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Improving Effects of Shilajit on Monosodium Glutamate Induced Testicular Changes in Male Mice

Hadeel Kamil Khaleel[a,*], Saja Muzahem Thaker[b], & Haneen Abdulsalam Ali[c]

[a]*Department of Basic Science, College of Dentistry, Al-Iraqia University, Baghdad, Iraq*
[b]*Department of Microbiology, College of Science, Al Karkh University of Science, Baghdad, Iraq*
[c]*College of Medicine, Ibn Sina University of Medical and Pharmaceutical Science, Baghdad, Iraq*

Abstract

Several health conditions are thought to be associated with monosodium glutamate (MSG). Nowadays, a lot of products depend on monosodium glutamate as a flavour enhancer and food additive. The usage of MSG is still debatable, even though it has been considered a safe food additive. A delayed excitotoxicity and tissue-damaging dietary additive, MS.G, can produce large quantities of free radicals that cause metabolic disturbance and damage several organs. The study aimed to investigate the impact of monosodium glutamate on the testis of mice and the significance of shilajit (Shi) in improving the damage. Fifty male albino mice weighing between 18–22g were used in the study. The animals were randomly selected into five groups. Group A received distilled water as a control, group B give (2g/ kg) from (MS.G), and group C gives (2g/ kg) of (MS.G) and (100mg\ kg) of shilajit (Shi), group D gives (4g/ kg) of (MS.G), and group E gives (4g/ kg) of (MS.G) and (200mg/ kg) of shilajit (Shi), orally for 2 weeks. Blood collected for hormone biochemical examination. The mice were sacrificed at the end of the experiment, and the testes were separated for histological study. The results showed a significant decrease in testis weight in groups B, C, and D compared to the control, while no change in group E. Oxidative stress, including MDA and SOD, showed a normal level in the group B and C, whereas increased significantly increased in the group D and E. TNF-α inflammatory cytokines increased significantly in the treated groups with low and high concentrations of MSG. The results indicated a significant decrease in the FSH and LH. While testosterone and progesterone levels showed a decrease in groups B, C, and D, group E was normal. The capsule thickness showed a significant increase in the tested groups, except for group E appeared relatively normal. Numerous histological alterations in the testis were discovered by the MSG groups, including disarray of seminiferous tubules, decreased sperm in the lumen, Hyalinization with necrosis of Leydig's cells and destruction of intermediate connective tissue cells, and increased thickness of the testis capsule. Spermatogonia cells slouching in the seminiferous tubule lumens, destruction of Sertoli cells, necrosis of spermatogonia, and macrophage infiltration in the seminiferous tubule lumen, degeneration and destruction of intermediate connective tissue cells, disorganization of spermatogonia and spermatocytes, slouching of spermatocytes in the seminiferous tubule lumen, Congestion of blood vessels, and infiltration of inflammatory cells in the lumen of tubules. While the E groups exhibit extremely typical seminiferous tubules, hyalinization of interstitial tissue with destruction of Leydig's cells, normal capsule, some vacuoles or spaces between spermatocytes, virtually typical spermatogonia, few amount of sperm, and relatively normal spermatocytes. In conclusion, shilajit (Shi) acts synergistically in reducing MSG-induced testicular changes via anti-inflammatory and anti-degeneration effects.

Keywords: ELISA, Histological, Hormones, Monosodium glutamate, Shilajit.

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1. Introduction

Unhealthy processed-food is now more widely available to society due to changes in lifestyle, work pressure, and socialization (Cortes et al., 2021). As well as, it has led to increasing exposure to taste enhancers, which improves the processed food's palatability (Toyama et al., 2008). One significant addition that is frequently used worldwide is monosodium glutamate. Glutamic acid and sodium combine to form monosodium glutamate (MSG) (Wijayasekara & Wansapala, 2017). Moreover, like naturally occurring glutamate in meals like stews and meat soups, it is most typically

* Corresponding author.
E-mail address: hadeel.k.khaleel@aliraqia.edu.iq

used as a flavouring enhancer in the production of foods, adding a taste to food to intensify its meaty, savoury flavour (Jinap & Hajeb, 2010).

Glutamic acid is an amino acid that occurs naturally in food (Nguyen Thuy et al., 2020). MSG is harmful to consumers when taken in large amounts, according to some reports (Sharma, 2015). Along with headaches, flushing, sweating, numbness, asthma, muscle sensitivity, and weakness are some of the symptoms that MSG may cause (Aanuoluwapo Adeleke et al., 2022). MSG consumption has been linked to several illnesses, including atopic dermatitis, ventricular arrhythmia, neuropathy, asthma, urticaria, and stomach pain, in addition to these MSG symptom complexes (Baad-Hansen et al., 2010). The mechanisms by which MSG consumption causes obesity include oxidative stress, leptin resistance, and hyperlipidaemia (Banerjee et al., 2021). In addition, a hypothalamic lesion, and elevated expression of the gamma and alpha peroxisome proliferator-activated receptors. In the same way, consumption of MSG reduced the amount of beta cells in the pancreas, raised metabolic rates, and oxidative stress (Api, 2004). Furthermore, it reduced the transport of glucose and insulin into skeletal muscle and adipose tissue, reduced insulin receptors, caused insulin insensitivity, and caused severe hyperinsulinemia (Nahok et al., 2021). Due to its alleged health benefits, shilajit, a natural chemical with ancient therapeutic roots, is being employed more and more in contemporary supplements (Alqarni et al., 2025). Comprehensive chemical characterization is lacking, nevertheless, especially when it comes to inorganic anions (Kamgar, Zembrzuska, Lorenc, et al., 2025). A natural ingredient used in both conventional and alternative medicine, shilajit has become well known as an essential part of dietary supplements (Kamgar, Zembrzuska, Zembrzuski, et al., 2025). Shilajit maintains the Bax/Bcl2 ratio, promotes germ cells proliferation, and Sertoli cells activity (N-Cadherin and β-Catenin). Moreover, enzymes such as 3β-HSD and 17β-HSD are activated. Further lowers oxidative stress, lipid peroxidation and raises antioxidant enzyme (Rajpoot et al., 2024). Lack of knowledge about its composition has raised questions about its usage in supplementary medicine. The study aims were to investigate whether administering shilajit (Shi) at low and high doses could mitigate the testicular damage caused by monosodium glutamate and to examine the potential of shilajit to enhance testicular tissue and antioxidant roles in male mice about the oxidative damage caused by MSG.

2. Materials and Methods

2.1. Ethical Approve

Experimental animals and assessments were permitted under the scientific council of the College of Dentistry\ Al-Iraqia University, ethical code number (ESA&HER-09-07-2025). 50 white male mice, aged between 60 to 70 days, were used and housed in typical cages of the specialized laboratory under controlled humidity. They lived in residences for animals with a light-dark cycle, and without difficulties obtaining water, and food in the laboratory (Khaleel, 2019).

2.2. Monosodium Glutamate preparation

To prepare monosodium glutamate, the powder was dissolved in distilled water to obtain the following 2 g and 4 g doses. The solution was stored in a container for use in the experiment. The concentrations were used according to the body weight (Abdulghani et al., 2022; Omogbiya et al., 2021).

2.3. Shilajit preparation

Each tablet contains 200 mg of pure shilajit. It contains 80% of humic acid and 20% of fatty acids, proteins, and amino acids(Kamgar, Kaykhaii, et al., 2025). In accordance with (Rajpoot et al., 2025), the tablets were dissolved in distilled water to obtain the subsequent 100 and 200 mg/kg of shilajit, then kept in a container that was utilized for the experiment. The brand of tablets from the pure Himalayan company, USA.

2.4. Experimental Design

The experimental mice were selected into 5 equal groups as follows:

a. Control A: received distilled water by gavage tube orally for 2 weeks.
b. Group B: administered 2g\kg of MS.G.
c. Group C: administered 2g\kg of MS.G and 100mg\ kg. of shilajit.
d. Group D: administered 4g\kg of MS.G.
e. Group E: administered 4g\kg of MS.G and 200mg\ kg. of shilajit.

Orally treated groups were administered by gavage tube for 2 weeks.

2.5. Samples Collection

The experimental animals were sacrificed after 12 hours following the final treatments. Heart punctures were used to obtain blood, then kept in a gel tube, and for parameter testing (Star et al., 2020). Serum was processed by being centrifuged at 4000 rpm for five minutes and stored at -80º C for 24 hours. The testes were surgically removed, meticulously cleaned to remove any remaining blood, and then precisely weighed (Hussein & Mustafa, 2024).

2.6. Determination of serum oxidative stress, inflammatory cytokines and hormone levels

Malondialdehyde (MDA) and Superoxide dismutase, TNF-α, hormones testosterone, progesterone, luteinizing (LH), and even follicle stimulating hormone (FSH) were determined using ELISA kits from Cusabio, USA. We employed quantitative sandwich enzyme immunoassay technology to apply assay concepts. A pre-selected antibody was placed on a microplate. Furthermore, samples and standards were pumped into the wells, where the fixed antibody binds to any antigen present (Mountjoy, 2021). Following the removal of any unconjugated materials, the wells were filled with the biotin-conjugated antibody. Once the wells had been cleaned, conjugated avidin horseradish peroxidase was added. Additionally, colour developed in responses to the interval of bound antigen during the first step, when a substrate the mixture was introduced to the wells, then washed to eliminate any unbound reagents. The intensity of colour was assessed, and colour development was halted (Ongaro et al., 2021). As directed, the reagent samples and standards were prepared. Each well received 100 µL of the standard or sample, which was incubated for two hours at 37 °C. Do not wash; the liquid was removed from each well. Moreover, each well received 100 µL of the biotin antibody (1x), which was incubated for one hour at 37 °C. Then, it was removed and cleaned three times (Huang et al., 2021). In addition, each well received 100 µL of horseradish peroxidase HRP-avidin (1x), which was incubated for one hour at 37°C. Furthermore, desired it five times and observed. Apply 90µL of tetra-methyl-benzidine to each well, then let's react for (15- 30) min at 37° C. Ultimately, (50µL) of the cease solution was inserted into each well. Readings were completed in five minutes at 450 nm (Tan et al., 2018).

2.7. Histological Examination

Testes were separated for analysis using histology techniques. The testes were fixed in a 10% formalin solution for 24 hours. The section preparation, which included dehydration by serial concentrations of ethyl. In addition, cleaning, and creating in paraffin to get the blocks, was employed to prepare fixed testis sections. The following histological dyes were used to stain sections by cut the blocks at an average of 5 µm (Suvarna et al., 2019). General histological investigations conducted with haematoxylin and eosin (H&E) (Khaleel & Haba, 2025). A Biobase compound microscope with a digital camera was utilized to examine the stained sections. Sections of the treated groups were compared with those of the control group for results evaluation.

2.8. Statistical Analysis

Statistical software 2019 was utilized to analyse the experimental data. One way assessment of variability test, and the least significant difference were used to evaluate the findings (Karim et al., 2021). The values were considered statistically significant when the probability (P) was less than or equal to 0.05. The mean ± standard error is used to display the data (Khaleel, 2019).

3. Results

The results indicated that, in compared to the control groups (0.37±0.01) g, the group B received MS.G at 2g/ kg had a substantial P≤ 0.05 decrease in the testis (0.29±0.01)g. In addition, group C gives 2g/ kg of (MS.G) with 100mg/ kg of (Shi.), which displayed a presumed decrease in the testis weight (0.32±0.01)g. When group D was give 4g/ kg of (MS.G), the testis weight (0.30±0.02)g was thought to have decreased. However, with the addition of 4g/ kg of (MS.G) and 200mg/ kg of (Shi.), group E showed normal in testis weight (0.36±0.01)g, based on figure 1.

The results of the oxidative stress in Figure 2A showed that there are no changes in the MDA concentration of group B (30.58±2.69) nmol/ml and C (27.34±2.96) nmol/ml compared to control (25.90±2.38) nmol/ml. Whereas, a high concentration of MSG caused a significant increase in MDA group D (43.56±4.54) nmol/ml and E (32.63±5.94) nmol/ml. SOD concentration in Figure 2B revealed that group B (120.22±6.96) U/ml and C (118.15±3.98) U/ml were the same in the control (106.71±5.19) U/ml. While the concentration of SOD increased significantly in group D (139.36±6.68) U/ml and E (132.12±4.11) U/ml.

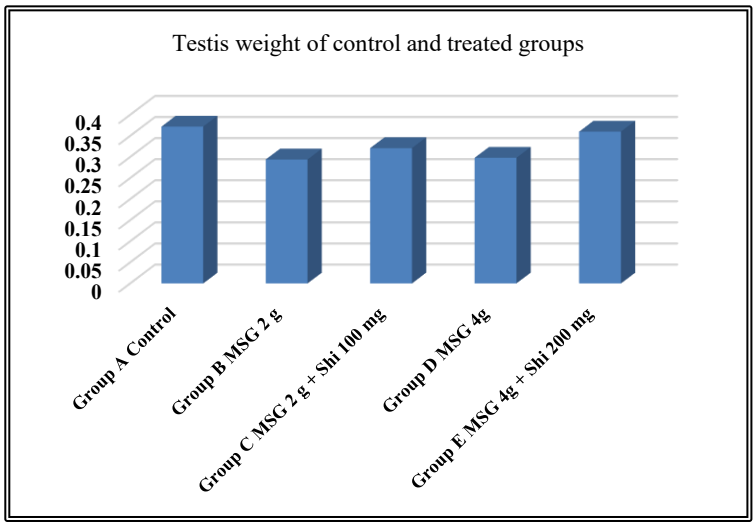

Figure 1. Effect of (MSG) and shilajit (Shi) on the testis weight.

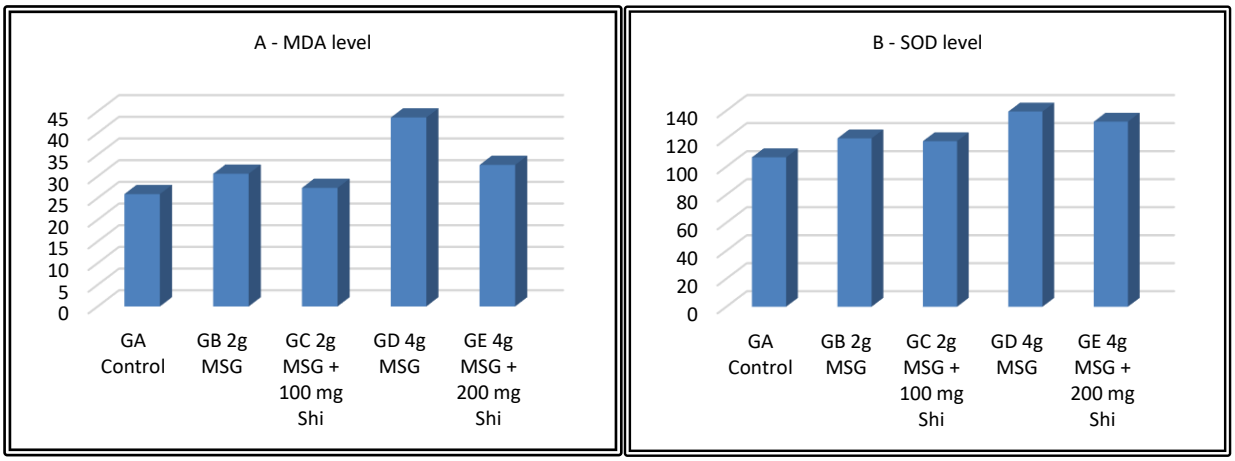

Figure 2. Effect of MSG and shilajit on the oxidative stress: A- MDA and B- SOD.

Administration of MSG had an adverse effect on inflammatory cytokines, TNF-α showed increased significantly in a group B (8.69±1.69) pg/ml and group C (6.77±1.09) pg/ml compared to control group (5.41±0.72) pg/ml. furthermore, at high concentration of MSG showed significant elevation in group D (12.54±1.47) pg/ml and E (8.00±1.82) pg/ml (Figure 3).

By contrast to the control group (45.31 ± 1.58) ng/ml, the mean correlated level of Follicle stimulating hormone (FSH) in group B (15.65 ± 0.52) was substantially decreased. Meanwhile, when adding 100 mg/kg of shilajit in group C with 2g/kg of MSG appears relatively decreased (17.03±0.50)ng/kg of FSH. The high concentration of MSG in group D caused a high decrease in FSH (23.40±1.35) ng/ml. In group E, the utilization of 200 mg/kg of shilajit along with 4g/kg of MSG results in a slight decrease (39.70 ± 2.77) ng/kg (Table1).

The mean associated level of Luteinizing hormone (LH) in group B (3.95± 0.17)ng/ml was significantly lower as relative of the control (7.89 ± 0.43) mIU/ml. In group C, the addition of 100 mg/kg of shilajit beside 2g/kg of MSG led to a relatively lower LH level (4.29± 0.25) mIU/ml. Group D elevated MSG content created a significant reduction in LH (5.93± 0.29)mIU/ml. Group E experiences a simple decrease (6.97 ± 0.19) mIU/ml when 200 mg/kg of shilajit and 4g/kg of MSG are used (Table 2).

Compared to the control group (4.38 ± 0.26)ng/ml, the group B mean associated level of testosterone (1.53 ± 0.09)ng/ml was considerably lower. In group C, a comparatively reduced testosterone level (2.55 ± 0.09)ng/ml resulted after administering the animals with 2g/ kg of (MS.G) and 100mg/ kg of (Shi). Testosterone levels in group D significantly decreased (3.48 ± 0.37) ng/ml. while, when use 200mg/ kg of shilajit plus 4g/ kg of (MS.G) showed normal seem the control (Table 3).

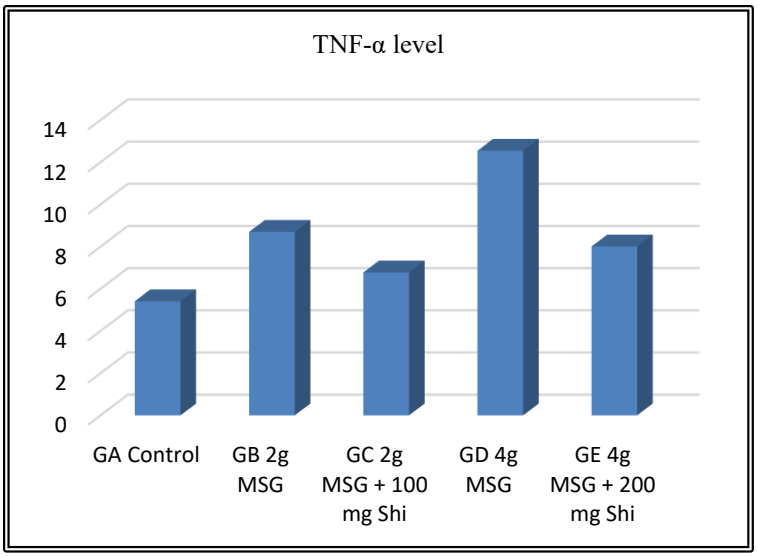

Figure 3. Effect of MSG and shilajit on the TNF-α.

Conversely, the group B mean associated level of progesterone (1.61 ± 0.15) ng/ml was significantly reduced than the control group (5.79 ± 0.45) ng/ml. In group C, mice were give 100 mg/kg of shilajit and 2 g/kg of MSG, which led to a relatively lower progesterone level (3.23 ± 0.22) ng/ml. Group D experienced a significant throw in progesterone levels (2.74 ± 0.29) ng/ml. However, when 200 mg/kg of shilajit and 4 g/kg of MSG were used, the progesterone levels were normal (Table 4).

Table 1. Effect of MSG and shilajit in FSH hormones

Groups	FSH. (ng/ ml.)	Maximum (ng/ ml.)	Minimum (ng/ ml.)	Standard Deviation
Group A control	45.31 ± 1.58a	51.44	33.58	5
Group B (2 g/kg) MSG	15.65 ± 0.52b	18.02	13.20	1.66
Group C (2 g/kg) MSG + 100 mg\km Shilajit	17.03 ± 0.50b	19.03	14.23	1.59
Group D (4 g/kg) MSG	23.40 ± 1.35c	29.45	18.32	4.27
Group E (4 g/kg) MSG + 200 mg/kg Shilajit	39.70 ± 2.77d	49.82	29.11	8.75
LSD	4.503			

Values are exhibited as mean ± SE, several letters in the same column varied significantly, P≤0.05.

Table 2. Effect of MSG and shilajit in LH hormones

Groups	LH. (mIU/ ml.)	Maximum (mIU/ ml.)	Minimum (mIU/ ml.)	Standard Deviation
Group A control	7.89 ± 0.43a	9.98	6.35	1.36
Group B (2 g/kg) MSG	3.95 ± 0.17b	4.85	3.29	0.55
Group C (2 g/kg) MSG + 100 mg\km shilajit	4.29 ± 0.25b	5.27	3.25	0.78
Group D (4 g/kg) MSG	5.93 ± 0.29c	7.43	4.22	0.90
Group E (4 g/kg) MSG + 200 mg/kg shilajit	6.97 ± 0.19d	7.88	6.03	0.61
LSD	0.801			

Values are exhibited as mean ± SE, several letters in the same column varied significantly, P≤0.05.

Table 3. Effect of MSG and shilajit in Testosterone hormones

Groups	Testosterone (ng/ ml.)	Maximum (ng/ ml.)	Minimum (ng/ ml.)	Standard Deviation
Group A control	4.38 ± 0.26a	5.33	2.48	0.82
Group B (2 g/kg) MSG	1.53 ± 0.09b	1.95	1.22	0.29
Group C (2 g/kg) MSG + 100 mg\km shilajit	2.55 ± 0.09c	2.99	2.13	0.28
Group D (4 g/kg) MSG	3.48 ± 0.37d	6.23	2.01	1.16
Group E (4 g/kg) MSG + 200 mg/kg shilajit	4.65 ± 0.51a	6.92	2.43	1.60
LSD	0.878			

Values are exhibited as mean ± SE, several letters in the same column varied significantly, P≤0.05.

Table 4. Effect of MSG and shilajit in progesterone hormones

Groups	Progesterone (ng/ ml.)	Maximum (ng/ ml.)	Minimum (ng/ ml.)	Standard Deviation
Group A control	5.79 ± 0.45a	7.78	4.25	1.41
Group B (2 g/kg) MSG	1.61 ± 0.15b	2.27	1.02	0.47
Group C (2 g/kg) MSG + 100 mg\km shilajit	3.23 ± 0.22c	4.06	2.17	0.69
Group D (4 g/kg) MSG	2.74 ± 0.29c	4.93	2.02	0.91
Group E (4 g/kg) MSG + 200 mg/kg shilajit	6.17 ± 1.02a	14.06	3.75	3.21
LSD	1.499			

Values are exhibited as mean ± SE, several letters in the same column varied significantly, P≤0.05.

The results showed that the capsule thickness (75.41±1.31)µm raised significantly (P≤ 0.05) in the treated group (B), which received (MS.G) at 2g/ kg, in comparison in the control groups (42.53±1.58)µm. Additionally, group c showed a supposed increase in the testis capsule thickness (47.62±0.68)µm after administering 2g/ kg of (MS.G) along with 100mg/ kg of (Shi.). The capsule thickness (76.67±1.12)µm was assumed to have elevated when group D received 4g/kg of MSG. But when 200mg/ kg of (Shi) and 4g/ kg of (MS.G) were added, group E testis capsule thickness increased to (54.25±1.74)µm (Figure 4).

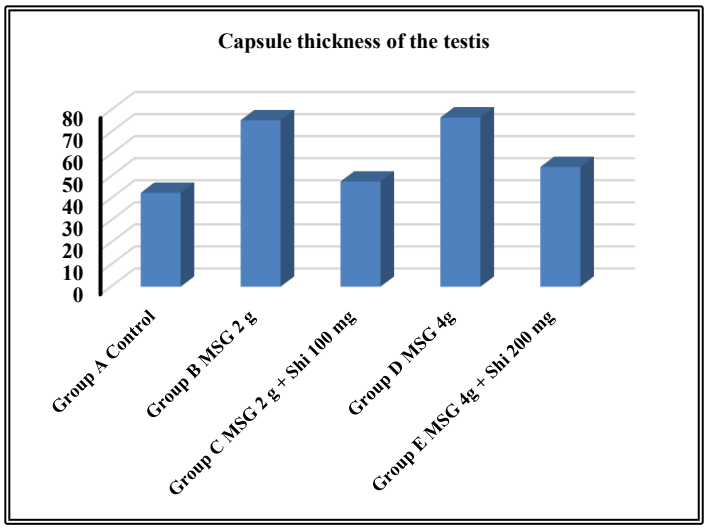

Figure 4. Effects of Monosodium Glutamate (MSG) and shilajit (Shi) on the capsule thickness of the testis

The results of the histological sectioned showed the testis of control group contain normal capsule, and normal seminiferous tubules. Each seminiferous tubule contains basal lamina, sertoli cells, spermatogonia, spermatocytes, spermatids, and sperms in the lumen (Fig 5). However, the animals in (group B) treated with 2g/ kg of (MS.G) displays disarray of seminiferous tubules, decreased sperms in the lumen, and Hyalinization with necrosis of Leydg's cells and destruction of intermediate connective tissue cells, increased thickness of the testis capsule. Spermatogonia cells slouching in the seminiferous tubule lumens, and destruction of sertoli cells, necrosis of spermatogonia, and macrophage infiltration in the seminiferous tubule lumen (Fig 6). On the other hand, group C treated by 2g/ kg of (MS.G) and 100mg/ kg of (Shi) performed disruption of seminiferous tubules with moderate sperms in the lumen, and hyalinization with necrosis of Leydg's cells and destruction of intermediate connective tissue cells, increased thickness of the testis capsule, necrosis of sertoli cells, Spermatogonia cells and spermatocytes slouching in the seminiferous tubule lumen (Fig 7). The testis of group D treated with 4g/ kg of (MS.G) appeared disarray of seminiferous tubules with a few amount of sperms in the lumen, and Hyalinization and destruction of intermediate connective tissue cells, increased thickness of the testis capsule, disorganization of spermatogonia and spermatocytes, slouching of spermatocytes in the seminiferous tubule lumen, Congestion of blood vessels, Infiltration of inflammatory cells in the lumen of tubules (Fig 8). While, the E groups treated with 4g/ kg of (MS.G) with 200mg /kg of (Shi) exhibit extremely typical seminiferous tubules, hyalinization of interstitial tissue with destruction of Leydig's cells, normal capsule, some vacuoles or spaces between spermatocytes, typical spermatogonia, few amount of sperms and relatively normal spermatocyte (Fig 9). The histological changes were summarized in the table 4.

Table 5. Histological changes scoring in the testicular of control and treated animals

Histological changes N = 10	GA Control		GB 2 g MSG		GC 2g MSG 100 mg shilajit		GD 4g MSG		GE 4g MSG 200 mg shilajit	
	N	%	N	%	N	%	N	%	N	%
Hyalinization	0	0	3	30	3	30	6	60	5	50
Necrosis	0	0	4	40	3	30	8	80	4	40
Sloughing	0	0	5	50	4	40	7	70	5	50
Congestion of blood vessels	0	0	7	70	5	50	9	90	6	60
Inflammatory cells infiltration	0	0	8	80	5	50	8	80	5	50
Capsule thickness	0	0	6	60	4	40	8	80	6	60

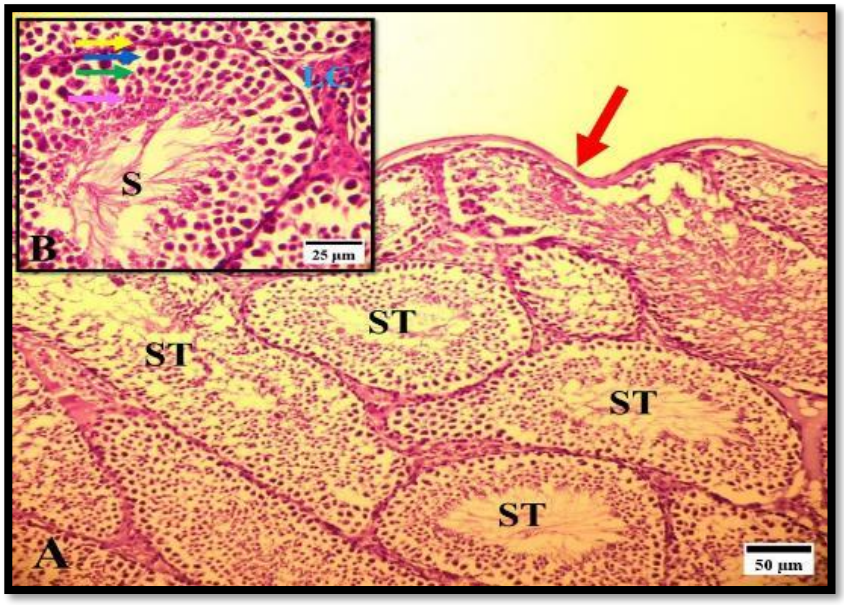

Figure 5. Micrograph in the testis of the control group displays: A: normal capsule (red arrow), and normally occurring seminiferous tubules tissue (ST). B: basal lamina (yellow arrow), spermatogonia (blue arrow), spermatocytes (green arrow), spermatids (pink arrow), and sperms (S), (H&E).

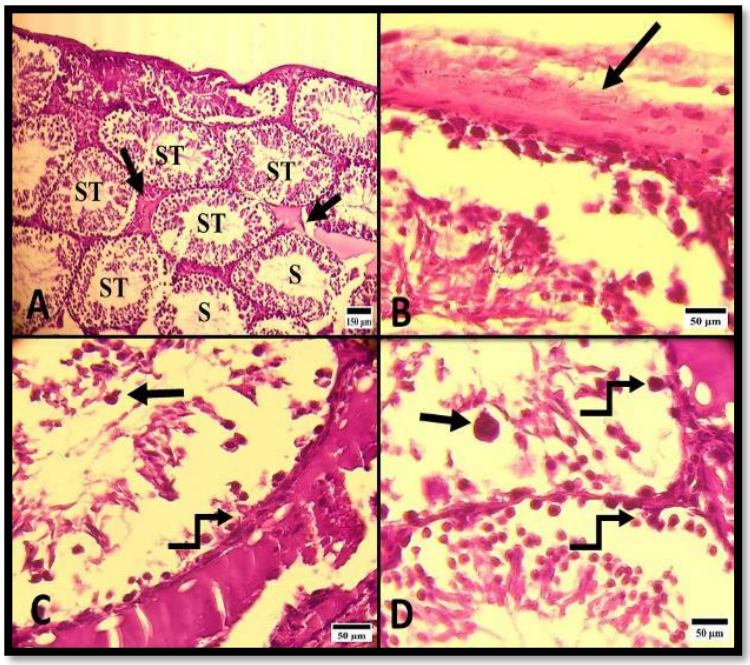

Figure 6. Micrograph in the testis of the group B displays: A: disarray of seminiferous tubules (ST), decreased sperms in the lumen (S), and Hyalinization with necrosis of Leydg's cells and degeneration of intermediate connective tissue cells (arrow). B: increased thickness of the testis capsule (arrow). C: Spermatogonia cells slouching in the seminiferous tubule lumen (arrow), and destruction of sertoli cells (elbow arrow). D: necrosis of spermatogonia (elbow arrows), Macrophage infiltration in the seminiferous tubule lumen (arrow), (H&E).

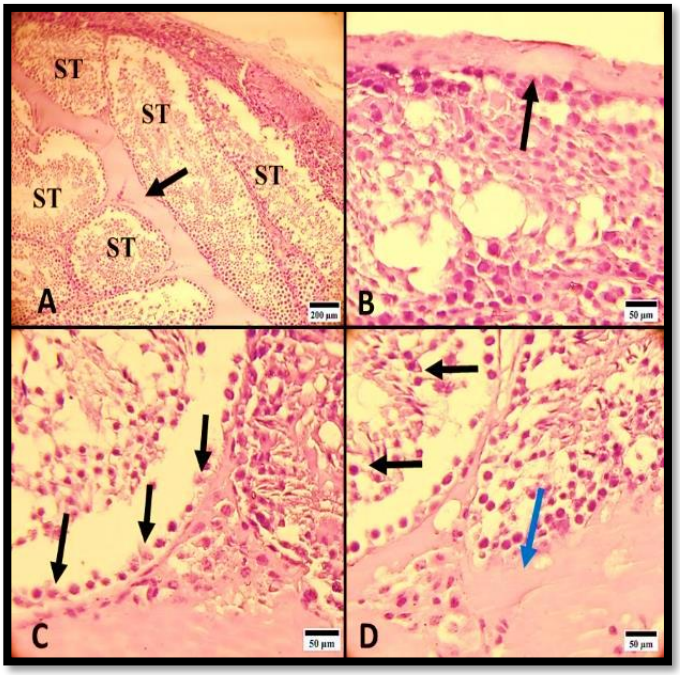

Figure 7. Micrograph in the testis of the group C displays: A: disarray of seminiferous tubules (ST) with moderate amount of sperms in the lumen, and Hyalinization with necrosis of Leydg's cells and destruction of intermediate connective tissue cells (arrow). B: Normally thickness of the testis capsule (arrow). C: necrosis of sertoli cells (arrows). D: Spermatogonia cells and spermatocytes slouching in the seminiferous tubule lumen (arrow), and hyalinization of interstitial tissue, (H&E).

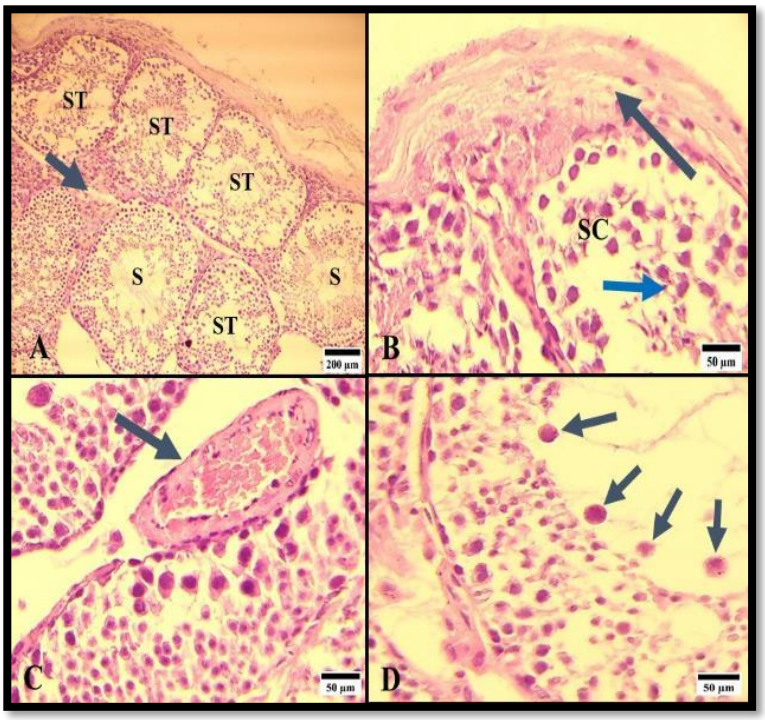

Figure 8. Micrograph in the testis of the group D displays: A: disarray of seminiferous tubules (ST) with a few amount of sperms in the lumen (S), and Hyalinization and degeneration of intermediate connective tissue cells (arrow). B: increased thickness of the testis capsule (arrow), disorganization of spermatogonia and spermatocytes (SC), slouching of spermatocytes in the seminiferous tubule lumen (blue arrow). C: Congestion of blood vessels (arrow). D: Infiltration of inflammatory cells in the lumen of tubules (arrows), (H&E).

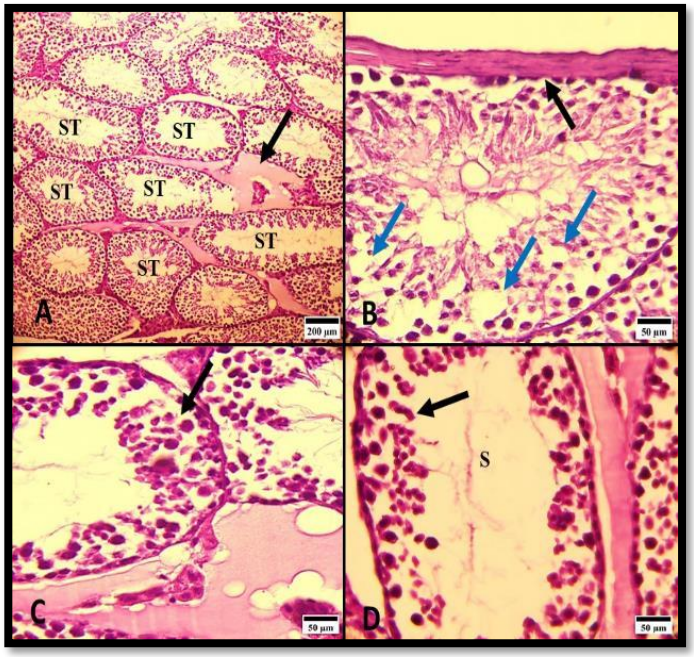

Figure 9. Micrograph in the testis of the group E displays: A: Extremely typical seminiferous tubules (ST), hyalinization of interstitial tissue with destruction of Leydig's cells (arrow). B: normal capsule (arrow), some vacuoles or spaces between spermatocytes (blue arrow). C: virtually typical spermatogonia (arrow). D: Few amount of sperms (S), relatively normal spermatocyte (arrow), (H&E).

4. Discussion

High concentration of MSG causes a decrease the testis weight due to response the tissue to MSG induced destruction (Ogunmokunwa & Ibitoye, 2025). *Nigella sativa* has tissue protective and antioxidant properties that served to minimize the testicular damage caused by MSG (Abd-Elkareem et al., 2021). Oxidative stresses, including MDA and SOD, increased significantly due to the high concentration of MSG, causing metabolic disorders and may cause the defences of antioxidants to decrease (M. Abdou et al., 2020). The current findings consistently showed an increase in TNF-α levels in the MSG-treated groups. This inflammatory induction following MSG administration is consistent with the findings of (M. Abdou et al., 2020), who noted that MSG administration caused oxidative stress in the treated animals. The consumed MSG led to a decrease the reproductive hormones, including FSH, LH, testosterone, and progesterone due to damage to testis tissue (Acikel-Elmas et al., 2023). According to other studies, animals tested on MSG showed decreased reproductive disorders when give L-carnitine because of its antioxidant actions (Koohpeyma et al., 2022). When additives damage GnRH cells, they lead to severe reproductive impairment and tissue changes in male rats (Wang et al., 2021). Testosterone and progesterone levels were reduced significantly in MSG groups, and elevated when selenium treated groups (Hamza & Diab, 2020). High concentration of food additives causes thickness of the capsule and hyalinization of the interstitial connective tissue between seminiferous tubules. These damages occur due to increased levels of oxidative stress markers and testis tissue responses, the damages can be reduced after vitamins C and E are taken (El Kotb et al., 2020). More researchers showed that the testis typical appearance was dispersed, and the seminiferous tubules showed varying degrees of atrophy overall, there was necrosis and degeneration in the spermatogenic cells, and the number of Leydig cells was reduced (Samir Nosseir et al., 2012). Testicular damage could be the cause of the alterations in the oxidant and hormonal levels, but when Roselle revealed ameliorative activity against the radicals (Gad et al., 2021). Glutathione and superoxide disorders cause testicular changes and degeneration when the animals give flavour enhancer. Apocynin is thought to avoid testicular deterioration by preserving the blood testis integrity (Acikel-Elmas et al., 2023). The levels of inflammatory markers were rise by MSG treatment, leading to infiltration of inflammatory cells in the seminiferous tubules, tissue damage of the testis, while Rutin's anti-inflammatory attributes supported mitigating the damage, suggesting that it had a protective impact (Abdel-Aty et al., 2025).

5. Conclusion

The ameliorative responses of shilajit against MSG induced testicular harmful effects in mice were noted in this work and can be assigned to their effective anti-inflammatory, and antioxidant behaviours. They act as a protective factor to the testis lesions brought on by dietary pollutants. A more thorough investigation of these compound molecular mechanism of action, long-term effects, and ideal dosage for therapeutic uses all sides need research.

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