




# Protective Effect of Tiger Nut (*Cyperus Esculentus*) Against Monosodium Glutamate-Induced Reproductive Dysfunction in Male Wistar Rats

## Yer Bademi (*Cyperus esculentus*)'nin Erkek Wistar Sıçanlarda Monosodyum Glutamat Kaynaklı Üreme Fonksiyon Bozukluğuna Karşı Koruyucu Etkisi

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Received/Geliş Tarihi: 24.09.2024  
Accepted/Kabul Tarihi: 20.02.2025  
Publication Date/Yayın Tarihi: 29.04.2025

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Cite this article: Ajani OS, Akinyemi AI, Akinniyi OO. Tiger nut (*Cyperus esculentus*) protects rats against monosodium glutamate-induced reproductive dysfunction in male wistar rats. *Vet Sci Pract.* 2025;20(1):16-23.

Atıf: Ajani OS, Akinyemi AI, Akinniyi OO. Yer Bademi (*Cyperus esculentus*)'nin Erkek Wistar Sıçanlarda Monosodyum Glutamat Kaynaklı Üreme Fonksiyon Bozukluğuna Karşı Koruyucu Etkisi. *Vet Sci Pract.* 2025;20(1):16-23.



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### ABSTRACT

Monosodium glutamate (MSG) is a widespread flavour enhancer linked to health risks, including male reproductive dysfunction. This study investigated tiger nut (*Cyperus esculentus*) as a potential protective agent against MSG-induced reproductive issues in male Wistar rats. Forty adult rats were divided into four groups: control, MSG-only (2 mg/g), tiger nut-only (500 mg/kg), and MSG+tiger nut combination (2 mg/g MSG + 500 mg/kg tiger nut). Treatments were administered orally for 28 days, with analyses conducted at days 14 and 28. Results showed significant variations in sperm parameters. At 14 days, the tiger nut group showed highest sperm motility (88.60±4.04%) and count (100.60±3.21×10<sup>6</sup>/mL), while MSG reduced sperm viability (70.00±4.69%). By 28 days, MSG significantly decreased sperm motility (41.80±4.92%) and viability (54.80±6.76%). MSG increased sperm abnormalities at 14 days (13.60±2.51%) but normalized by 28 days. The MSG+tiger nut combination eliminated certain sperm abnormalities like coiled tail and tail-without-head. Gonadometric parameters remained stable throughout the study, indicating tiger nut's ability to maintain testicular architecture despite MSG exposure. Initial body weight increases in the MSG group normalized by weeks 3-4. The study concludes that tiger nut juice significantly protects against MSG-induced low sperm quality in male Wistar rats, suggesting its potential as a protective supplement for populations with unavoidable MSG exposure. Future research should explore long-term effects and cellular mechanisms.

**Keywords:** *Cyperus esculentus*, fertility, monosodium glutamate, rat, spermatotoxicity.

### ÖZ

Monosodyum glutamat (MSG), erkek üreme fonksiyon bozukluğu da dahil olmak üzere sağlık riskleriyle ilişkilendirilen yaygın bir lezzet arttırıcıdır. Bu çalışmada, yer bademinin (*Cyperus esculentus*) erkek Wistar sıçanlarında MSG kaynaklı üreme sorunlarına karşı potansiyel koruyucu etkisi araştırılmıştır. Kırk yetişkin sıçan, kontrol, MSG (2 mg/g), yer bademi (500 mg/kg) ve MSG + yer bademi kombinasyonu (2 mg/g MSG + 500 mg/kg yer bademi) olmak üzere dört gruba ayrılmıştır. Uygulamalar 28 gün boyunca ağız yoluyla gerçekleştirilmiş; analizler 14. ve 28. günlerde yapılmıştır. Sonuçlar sperm parametrelerinde önemli değişiklikler olduğunu göstermiştir. 14. günde, yer bademi grubu en yüksek sperm hareketliliği (%88,60±4,04) ve sayısı (100,60±3,21×10<sup>6</sup>/mL) gösterirken, MSG uygulaması sperm canlılığını (%70,00±4,69) azaltmıştır. MSG, 28. günde sperm hareketliliğini (%41,80±4,92) ve canlılığını (%54,80±6,76) önemli ölçüde azaltmıştır. MSG, 14. günde sperm anormalliklerini artırmış (%13,60±2,51); ancak 28. günde normale dönmüştür. MSG+yer bademi kombinasyonu ise kıvrık kuyruk ve başsız kuyruk gibi bazı sperm anormalliklerini ortadan kaldırmıştır. Gonadometrik parametreler çalışma boyunca sabit kalmış, bu da yer bademinin MSG maruziyetine rağmen testis yapısını koruma yeteneğini göstermektedir. MSG grubundaki başlangıçtaki vücut ağırlığı artışları, 3-4. haftalarda normale dönmüştür. Bu çalışma, yer bademi suyunun erkek Wistar sıçanlarında MSG kaynaklı düşük sperm kalitesine karşı önemli ölçüde koruma sağladığı sonucuna vararak, kaçınılmaz MSG maruziyeti olan popülasyonlar için koruyucu bir takviye olarak potansiyelini ortaya koymaktadır. Gelecekteki araştırmalar uzun vadeli etkileri ve hücresel mekanizmaları araştırmalıdır.

**Anahtar Kelimeler:** *Cyperus Esculentus*, fertilité, monosodyum glutamat, sıçan, spermatotoksisite

## INTRODUCTION

Monosodium glutamate (MSG) is a widely used flavour enhancer, particularly prevalent in oriental cuisine and processed foods worldwide.<sup>1</sup> As a sodium salt of glutamic acid, it provides the distinctive umami taste that enhances food palatability.<sup>2</sup> Despite its Generally Recognized as Safe (GRAS) status by food safety regulatory agencies, mounting evidence suggests potential health risks associated with MSG consumption, including obesity, metabolic disorders, and notably, reproductive dysfunction.<sup>3</sup> Of particular concern are studies linking MSG to male reproductive health issues, including oligozoospermia, abnormal sperm morphology, and testicular damage.<sup>4,5</sup>

The search for protective agents against MSG-induced reproductive toxicity has gained importance, especially given MSG's ubiquity in modern diets. Tiger nut (*Cyperus esculentus*), a nutrient-rich tuber, has emerged as a promising candidate due to its unique nutritional profile.<sup>6</sup> Rich in fibre, proteins, essential fatty acids, vitamins C and E, and various minerals, tiger nut has traditionally been consumed in various forms, including raw, roasted, or as a beverage called "Horchata".<sup>7,8</sup> While its nutritional benefits are increasingly recognized, tiger nut's potential protective effects against reproductive toxicants remain largely unexplored.

Previous studies have examined either MSG's adverse effects on male reproduction or tiger nut's general health benefits separately. MSG has been shown to cause testicular damage through oxidative stress and inflammatory pathways.<sup>9-11</sup> While tiger nut has demonstrated antioxidant and anti-inflammatory properties,<sup>12</sup> However, the potential protective role of tiger nut against MSG-induced reproductive dysfunction represents a novel area of investigation. This research gap is particularly significant given the increasing exposure to MSG in modern diets and the need for practical, accessible protective measures.

Our study uniquely investigates the concurrent administration of MSG and tiger nut, examining whether tiger nut's nutritional and antioxidant properties could mitigate MSG-induced reproductive dysfunction in male Wistar rats. This approach differs from previous research by evaluating the protective rather than merely therapeutic potential of tiger nut, focusing on practical, readily measurable reproductive parameters, and examining the interaction between MSG and tiger nut in a controlled experimental setting. We hypothesized that tiger nut supplementation protects against MSG-induced reproductive dysfunction by improving sperm parameters

and maintaining normal gonadometric measures. This investigation aims to provide practical insights for populations with unavoidable MSG exposure and potential applications in animal breeding programs.

## MATERIALS AND METHODS

### Experimental Animals

This study was carried out according to the regulations and principles that govern both care and use of experimental animals for research purposes by the Animal Care and Use Ethics Committee at the University of Ibadan (Date: 16/5/2018, Approval No: UI-ACUREC/17/0069). Forty adults male Wistar rats (150 ± 10g) were housed under controlled conditions (35-36°C, 50 ± 15% humidity, 12:12 light-dark cycle). Animals were monitored daily for signs of stress or adverse reactions, including changes in feeding behaviour, activity levels, and coat condition. No unexpected events or adverse reactions were observed during the study period.

### Dosage Selection and Treatment

The MSG dosage (2 mg/g) was established through a weight-based calculation accounting for average daily dietary MSG exposure scaled to rat metabolism. This dose represents a moderate exposure level that allows for observation of potential reproductive effects while remaining within physiologically relevant bounds for food additive consumption. The tiger nut dose (500 mg/kg) was selected based on the measured nutrient content of tiger nut juice, specifically its antioxidant components and bioactive compounds, to provide sufficient protective capacity while maintaining safe consumption levels for daily administration. Both doses were determined to be within the practical oral administration range for rats of this size and age. Treatments were administered orally for 28 days, with careful monitoring of administration technique to ensure complete dosing. The 28-day duration was chosen to span at least two complete cycles of spermatogenesis in rats, allowing for observation of potential effects on sperm production and maturation.

### Materials and Preparation

MSG (98%) and tiger nut were sourced from Bodija Market, Ibadan, Nigeria (7.3775°N, 3.9470°E). All other chemicals used in the study were of the highest available grade in Nigeria. The tiger nut juice was prepared by crushing the tubers, followed by manual extraction of the juice (milk) using a 0.075 mm plastic sieve.

### Study Design

The 40 adult male Albino rats (Wistar strain) were randomly grouped into four (n=10). Group A was the

Control and received 0.5ml of distilled water orally. Group B, received oral MSG at 2 mg/g body weight orally, Group C received oral tiger nut at 500 mg/kg body weight while Group D received a combination of MSG at 2 mg/g and tiger nut at 500 mg/kg body weight. All treatments were administered for 28 days. Five rats from each group were humanely sacrificed 24 hours after both 14 and 28 days of treatment. Following the sacrifice, the testes and epididymis were harvested, and semen samples were collected for analysis. Additionally, gonadometric assessments were conducted.

### Gonadometric Analysis

The testes were detached from the epididymis as outlined by Ajani and Omoyeni.<sup>12</sup> Both the testes and epididymis were weighed using an Electronic Weight Scale (Ohaus Corporation, USA), while the length and diameter of the testes were determined using an Electronic Veiner Caliper (Mitutoyo Corporation, Japan) following the methodology outlined by Ajani and Omoyeni.<sup>12</sup>

### Semen Analysis

Semen samples were collected from the left caudal epididymis using a surgical incision method as described by Oyeyemi and Fayomi.<sup>13</sup> Briefly, the epididymis was carefully excised and placed in a pre-warmed (37°C) petri dish containing 1 mL of physiological saline (0.9% NaCl). Multiple incisions were made in the cauda epididymis using a sterile surgical blade (size 11) to release the spermatozoa.

For morphological assessment, a drop of semen was placed on a clean, pre-warmed (37°C) glass slide and stained using Wells and Awa staining method (Wells and Awa stain). The stained slides were examined under a light microscope at 400× magnification to evaluate sperm morphology.

Sperm viability was assessed using the eosin-nigrosin staining technique. The staining solution was prepared using 5% eosin Y and 10% nigrosin in distilled water. One drop of semen was mixed with two drops of the stain on a

pre-warmed slide, and a thin smear was prepared. After air-drying, the slides were examined under a light microscope at 400× magnification. Live spermatozoa remained unstained (white), while dead spermatozoa appeared pink.

For sperm motility assessment, 2-3 drops of semen were mixed with an equal volume of pre-warmed (37°C) 2.9% sodium citrate buffer (pH 7.4) on a clean glass slide as described by Zemjanis.<sup>14</sup> The preparation was immediately examined under a light microscope at 400× magnification using a pre-warmed (37°C) stage. The percentage of motile sperm was calculated by counting both motile and immotile spermatozoa in several microscopic fields, evaluating a minimum of 200 sperm cells.

### Statistical Analysis

The Shapiro–Wilk test was done and it showed that the parameters were Gaussian. Data were expressed as mean  $\pm$  SD and analysed using One-Way ANOVA, followed by the post-hoc Tukey's test to evaluate significant differences within the groups. GraphPad Prism version 9.0 (GraphPad Software, San Diego, California, USA) was used for the statistics analysis. Value of  $P \leq .05$  was considered significant.

## RESULTS

### Body Weight Changes in Response to MSG Treatment Over a Four-Week Period

The initial body weights showed no significant differences ( $P > .05$ ) between all groups. In week one and two, the MSG-only group showed significantly higher body weights (179.57 $\pm$ 12.45g and 182.58 $\pm$ 17.64g respectively,  $P < .05$ ) compared to other groups. However, by weeks three and four, there were no significant differences ( $P > .05$ ) in body weights across all groups, suggesting that the initial weight gain effect of MSG normalized over time (Table 1).

Table 1. Body Weight of the Male Wistar Rats in Different treatment groups across experiment duration

Body Weight (g)	A Control	B MSG only	C Tiger nut only	D MSG & Tiger nut
Initial Weight	147.56 $\pm$ 4.63	147.75 $\pm$ 16.52	155.8 $\pm$ 2.61	146.1 $\pm$ 3.15
Week One	157.83 $\pm$ 9.07 <sup>a</sup>	179.57 $\pm$ 12.45 <sup>b</sup>	160.79 $\pm$ 10.60 <sup>a</sup>	162.16 $\pm$ 5.90 <sup>a</sup>
Week Two	155.78 $\pm$ 11.07 <sup>a</sup>	182.58 $\pm$ 17.64 <sup>b</sup>	161.65 $\pm$ 11.33 <sup>a</sup>	163.76 $\pm$ 9.96 <sup>a</sup>
Week Three	185.48 $\pm$ 18.27	203.93 $\pm$ 17.42	197.17 $\pm$ 7.29	201.92 $\pm$ 11.06
Week Four	203.52 $\pm$ 19.42	214.76 $\pm$ 20.30	204.24 $\pm$ 14.27	205.68 $\pm$ 9.10

MSG: Monosodium glutamate, Values expressed as Means  $\pm$  Standard Deviation (SD), <sup>a,b</sup>Means with different superscripts within row are significantly different ( $P < .05$ ).

### Effects of Tiger Nut and MSG on Sperm Characteristics and Morphology at 14 and 28 Days Post-Treatment

The tiger nut only group showed significantly higher sperm motility ( $88.60 \pm 4.04\%$ ,  $P < .05$ ) compared to other groups. Sperm viability was significantly lower in the MSG-

only group ( $70.00 \pm 4.69\%$ ,  $P < .05$ ). Sperm count was significantly highest in the tiger nut group ( $100.60 \pm 3.21 \times 10^6$  sperm cells/mL,  $P < .05$ ) and lowest in the MSG-only group ( $57.00 \pm 23.82 \times 10^6$  sperm cells/mL,  $P < 0.05$ ) (Table 2).

Table 2. Semen characteristics of the male Wistar rats in different treatment groups at 14 days post-treatment

Parameters	A (Control)	B (MSG only)	C (Tiger nut only)	D (MSG + Tiger nut)
Sperm Motility (%)	$75.00 \pm 12.25^a$	$59.40 \pm 11.15^b$	$88.60 \pm 4.04^c$	$71.40 \pm 8.20^a$
Sperm viability (%)	$81.20 \pm 11.43^{ab}$	$70.00 \pm 4.69^c$	$89.20 \pm 5.26^a$	$72.20 \pm 11.43^{bc}$
Sperm count $\times 10^6$ sperm cells/mL	$83.40 \pm 6.35^a$	$57.00 \pm 23.82^b$	$100.60 \pm 3.21^c$	$80.00 \pm 9.87^a$

MSG: Monosodium glutamate, Values expressed as Means  $\pm$  Standard Deviation (SD), <sup>a,b,c</sup>: Means with different superscripts within row are significantly different ( $P < .05$ ).

After 28 days, the MSG-only group showed significantly reduced sperm motility ( $41.80 \pm 4.92\%$ ,  $P < .05$ ) and viability ( $54.80 \pm 6.76\%$ ,  $P < .05$ ) compared to other groups. The tiger nut group maintained the highest values for these

parameters. Sperm count was significantly lower in the MSG-only group ( $51.80 \pm 10.64 \times 10^6$  sperm cells/mL,  $P < .05$ ) compared to other groups (Table 3).

Table 3. Semen characteristics of the male Wistar rats in different treatment groups at 28 days post-treatment

Parameters	A (Control)	B (MSG only)	C (Tiger nut only)	D (MSG + Tiger nut)
Sperm Motility (%)	$80.80 \pm 7.46^a$	$41.80 \pm 4.92^b$	$92.40 \pm 3.36^a$	$79.40 \pm 7.80^a$
Sperm viability (%)	$78.40 \pm 12.66^a$	$54.80 \pm 6.76^b$	$91.80 \pm 3.83^a$	$82.20 \pm 5.40^a$
Sperm count $\times 10^6$ sperm cells/mL	$78.80 \pm 8.67^a$	$51.80 \pm 10.64^b$	$100.20 \pm 14.8^a$	$87.80 \pm 14.29^a$

MSG: Monosodium glutamate, Values expressed as Means  $\pm$  Standard Deviation (SD), <sup>a,b</sup>: Means with different superscripts within row are significantly different ( $P < 0.05$ ).

The MSG-only group showed significantly higher total abnormal cells ( $13.60 \pm 2.51\%$ ,  $P < .05$ ) and percentage abnormality ( $3.14 \pm 0.38\%$ ,  $P < .05$ ) compared to other groups. Individual morphological abnormalities showed no

significant differences ( $P > .05$ ) between groups. The total cells counted were comparable across all groups ( $P > .05$ ) (Table 4).

Table 4. Sperm morphology of the male Wistar rats in different treatment groups at 14 days post-treatment

Parameters (%)	A (Control)	B (MSG only)	C (Tiger nut only)	D (MSG + Tiger nut)
Abnormal head	$2.00 \pm 0.00$	$2.60 \pm 0.89$	$2.50 \pm 0.58$	$3.00 \pm 0.00$
Coiled tail	$1.67 \pm 0.58$	$3.00 \pm 0.00$	$2.00 \pm 0.00$	$1.00 \pm 0.00$
Bent tail	$2.00 \pm 0.82$	$3.30 \pm 1.26$	$2.50 \pm 0.58$	$2.25 \pm 0.00$
Head without tail	$2.00 \pm 0.00$	$2.33 \pm 0.58$	$2.00 \pm 0.00$	$2.75 \pm 0.96$
Tail without head	$3.33 \pm 0.58$	$2.67 \pm 1.15$	$1.00 \pm 0.00$	$2.00 \pm 1.41$
Looped tail	$2.00 \pm 0.00$	$3.00 \pm 1.00$	$2.00 \pm 0.82$	$2.00 \pm 1.41$
Rudimentary tail	$2.25 \pm 0.50$	$2.33 \pm 1.53$	$0.00 \pm 0.00$	$0.00 \pm 0.00$
Double head	$0.00 \pm 0.00$	$0.00 \pm 0.00$	$0.00 \pm 0.00$	$0.00 \pm 0.00$
Curved mid-piece	$2.00 \pm 0.00$	$2.50 \pm 0.71$	$2.67 \pm 1.52$	$1.00 \pm 0.00$
Total abnormal cells	$8.00 \pm 1.00^a$	$13.60 \pm 2.51^b$	$8.60 \pm 1.82^a$	$9.75 \pm 1.26^a$
Total cells counted	$422.20 \pm 11.43$	$435.20 \pm 38.69$	$463.60 \pm 47.22$	$427.75 \pm 10.69$
Percentage abnormality	$1.90 \pm 0.28^a$	$3.14 \pm 0.38^b$	$1.86 \pm 0.54^a$	$2.28 \pm 0.26^a$

MSG: Monosodium glutamate, Values expressed as Means  $\pm$  Standard Deviation (SD), <sup>a,b</sup>: Means with different superscripts within row are significantly different ( $P < .05$ ).

Table 5 examined sperm morphology across four groups (Control, MSG only, Tiger nut only, and MSG + Tiger nut) over 28 days. The MSG + Tiger nut combination eliminated

coiled tail and tail-without-head abnormalities ( $P < .05$ ), but also showed the lowest total sperm count ( $429.50 \pm 19.09$ ) and highest overall abnormality rate ( $2.20 \pm 0.42\%$ ). The

Tiger nut only group demonstrated the lowest percentage of total abnormalities ( $1.66 \pm 0.56\%$ ), while most other sperm parameters showed no significant differences between groups.

#### Gonadometric Assessment Following MSG and Tiger Nut Treatment at 14 and 28 Days Post-Treatment

No significant differences ( $P > .05$ ) were observed in any gonadometric parameters (testicular weights, lengths,

diameters, and epididymal weights) between groups after 14 days of treatment (Table 6).

Similar to the 14-day results, no significant differences ( $P > .05$ ) were observed in any gonadometric parameters between groups after 28 days of treatment, suggesting that neither MSG nor tiger nut significantly affected testicular size and epididymal weight (Table 7).

Table 5. Sperm morphology of the male Wistar rats in different treatment groups at 28 days post-treatment

Parameters (%)	Control	MSG only	Tiger nut only	MSG + Tiger nut
Abnormal head	$2.75 \pm 0.96$	$2.25 \pm 0.50$	$3.00 \pm 0.00$	$2.00 \pm 0.00$
Coiled tail	$3.00 \pm 0.00^a$	$2.00 \pm 1.00^{ab}$	$2.00 \pm 0.00^{ab}$	$0.00 \pm 0.00^b$
Bent tail	$2.00 \pm 0.00$	$3.00 \pm 0.00$	$2.50 \pm 0.71$	$2.00 \pm 0.00$
Head without tail	$2.00 \pm 0.00$	$1.67 \pm 0.57$	$2.00 \pm 0.00$	$1.50 \pm 0.71$
Tail without head	$2.00 \pm 0.00^a$	$2.33 \pm 1.15^a$	$2.50 \pm 0.71^a$	$0.00 \pm 0.00^b$
Looped tail	$2.33 \pm 0.58$	$2.00 \pm 0.00$	$2.33 \pm 0.58$	$2.00 \pm 0.00$
Rudimentary tail	$2.00 \pm 1.41$	$2.50 \pm 0.71$	$2.50 \pm 0.71$	$3.00 \pm 0.00$
Double head	$0.00 \pm 0.00$	$0.00 \pm 0.00$	$0.00 \pm 0.00$	$0.00 \pm 0.00$
Curved mid-piece	$1.00 \pm 0.00^b$	$2.25 \pm 0.96^a$	$2.00 \pm 1.41^{ab}$	$1.50 \pm 0.71^{ab}$
Total abnormal cells	$9.75 \pm 1.50$	$9.60 \pm 1.14$	$7.80 \pm 2.95$	$9.50 \pm 2.12$
Total cells counted	$510.50 \pm 62.67^a$	$491.40 \pm 27.80^a$	$470.0 \pm 53.61^a$	$429.50 \pm 19.09^b$
Percentage abnormality	$1.93 \pm 0.35^{ab}$	$1.98 \pm 0.16^{ab}$	$1.66 \pm 0.56^b$	$2.20 \pm 0.42^a$

MSG: Monosodium glutamate, Values expressed as Means  $\pm$  Standard Deviation (SD), <sup>a,b</sup>: Means with different superscripts within row are significantly different ( $P < .05$ ).

Table 6. Gonadometric assessment of the male Wistar rats in different treatment groups at 14 days post-treatment

Parameters	A Control	B MSG only	C Tiger nut only	D(MSG $\pm$ Tiger nut)
LTW (g)	$1.01 \pm 0.09$	$1.11 \pm 0.15$	$1.10 \pm 0.06$	$1.11 \pm 0.08$
RTW(g)	$1.06 \pm 0.08$	$1.08 \pm 0.16$	$1.04 \pm 0.10$	$1.11 \pm 0.07$
LTL (mm)	$18.20 \pm 1.22$	$18.44 \pm 0.97$	$18.48 \pm 0.19$	$18.08 \pm 0.41$
RTL (mm)	$18.56 \pm 0.54$	$18.18 \pm 1.06$	$17.98 \pm 0.74$	$17.40 \pm 0.71$
LTD (mm)	$10.34 \pm 0.82$	$10.40 \pm 0.26$	$10.72 \pm 0.69$	$10.42 \pm 0.08$
RTD (mm)	$11.16 \pm 0.88$	$10.38 \pm 0.52$	$10.64 \pm 0.67$	$10.46 \pm 0.53$
Ep. Wt. (g)	$0.35 \pm 0.18$	$0.30 \pm 0.06$	$0.29 \pm 0.07$	$0.39 \pm 0.06$

MSG: Monosodium glutamate, LTW: left testes weight, RTW: right testes weight, LTL: left testes length, RTL: right testes length, LTD: left testes diameter, RTD: right testes diameter, Ep. Wt.: Epididymal Weight, Values expressed as Means  $\pm$  Standard Deviation (SD).

Table 7. Gonadometric assessment of the male Wistar rats in different treatment groups at 28 days post-treatment

Parameters	A Control distilled water for 28 days	B MSG only for 28 days	C Tiger nut only	D (MSG + Tiger nut)
LTW (g)	$1.09 \pm 0.17$	$1.11 \pm 0.14$	$1.17 \pm 0.11$	$1.21 \pm 0.10$
RTW(g)	$1.12 \pm 0.16$	$1.13 \pm 0.15$	$1.21 \pm 0.06$	$1.24 \pm 0.07$
LTL (mm)	$18.12 \pm 1.13$	$18.76 \pm 0.83$	$19.24 \pm 0.84$	$19.76 \pm 0.46$
RTL (mm)	$18.20 \pm 1.10$	$18.56 \pm 0.95$	$19.24 \pm 0.49$	$19.58 \pm 0.82$
LTD (mm)	$10.60 \pm 0.25$	$11.10 \pm 0.72$	$11.28 \pm 0.61$	$11.18 \pm 0.70$
RTD (mm)	$10.72 \pm 0.73$	$11.10 \pm 0.66$	$11.38 \pm 0.30$	$11.42 \pm 0.40$
Ep. Wt. (g)	$0.47 \pm 0.10$	$0.44 \pm 0.09$	$0.41 \pm 0.05$	$0.52 \pm 0.06$

MSG: Monosodium glutamate, LTW: left testes weight, RTW: right testes weight, LTL: left testes length, RTL: right testes length, LTD: left testes diameter, RTD: right testes diameter, Ep. Wt.: Epididymal Weight, Values expressed as Means  $\pm$  Standard Deviation (SD).



## DISCUSSION

Our study reveals significant insights into the protective effects of tiger nut against MSG-induced reproductive dysfunction in male Wistar rats. The findings contribute to both the understanding of MSG's reproductive toxicity and the potential protective role of natural supplements in maintaining reproductive health.

The observed initial increase in body weight following MSG treatment aligns with established literature on MSG's orexigenic effects.<sup>15-17</sup> This finding supports previous research demonstrating MSG's potential role in obesity development through altered feeding patterns and metabolic changes. However, the normalization of body weight by weeks 3-4 suggests potential adaptation mechanisms that warrant further investigation.

A key finding of our study is the marked improvement in sperm parameters following tiger nut administration. The significant enhancement in sperm motility, viability, and count in the tiger nut-treated groups extends beyond previous findings by Ekaluo et al.,<sup>18</sup> who reported only motility improvements. Our comprehensive analysis demonstrates tiger nut's broader positive impact on overall semen quality, suggesting multiple mechanisms of action that could include enhanced antioxidant protection through vitamin C and E content, improved cellular membrane integrity from essential fatty acids, and potential hormonal modulation through bioactive compounds.<sup>19,20</sup>

The protective effect of tiger nut against MSG-induced spermatotoxicity represents a novel finding. While MSG administration alone resulted in significant reductions in sperm motility and count, concurrent tiger nut supplementation maintained these parameters near control levels. This protective effect was particularly evident in sperm morphology, where tiger nut treatment significantly reduced the incidence of abnormal forms typically associated with MSG exposure. These findings build upon previous work by Nosseir et al.<sup>21</sup> and Onakewhor et al.,<sup>22</sup> suggesting tiger nut's potential role in maintaining normal sperm development and maturation.

Regarding gonadometric parameters, the absence of significant changes between groups requires careful interpretation. Rather than indicating a lack of effect, this finding suggests that tiger nut supplementation may help maintain normal testicular architecture even in the presence of MSG exposure.<sup>23-25</sup> This maintenance of normal gonadometric parameters, combined with improved sperm quality, indicates that tiger nut's protective effects may operate through biochemical and cellular mechanisms that

preserve functional capacity without necessarily altering gross anatomical measures.

The practical implications of our findings are significant, particularly for population groups with high MSG exposure through dietary habits, animal breeding programs where reproductive efficiency is crucial, and the development of natural protective supplements for reproductive health. While our study provides valuable insights, we acknowledge several limitations that suggest directions for future research. The 28-day treatment period, while sufficient to demonstrate acute effects, could be extended to cover complete spermatogenic cycles. Inclusion of histological analysis would provide cellular-level insights into protective mechanisms. Hormonal profiling would help elucidate potential endocrine-mediated effects. Dose-response studies could optimize protective effects. These limitations do not diminish the significance of our findings but rather highlight opportunities for more comprehensive future investigations.

As a result, our findings establish a foundation for understanding tiger nut's protective potential against reproductive toxicants and suggest practical applications in both human and animal reproductive health. Tiger nut demonstrates significant promise as a natural protective agent against MSG-induced reproductive dysfunction. The improvement in sperm parameters and maintenance of normal gonadometric measures support its potential use as a supplement in contexts where MSG exposure is concerning. These findings contribute to the growing body of evidence supporting natural dietary interventions in reproductive health maintenance and suggest practical applications in both human and animal reproductive health management.

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**Ethics Committee Approval:** This study was carried out according to the regulations and principles that govern both care and use of experimental animals for research purposes by the Animal Care and Use Ethics Committee at the University of Ibadan (Approval No: UI-ACUREC/17/0069), with strict adherence to guidelines regarding the well-being of the animals, such as proper housing, standard feeding, humane handling and disease prevention and control (Date: 16/5/2018).

**Peer-review:** Externally peer-reviewed.

**Author Contributions:** Concept - OSA, AIA; Design - OSA, AIA; Supervision - OOA, AIA; Resources - OSA; Data Collection and/or Processing - OSA; Analysis and/or

Interpretation - AIA, OOA, OSA; Literature Search - OSA; Writing Manuscript - OSA; Critical Review - AIA, OOA, OSA

**Declaration of Interests:** The authors declare that there is no conflict of interest.

**Funding:** The authors declared that this study has received no financial support

Bu çalışma, Ibadan Üniversitesi Hayvan Bakımı ve Kullanımı Etik Kurulu'nun (Onay No: UI-ACUREC/17/0069, Tarih: 16/05/2018) araştırma amaçlı deneysel hayvanların bakımı ve kullanımına ilişkin düzenlemeler ve ilkeler doğrultusunda gerçekleştirilmiştir.

**Hakem Değerlendirmesi:** Dış bağımsız.

**Yazar Katkıları:** Fikir – OSA, AIA; Tasarım – OSA, AIA; Denetleme – OOA, AIA; Kaynaklar - OSA; Veri Toplanması ve/veya İşlemesi - OSA; Analiz ve/veya Yorum – AIA, OOA, OSA; Literatür Taraması - OSA; Yazıyı Yazan - OSA; Eleştirel İnceleme – AIA, OOA, OSA

**Çıkar Çatışması:** Yazarlar çıkar çatışması olmadığını beyan ederler.

**Finansal Destek:** Yazarlar bu çalışma için herhangi bir mali destek almadıklarını beyan etmişlerdir.

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