, fibrosis, and P53 proapoptotic protein expression in rats. Environ Sci Pollut Res 27, 20014-20024.

Kadoglou NP, Panayiotou C, Vardas, M, Balaskas N, Kostomitsopoulos NG, *et al.*, 2022. comprehensive review of the cardiovascular protective properties of Silibinin/Silymarin: A new kid on the block. Pharm. 15(5), 538.

Koushki M, Yekta RF, Amiri-Dashatan N, 2023. Critical review of therapeutic potential of silymarin in cancer: A bioactive polyphenolic flavonoid. J Funct Foods, 104, 105502.

Li L, Zeng, Z. Live imaging of innate and adaptive immune responses in the liver. Frontiers Immunol, 2020; 11, 564768.

Liu Y, Hao C, Li L, Zhang H, Zha W, Ma L *et al.,* 2023. The role of oxidative stress in the development and therapeutic intervention of hepatocellular carcinoma. Cur Can Drug Tar. 23(10), 792–804.

Marceddu R, Dinolfo L, Carrubba A, Sarno M, Di Miceli G, 2022. Milk thistle (*Silybum marianum* L.) as a novel multipurpose crop for agriculture in marginal environments: A review. Agronomy, 12(3), 729.

Mudd TW, Guddati AK, 2021. Management of hepatotoxicity of chemotherapy and targeted agents. Am J Cancer Res. 15;11(7):3461-3474.

Oda SS, El-Ashmawy IM, 2012. Protective Effect of Silymarin on Mercury-Induced Acute Nephro-Hepatotoxicity in Rats. Global Veter. 9 (4): 376-383.

Omogbiya AI, Ben-Azu B, Eduviere AT, Eneni AO, Nwokoye PO *et al.,* 2020. Monosodium glutamate induces memory and hepatic dysfunctions in mice: ameliorative role of Jobelyn® through the augmentation of cellular antioxidant defense machineries. Toxicol Res. 23;37(3):323-335.

Pal LC, Kumar A, Pande V, Rao ChV, 2020. Hepatoprotective Effect of Bioactive Fraction of Lagerstroemia speciosa (L.) Pers. Bark Against Monosodium Glutamate-Induced Phcogj.com Liver Toxicity. Pharmacogn J.;12(6)1630-40.

Rotruck JT, Pope AL, Ganther HE, 1973. Selenium: Biochemical role as a component of glutathione peroxidase purification assay. Sci.; 179:588–90

Sedlak J, Lindsay RH, 1968. Estimation of total, protein-bound, and nonprotein sulfhydryls groups in tissue with Ellman's reagent. Anal Biochem. 25:192–205.

Shakya AK, 2020. Drug-induced Hepatotoxicity and Hepatoprotective Medicinal Plants: A Review. Indian J of Pharm Edu and Res. 54(2):234-50

Shinde M, Mohan M. 2022, Protective efficacy of Murraya koenigii aqueous extract against monosodium glutamate-induced hepatotoxicity in Wistar rats. Indian J Nat Prod and Res; 13(2), 188-196

Sreejesh PG, Sreekumaran E, 2018. Effect of monosodium glutamate on striato-hippocampal acetylcholinesterase level in the brain of male Wistar albino rats and its implications on learning and memory during aging. Biosci Biotech Res Comm., 11, 76-82,

Sun M, Zigman S, 1978. An improved spectrophotometric assay for superoxide dismutase based on epinephrine autoxidation, Anal Biochem, Vol 90 (1)81-89.

Tao L, Qu X, Zhang Y, Song Y, Zhang S X, 2019. Prophylactic therapy of silymarin (milk thistle) on antituberculosis drug-induced liver injury: A meta-analysis of randomized controlled trials. Canadian J Gastroenterol and Hepatol, 3192351.

Zanfirescu A, Ungurianu A, Tsatsakis AM, Nițulescu GM, Kouretas D *et al.,* 2019. A review of the alleged health hazards of monosodium glutamate. Compr Rev Food Sci Food Saf. J18(4):1111-1134.