



DOI: <https://doi.org/10.25130/tjas.25.1.15>

## Physiological effects of *Boswellia serrata* resin extract on reproductive and metabolic hormones in adult male rats

Entissar Mansour Abdul Rasool<sup>1\*</sup> , and Man Samir Balola<sup>2</sup> 

<sup>1</sup>Department of Basic science, College of Dentistry, Al-Iraqia University, Baghdad, Iraq

<sup>2</sup>Department of Physiology, Biochemistry and pharmacology, College of Veterinary Medicine, University of Mosul, Mosul, Iraq

\*Correspondence email: [entissar.m.abdulrasool@aliraqia.edu.iq](mailto:entissar.m.abdulrasool@aliraqia.edu.iq)

### KEY WORDS:

Boswellia, spermatogenesis, testosterone, oxidative stress, lipid profile

Received: 14/12/2024

Revision: 22/02/2025

Proofreading: 22/03/2025

Accepted: 16/03/2025

Available online: 31/03/2025

© 2025. This is an open access article under the CC by licenses <http://creativecommons.org/licenses/by/4.0>



### ABSTRACT

*Boswellia serrata* (*B.serrata*) is commonly defined as a resin, widely used because of the healing properties of inflammations and other diseases due to its powerful antioxidant action. This study investigates the effect of *B.serrata* on reproductive system and metabolic hormones in adult male rats. Four groups of adult male rats (10 rats each) were subjected to the trial. Group 1 (control) rats were fed on normal diet (ND), group (2) rats were fed on high fat diet (HFD), group (3) rats were fed ND and aqueous *B.serrata* extract (1000 mg/kg body weight/day), and group (4) rats fed on HFD and aqueous *B.serrata* extract at the same dose respectively for the duration of 60 days. The results indicated that feeding on HFD resulted in hyperlipidemia, significant ( $p \leq 0.05$ ) reduced level of testosterone, insulin, TSH and the antioxidant enzymes (GSH, SOD, CAT), significant ( $p \leq 0.05$ ) elevated levels of lipid profile, oxidative markers (MDA) and cytokines (TNF- $\alpha$ , IL-6 and IL-1 $\beta$ ) concentration. Negative effects on spermatogenesis was also observed, confirmed by significant ( $p \leq 0.05$ ) decrease in sperm concentration and sperm motility in addition to significant ( $p \leq 0.05$ ) increase in the percentage of dead and abnormal sperm. Daily aqueous *B.serrata* extract administration for 60 days managed to adjust all the metabolic disorders and enhanced rats fertility. The study concluded that the administration of *B.serrata* extract improves the adverse effects of high fat diet by acting as antidiabetic, anti-inflammatory and antioxidant agent. *B. serrata* extract is also able to correct the disrupted metabolic pathways, with positive effect on male fertility.

## التأثيرات الفسلجية لمستخلص لبان الذكر على الجهاز التكاثري والهرمونات الايضية في ذكور الجرذان البالغة

<sup>1</sup>انتصار منصور عبد الرسول و<sup>2</sup>معن سمير بلوله

<sup>1</sup>فرع العلوم الاساسية، كلية طب الاسنان، الجامعة العراقية، بغداد، العراق

<sup>2</sup>فرع الفسلجة الكيمياء والحياتية والادوية، كلية الطب البيطري، جامعة الموصل، الموصل، العراق

### الخلاصة

يُعرّف لبان الذكر بأنه مادة راتنجية تُستخدم على نطاق واسع في علاج الحالات الالتهابية وامراض اخرى بسبب خصائصه العلاجية كمضاد للالتهابات ومضاد قوي للاكسدة. صممت هذه الدراسة لمعرفة التأثيرات الفسلجية للمستخلص المائي للبان الذكر على الجهاز التناسلي والهرمونات الايضية في ذكور الجرذان البالغة. خضعت أربع مجاميع من ذكور الجرذان البالغة (10 جرذان لكل مجموعة) للتجربة. المجموعة الاولى (السيطرة) تم تغذيتها على علف طبيعي، المجموعة الثانية تمت تغذيتها على علف عالي الدهون، المجموعة الثالثة تمت تغذيتها على علف طبيعي فضلا عن تجريعها بمستخلص لبان الذكر 1000 ملغم/كغم يوميا لمدة 60 يوما، المجموعة الرابعة تمت تغذيتها على علف غني بالدهون فضلا عن تجريعها بمستخلص لبان الذكر 1000 ملغم/كغم يوميا لمدة 60 يوما. اظهرت النتائج إلى أن المعاملة بالعلف عالي الدهون أدت إلى حدوث فرط الدهون في الدم رافقه انخفاضاً معنوياً ( $p \leq 0.05$ ) في مستوى هرمون التستوستيرون، هرمون الأنسولين وهرمون المحفز للغدة الدرقية، فضلا عن انخفاض معنوي ( $p \leq 0.05$ ) في مستوى انزيمات مضادات للاكسدة (الكلوتاثيون، سوبر اوكسايدي ديسميوتيز وانزيم الكاتاليز) مصحوبا بارتفاع معنوي ( $p \leq 0.05$ ) في مستوى المألوندايديهايد والساييتوكينات مثل (انترلوكين 6، انترلوكين 1-بيتا، عامل نخر الورم الفا) في الدم. كما لوحظ تأثير مثبت للخصوبة تمثل في انخفاض معنوي ( $p \leq 0.05$ ) في عدد الحيامن ونسبة الحركة ونسبة الحيامن الحية مع ارتفاع معنوي ( $p \leq 0.05$ ) في نسبة الحيامن الميتة والمشوه. ادت المعاملة بالمستخلص المائي للبان الذكر إلى تحسين الخلل المحدث أعلاه على مستويات الخصوبة، الابيض ومعايير الإجهاد التأكسدي. يستنتج من الدراسة الحالية أن للبان الذكر ومستخلصاته تأثيرات إيجابية على عدة مستويات في الجسم منها معايير الخصوبة لدى الذكور فضلا عن تأثيره المضاد للاكسدة والمضاد للالتهاب والخافض لدهون الدم.

**الكلمات الافتتاحية:** لبان الذكر، عملية تكوين الحيامن، هرمون التستوستيرون، الاجهاد التأكسدي، صور الدهون.

## INTRODUCTION

Male infertility has recently been identified as a one of the serious challenges for mankind, and about 50% of infertilities have a male component (WHO,2021).The current lifestyle is considered as a critical reason for the decreased fertility in men, which manifested by a sharp decrease in sperm count, poor sexual performances and loss of libido (Emokpae and Brown,2021) .One of the features of the current lifestyle is the high consumption of foods rich in energy, high in fat and sugar, which predominantly leads to the development of obesity (Shen *et al.*,2016 ).Numerous studies link hormonal disruption, in addition to increased tissue oxidative and inflammatory damage, to the impact of a high-fat diet on male reproduction (Barbagallo *et al.*,2021; Leisegang *et al.*,2021).Furthermore, spermatogenesis can be reduced by consuming high fat diet through affecting sperm maturation in the epididymis and hormonal disruption(Ruiz-Valderrama *et al.*, 2025). In recent years, Plants and/or their extracts have long been recognized to have a powerful influence on a variety of illnesses (Akram and Mahmood, 2024). The wide use of plant products is probably attributed to their minor toxic effect compared to the chemical medicines, in addition to the reduced tissue damage, resulting from plants, thus treatment can be performed with less side effects (Al-Yasiry and Kiczorowska, 2016).

One of the species have been identified and utilized in conventional medicine is *Boswellia* genus, also known as frankincense (Schmiech *et al.*,2019). *B. serrata* is a large tree of mountainous areas; the resin of this plant was recognized to have a variety of therapeutic actions on the body, such as anti-inflammatory, antioxidant and antibacterial potential (Purabiya *et al.*,2023). Numerous desired curative properties of *B. serrata* have also been illustrated, including; hypoglycemic effect (Karimi *et al.*, 2024), promoting healing of peptic ulcer (Sherif *et al.*, 2019), anticancer (Trivedi *et al.*,2023) and treating of osteoarthritis and improving learning problems (Beheshti *et al.*, 2018) ,furthermore to rheumatoid arthritis(Kumar *et al.*,2019) .

Due to the composition of the resin collected from frankincense, recently, it has been noted that *B. serrata* has effects on fertility. Different extracts have been studied for their effects on infertility in both human and animal models. Due to the extract's strong antioxidant properties and steroid-like properties, it has been reported that aqueous and/or alcoholic extracts of *B. serrata* may cure or improve male infertility in animals, which predominantly increase the number of active sperms (Alyahya and Asad, 2020; Emtenan *et al.*, 2023). Another study explained the prophylactic effect of *B. sacra* extract in the testis is mediated by its antioxidant action, in addition to increasing the number of spermatocytes in the seminiferous tubules, along with raising blood testosterone levels, it also improved sperm motility and count (Alharbi *et al.*, 2022)

In this study, we used *B. serrata* extract to investigate their effects on male fertility and metabolic aspects in rats exposed to a high fat diet, which is supposed to be a deleterious factor, to disturb cell oxidative status.

## MATERIALS AND METHODS

### Ethical approval.

Approval for the study protocol was issued by Al\_Iraqia University- College of Dentistry (No. ESA&HER-06 Date:22/06/2024). All lab animal experiments were undertaken according to the guide issued by the National Institutes of Health.

### Animals

*Albino* adult male rats ( $90 \pm 10$  days), with body weight range ( $210 \pm 20$  g) were subjected to the experiment. Animals were obtained from the animal house at AL-Nahrain University/ Biotechnology Research Center. Rats were kept in a healthy and normal environment, including  $22^\circ\text{C} \pm 2^\circ\text{C}$ , 55-60% humidity and a 12 h light/dark cycle. Rats were exposed to a –one week- duration of adaptation before being divided into experimental groups.

### Diet

The diet used in this research was procured from local markets in accordance with the nutritional needs of the rats. High-fat diet (Panchal& Brown, 2010), was formulated by adding saturated fats to the ingredients to meet the percentage of 60% of total energy in the diet, gained from fats.

### Preparation of *B. serrata* aqueous extract

*B. serrata* resins used in this investigation were procured from local marketplaces in Baghdad, Iraq. 10 g of *B. serrata* resin was infused in 10 ml of distilled water overnight at  $4^\circ\text{C}$ , then filtered using a clean cloth. The percentage of the dry material was determined using the lyophilizing technique, and the stock concentration obtained during this protocol was diluted to the concentration of 300 mg/ml, provided daily to the rats orally at a dosage of 1000 mg/kg of body weight.

### Experimental design

Forty adult male rats were split into four groups at random, with ten rats in each group

**Group 1:** Negative control: Normal rats administered 1 ml distilled water orally /day by gavage needle and fed normal diet *ad libitum* for 60 days.

**Group 2:** High-fat diet (HFD) group rats fed on high fat diet (60% of the total energy) *ad libitum* for 60 days.

**Group 3:** rats fed normal diet with *B. serrata* aqueous extract (1000 mg/kg/day) orally by gavage needle for 60 days

**Group 4:** rats fed on high-fat diet (HFD) with *B. serrata* aqueous extract (1000mg/kg/day) orally by gavage needle for 60 days

### Specimen's collection

At the end of the 60 days of treatments, fasting was applied to the rats for 12 hours. Blood was collected from the retro-orbital puncture, under deep anesthesia with diethyl ether, using capillary tubes (without heparin). After allowing the blood to coagulate, it was centrifuged for 15 minutes at 3000 rpm. Before doing the biochemical analysis, the serum was moved into Eppendorf tubes and kept at  $-20^\circ\text{C}$ .

### Epididymis Preparation for Sperm parameters

After blood collection, rats were sacrificed, postmortem (PM) was undertaken to obtain the genitalia for the spermatogenesis protocol. The prostate gland, seminal vesicles, and epididymis were removed, cleaned, and weighed together with the testis. The head of epididymis was removed, sectioned, and its contents were promptly gently pressed

in a sterile watch glass that held 9.8 milliliters of buffer formalin and 0.1 milliliters of 5% eosin stain. Using the hemocytometric approach, this was utilized to count the sperm (Yokoi& Mayi,2004). Eosin-nigrosin stain mixed with 3% sodium citrate was used to measure the proportion of living and morphologically aberrant sperm in a smear made from epididymal tail content (Sakamoto& Hashimoto,1986).

### Biochemical analyses

The serum lipid profile levels, including total cholesterol (TC), triglycerides (TG), High density lipoproteins (HDL) and low density lipoproteins (LDL) were carried out photometrically using spectrophotometer, very low density lipoproteins (VLDL) was mathematically derived by dividing the TG value by 5.

Serum hormone levels of insulin ,Triiodothyronine (T3), Thyroxine (T4),Thyroid-stimulating hormone (TSH) and Testosterone (T), as well as cytokines concentration including Tumor Necrosis Factor-  $\alpha$  (TNF- $\alpha$ ), Interleukin-6 (IL-6) Interleukin-1 $\beta$  (IL-1 $\beta$ ) and oxidative stress markers in serum such as malondialdehyde (MDA), glutathione (GSH), superoxide dismutase (SOD) and catalase (CAT) were carried out using ELISA technique.

All photometric methods applied on spectrophotometer used reagents supplied from (Biolabo, France), measured using a spectrophotometer (Shimadzu,Japan), whereas ELISA techniques were undertaken using kit reagents supplied by (SunLong Biotech Co. LTD, China) and read by (HumaReader HS, Germany) reader ELISA.

### Statistical Analysis

To identify significant values, all data were submitted to a one-way analysis of variance test (ANOVA), Using the Duncan multiple range test, groups were compared. Differences across groups were considered significant when ( $P \leq 0.05$ ). The IBM SPSS Statistics, version 23, was used to conduct the investigation.

## RESULTS AND DISCUSSION

The focus of the current study was on the influence of *B. serrata* aqueous extract on different metabolic aspects in adult male rats, exposed to high fat diet (HFD). In addition, the antioxidant capacity was also assessed. Our experiments were undertaken in normal rats and rats fed on HFD as a challenge to investigate the efficacy of the extract in abnormal metabolic status. All results of the study are described below in detail. Table 1 indicates that the HFD therapy was a noteworthy ( $p \leq 0.05$ ) influence in the rise in body weight (B.W.) above the control group mean at the conclusion of the trial. With or without HFD, treatment with *B. serrata* aqueous extract effectively reversed this action.

Table( 2) describes the weight of genitalia of the experimental animals. No significant effect was detected in the weight of such testis, epididymis, seminal vesicle and prostate gland as a result of the treatment with HFD and/or *B. serrata* aqueous extract. All the values were around the statistical level of the normal control group.

Table1: Impact of *B. serrata* aqueous extract on B.W. of adult male rats exposed to HFD

Treatments	Parameters			
	Initial body weight(g)		Final body weight(g)	
Control(ND)	208.3 $\pm$ 10.36	a	240.4 $\pm$ 11.58	b
HFD	229.2 $\pm$ 15.95	a	275.4 $\pm$ 8.82	a
<i>B. serrata</i>	218.2 $\pm$ 9.7	a	245.2 $\pm$ 6.72	b
HFD + <i>B. serrata</i>	225. 8 $\pm$ 11.9	a	258.8 $\pm$ 9.73	ab

The values are shown as mean  $\pm$ SEM; Statistical differences are shown by different letters in the same column ( $p \leq 0.05$ )

Table 2: Impact of *B. serrata* aqueous extract on weight of genital organs in male rats exposed to HFD.

Treatments	Parameters			
	Testis weight (mg/100g B.W.)	Epididymis weight (mg/100gB.W)	Seminal vesicle (mg /100g B.W.)	Prostate gland (mg/100g B.W.)
Control (ND)	540±21 a	160±23 a	110.11±18 a	445.16±13 a
HFD	553±53 a	164±12 a	92.90±22 a	460.67±22 a
<i>B. serrata</i>	534±34 a	155±30 a	106.45±30 a	439.23±19 a
<i>HFD+B.serrata</i>	541±47 a	148±21 a	89.72±11 a	450.88±31 a

The values are shown as mean ±SEM; Statistical differences are shown by different letters in the same column (p≤0.05)

The current study is a complementary step to previous studies by other researchers, to investigate the different actions of *B. serrata* on a variety of metabolic parameters to speculate the prospective mechanism of action as well as to confirm the findings by others and highlight the advantages behind using *B. serrata* in alternative medicine. In this context, we analyzed a broad range of parameters, representing different metabolic processes for the effect of the extract against the high fat diet (HFD), administered to male rats as an abnormal metabolic condition. As described in the results section, the overall body weight of rats was increased by feeding on HFD, which is an inevitable result due to the increase in the energy intake and body fat mass, which is a result similar to the search result arch (Mahdi & Khalil, 2022 ; Ayed *et al.*,2024) . This is consistent with (Alharbi *et al.*,2022) ,who also found no alteration in the weight of testis and accessory glands, which has been confirmed by our study. This effect is potentially attributed to the insufficient duration of treatment to affect the physiological function of the reproductive system (tables 1 and 2).

Data presented in Table (3) refer to the numbers of sperms as well as the sperms motility observed from the experimental animals. Following HFD therapy, both number and sperm activity were considerably (p≤0.05) decreased; however, following *B. serrata* extract treatment, the effect was noteworthy (p≤0.05) restored to its normal value. The sperms characters described in Table (4) showed that the treatment with HFD significantly (p≤0.05) reduced the percentage of the live sperms to the least level amongst the four groups. However, the intubation of *B. serrata* extract to the HFD- treated rats managed to modify the above effect, hence elevation of the level over the HFD group was obtained. The same adverse effect was observed regarding the percentage of the dead sperms, which was also elevated in the HFD group and amended in the HFD plus *B. serrata* extract group. In contrast to the live and dead sperms, the only elevation (p≤0.05) in the percentage of abnormal sperms was detected in the HFD group.

Table3:Impact of *B.serrata* aqueous extract on sperm count and motility in rats exposed to HFD

Treatments	Parameters	
	(No. sperms×10 <sup>6</sup> ) of head epididymis	Sperms motility%
Control(ND)	1.62± 0.05 a	87.80±0.12 a
HFD	0.905±0.1 b	60.65±2.4 b
<i>B. serrata</i>	1.79±0.07 a	94.98±1.8 a
<i>HFD + B. serrata</i>	1.54±0.23 a	84.6±0.9 a

The values are shown as mean ±SEM; Statistical differences are shown by different letters in the same column (p≤0.05)

Table 4: Impact of *B. serrata* aqueous extract on sperm characters of rats exposed to HFD

Treatments	Parameters		
	Live sperms %	Dead sperms %	Abnormal sperms%
Control(ND)	81.4±0.8 a	18.40±0.8 c	8.39 ± 2.41 b
HFD	64.55 ±1.5 c	35.45±1.5 a	20.10 ±1.90 a
<i>B. serrata</i>	83.81 ± 2.4 a	16.19±2.4 c	7.24 ± 1.01 b
<i>HFD + B. serrata</i>	76.28± 3.1 b	23.72± 3.01 b	12.53 ± 3.6 b

The values are shown as mean ±SEM; Statistical differences are shown by different letters in the same column (p≤0.05)

A significant improvement in terms of sperms concentration, motility and live and dead sperms, have also been observed by the treatment with *B. serrata* after being negatively affected by the feeding on HFD (tables 3 and 4). These results confirm the findings of (Emtenan *et al.*, 2023), and it is likely to be a result of the antioxidant properties of *B. serrata* ingredients. Our results are also in agreements with Bataineh and Nusier (2005), who attributed the effect of the HFD on reproductive characters to reduced sperms intensity and changes in the epithelium of the epididymis, the effect which has been adjusted by the extract. It has also been found that the concentrations of TG and VLDL negatively affect sperm count, live sperm through affecting Leydig cell's function, whereas increased HDL concentration can reverse those effects and improve the testicular function (Louei Monfared, 2013) . This action requires more studies to identify the underlying mechanisms of the effect. It has been reported that the HFD is a potential factor inducing oxidative stress status, the condition entails a decrease in the body's natural antioxidant capacity (Al-Samarai& Taha, 2024) as well as the uncontrolled generation of reactive oxygen species (ROS). This condition is an additional factor to affect the sperm function and enhance apoptosis, since sperms have a high content of polyunsaturated fatty acids, which are a substrate favors lipid peroxidation (Wagner *et al.*,2019). Furthermore, the adjustment of the testosterone concentration after the treatment with *B. serrata* is an additional factor to be considered for the improvement of the spermatogenesis process (table 6). The testosterone adjustment is probably due to the inhibition of aromatase activity, already activated as a result of the HFD (Phillips & Tanphaichitr, 2010). One more assumption for the testosterone elevation by *B. serrata* extract is linked to the reduced total cholesterol (TC) (table 5), suggesting the utilization of cholesterol as the precursor for the testosterone biosynthesis. This action is a rational reason for decreased total cholesterol and LDL concentrations yielded from the treatment by the *B. serrata* extract.

The results of lipid profile presented in Table (5) revealed that the treatment with HFD resulted in a significant ( $p \leq 0.05$ ) elevation in triglycerides concentration, and the consequent VLDL. Whereas the group received both *B. serrata* extract with the HFD demonstrated a noteworthy ( $p \leq 0.05$ ) reduction in contrast with the HFD group, however, it is higher than the control and *B. serrata* groups. Both TC and LDL followed a parallel line when the only elevation observed was in the HFD group, with no changes in the other treatments. However, as compared to the other three groups, the HDL concentration only dropped in the HFD group.

**Table5: Impact of *B. serrata* aqueous extract on lipid profiles of male rats exposed to HFD**

Treatments	Parameters				
	TG (mg/dl)	VLDL(mg/dl)	LDL(mg/dl)	HDL(mg/dl)	TC(mg/dl)
Control (ND)	60.48±2.6 c	12.09±0.52 c	38.06±1.34 b	47.48±1.47a	97.28±1.9 b
HFD	140.83±10.87 a	28.16±2.17 a	93.54±14.5 a	31.41±5.5 b	153.12±18.39 a
<i>B. serrata</i>	60.63±1.3 c	12.12±0.16 c	33.14±1.32 b	51.21±2.23a	96.48±2.81 b
<i>HFD+B.serrata</i>	103.20±3.96 b	20.63±0.79 b	37.92±2.99 b	49.98±2.49a	108.54±5.21 b

The values are shown as mean ±SEM; Statistical differences are shown by different letters in the same column ( $p \leq 0.05$ )

It is not surprising that feeding rats on HFD elevates the concentration of TG and the mathematically- derived VLDL. The HFD is rich in TG and free fatty acids, which are absorbed from the intestinal mucosa by chylomicrons to turn to intermediate density lipoprotein (IDL) and consequently VLDL and LDL in the blood. However, HDL concentration possesses an inverse relationship with LDL, since HDL function is to transport cholesterol from the adipose tissue to organs to be utilized as metabolites. The above data agree with Ahmed *et al.* (2015), who undertook an in vitro study and found an antidyslipidemic effect of similar extracts. Furthermore, similar findings have also been described by Gomaa *et al.*( 2019). To support our data in regards of lipid profile, we estimated the insulin concentration (table 6), which indicated an improvement due to the administration of *B. serrata*. This improvement might be because of the direct action of *B. serrata* extract on pancreatic B cells. Insulin is an anabolic hormone which can restore HFD- induced hypercholesterolemia, thereby, a proper utilization of energy is expected (Shirwaikar *et al.*,2004)

The analysis of hormones revealed different effects depending on the hormonal function and the target of our treatment. The treatment with *B. serrata* extract induced an elevation ( $p \leq 0.05$ ) in testosterone hormone over the value of the control. The later action was followed by the administration of the extract with HFD, which is similar to the

control. The treatment with HFD caused a sharp decline ( $p \leq 0.05$ ) in the testosterone level beyond all treatments. The treatment with *B. serrata* extract and /or HFD did not affect the concentration of both T3 and T4 hormones, while feeding rats on HFD increased the TSH concentration over the other three groups of the experiment. Finally, the HFD group showed a decrease in the insulin level, which was then adjusted after the treatment with *B. serrata extract* (table 6).

Table 6: Impact of *B. serrata* aqueous extract on metabolic hormones of male rats exposed to HFD

Parameters	Treatments			
	Control (ND)	HFD	<i>B. serrata</i>	HFD+ <i>B. serrata</i>
T (ng/ml)	7.78±0.31 b	5.18±0.4 c	9.24±0.28 a	6.88±0.43 b
T4(pg/ml)	5.9±0.5 a	6.3± 0.1 a	5.4±0.9 a	6.02±0.23 a
T3(pg/ml)	0.95±0.01 a	0.82±0.08 a	1.02±0.1 a	0.86±0.05 a
TSH( $\mu$ U/ml)	0.9±0.5 b	1.74 ±0.21 a	0.7±0.12 b	1.02±0.23 b
Insulin (ng/ml)	1.70±0.27 a	1.01±0.15 b	1.58±0.17 a	1.32±0.4 ab

The values are shown as mean  $\pm$ SEM; Statistical differences are shown by different letters in the same column ( $p \leq 0.05$ )

Further to the data presented above, we also analyzed relevant cytokines to confirm the efficacy of the *B. serrata* extract in correcting prospective disorders, undertaken by the administration of HFD. We found that such TNF- $\alpha$ , IL-1 $\beta$  and IL-6 concentrations were significant ( $p \leq 0.05$ ) elevated over the control after feeding rats on HFD, which was significant ( $p \leq 0.05$ ) reduced by the effect of *B. serrata* extract in group 4. In the same time, the oral intubation of normal rats with *B. serrata* yielded no discernible difference as compared to the control group (table 7).

Data presented in Table 8 confirm the oxidative stress markers of the *B.serrata* extract, which adjusted the HFD-induced elevated MDA, in comparison to the control group as a measure of oxidative damage. On the other hand, the antioxidant markers (GSH, SOD and CAT) estimated in the serum of experimental rats, showed a correction for the significant ( $p \leq 0.05$ ) reduced value of the HFD group taking it to the same significant ( $p \leq 0.05$ ) as a consequence of the extract treatment at the control level.

Table7: Impact of *B. serrata* aqueous extract on cytokines concentration of male rats exposed to HFD

Parameters	Treatments			
	Control (ND)	HFD	<i>B. serrata</i>	HFD+ <i>B. serrata</i>
TNF- $\alpha$ (pg/ml)	10.46±0.45 c	18.30±1.2 a	9.98±0.5 c	14.18±1.02 b
IL-1 $\beta$ (pg/ml)	42.52±4.3 c	69.72±1.2 a	41.35±1.18 c	56.12±3.2 b
IL-6(pg/ml)	59.38±1.94 c	84.96±2.73 a	58.86±3.6 c	72.00±2.02 b

The values are shown as mean  $\pm$ SEM; Statistical differences are shown by different letters in the same column ( $p \leq 0.05$ )

Table 8: Impact of *B.serrata* aqueous extract on Oxidative Stress markers in serum rats exposed to HFD

Treatments	Parameters			
	MDA(nmol/l)	GSH( $\mu$ mol/l)	SOD (U/ml)	CAT(U/ml)
Control (ND)	7.50±0.33 c	19.23±0.54 a	90.24±2.21 a	29.02±0.74 a
HFD	18.68±0.71 a	12.79±0.91 b	75.20±3.61 b	17.12±1.12 b
<i>B. serrata</i>	7.51±0.53 c	20.44±0.34 a	94.56±1.88 a	31.44±1.44 a
HFD + <i>B. serrata</i>	13.98±0.49 b	17.65±0.40 a	92.94±3.83 a	27.86±1.38 a

The values are shown as mean  $\pm$ SEM; Statistical differences are shown by different letters in the same column ( $p \leq 0.05$ )

The improvement of the oxidative stress status observed via the treatment with *B. serrata* extract integrates with our findings discussed above. It has been reported that feeding lab animals on HFD is a reason for induction of oxidative stress (Lasker *et al.*, 2019; Mani *et al.*, 2022) However, the ingredients contained in the extract, mainly

Boswellic acid efficiently improved the oxidative status by reducing MDA and elevation such GSH, SOD and CAT, which is in accordance with (Khan *et al.*,2022).It has been demonstrated that the administration of *B. serrata* extract inhibits the expression of genes responsible for the antioxidant enzymes synthesis, consequently, decreases the ROS pool in the cell (Alyahya & Asad, 2020). Moreover, scavenging of the endogenous ROS is also expected by the extract due to the potent antioxidant activity. The current study suggests the antioxidant activity carried out by ingredients contained in *B. serrata*, namely Boswellic acid, to be the key factor, exhibiting an improvement in the different metabolic functional status analyzed in our results.

## CONCLUSION

The data analysis from this investigation lends credence to the potential therapeutic use of *B. serrata* aqueous extract, induced by the administration of HFD on levels of reproductive function, lipid profile and oxidative damage. However, more investigations in this context are recommended for a deeper insight into the individual ingredients and their mechanisms of actions.Worthy mentioned that the extracts affected neither the cytokines and the thyroid hormones in this study. In addition, we found that the *B. serrata* aqueous extract shows the maximum effect during stress conditions, with almost no effect on normal rats.

## CONFLICT OF INTEREST

The authors declare no conflicts of interest associated with this manuscript.

## ACKNOWLEDGMENTS

Al-Iraqia University / College of Dentistry is acknowledged by the author for providing facilities for this research.

## REFERENCES

- Ahmed, H. H., Abd-Rabou, A. A., Hassan, A. Z. and Kotob. S. E. (2015). 'Phytochemical Analysis and Anti-cancer Investigation of Boswellia serrata Bioactive Constituents In Vitro', Asian Pac J Cancer Prev, 16: 7179-88.
- Akram,M. and Mahmood,K.(2024). Awareness and current knowledge of medicinal plants. RPS Pharmacy and Pharmacology Reports.3, rqa023<https://doi.org/10.1093/rpspp/rqa023>
- Alharbi, S.A.,Asad, M., Abdelsalam, K.E.A., Ibrahim, M.A., Chandy, S.(2022). Beneficial Effect of Methanolic Extract of Frankincense (Boswellia Sacra) on Testis Mediated through Suppression of Oxidative Stress and Apoptosis. Molecules , 27: 4699.
- Al-Samarai, A.M.& Taha, A.T.(2024). The Protective Effects of L-Carnitine Against Oxidative Toxicity in Adult Male New Zealand Rabbits. Tikrit Journal for Agricultural Sciences. 24 (3):1-12
- Alyahya, A.A., Asad, M. (2020).Repeated 28-DAY oral dose study on Boswellia sacra oleo gum resin extract for testicular toxicity in rats. J. Ethnopharmacol. 258, 112890.
- Al-Yasiry,A.and Kiczorowska,B.(2016). Frankincense-therapeutic properties. Adv. Hyg. Exp. Med. 70: 380–391.
- Ayed, H.S, Shihab,O.B.& Jassim, M.A. (2024). Level of lipid profile and liver enzymes hypercholesterolemic rats treated with actimel milk.Tikrit Journal for Agricultural Sciences.24(4):235-245
- Barbagallo, F., Condorelli, R.A. , Mongioì , L.M., Cannarella, R. , Cimino, L. , Magagnini, M.C. , Crafa, A., Vignera, S. , Calogero, A.E. (2021) Molecular mechanisms underlying the relationship between obesity and male infertility. Metabolites:11,840
- Bataineh , H. N. and Nusier, M. K.(2005). Effect of cholesterol diet on reproductive function in male albino rats. Saudi Med J;26(3):398-404.
- Beheshti, S., Ghorbanpour Skakakomi, A., Ghaedi, K., Dehestani, H. (2018). Frankincense upregulates the hippocampal calcium/calmodulin kinase II- $\alpha$  during development of the rat brain and improves memory performance. International journal of developmental neuroscience. 69(1):44-48.



- Emokpae, M.P. and Brown, S.I. (2021) Effects of lifestyle factors on fertility: practical recommendations for modification. *Reproduction and Fertility* : 1 R13–R26
- Ementan, M., Heba, H., Mohamed, T., Reda, M., Korany, S., Walid, S. El Natat , Manal, M., Seham, S., Al Shafei, M. and Dina, M. (2023). Effect of Frankincense (*Boswellia Carterii*) on Animal Reproductive Performance Egypt. *J. Chem.* 66( 2): 213- 223.
- Gomaa, A. A., Farghaly, H. S. M., El-Sers, D. A. ,Farrag, M. M. and Al-Zokeim, N. I. (2019). 'Inhibition of adiposity and related metabolic disturbances by polyphenol-rich extract of *Boswellia serrata* gum through alteration of adipo/cytokine profiles', *Inflammopharmacology.* 27: 549-59.
- Karimi, M., Vakili, K., Rashidian, P., Razavi-Amoli, S.K., Akhbari, M. and Kazemi, K. (2024) Effect of boswellia (*Boswellia serrata* L.) supplementation on glycemic markers and lipid profile in type 2 diabetic patients: a systematic review and meta-analysis. *Front. Clin. Diabetes Healthc.* 5:1466408. doi: 10.3389/fcdhc.2024.1466408
- Khan, A., Khan, I., Halim, S. A. , Rehman, N. U. , Karim, N., Ahmad, W. , Khan, M., Csuk, R. and Al-Harrasi, A. (2022). 'Anti-diabetic potential of  $\beta$ -boswellic acid and 11-keto- $\beta$ -boswellic acid: Mechanistic insights from computational and biochemical approaches', *Biomed Pharmacother.* 147: 112669.
- Kumar, R., Singh, S., Saksena, A.K., Pal, R., Jaiswal, R. and Kumar, R. (2019) Effect of *Boswellia serrata* extract on acute inflammatory parameters and tumor necrosis factor- $\alpha$  in complete Freund's adjuvant-induced animal model of rheumatoid arthritis. *Int J App Basic Med Res* 9:100-6.
- Lasker, S., Rahman, M. M. , Parvez, F. , Zamila, M. , Miah, P. , Nahar, K. , Kabir, F. , Sharmin, S. B. , Subhan, N. , Ahsan, G. U. and Alam. M. A. ( 2019). 'High-fat diet-induced metabolic syndrome and oxidative stress in obese rats are ameliorated by yogurt supplementation', *Sci Rep*, 9: 20026.
- Leisegang, K., Sengupta, P., Agarwal, A. & Henkel, R. (2021). Obesity and male infertility: mechanisms and management. *Andrologia.* 53(1):e13617.
- Louei Monfared A. (2013). Correlation of serum lipid profile with histological and seminal parameters of testis in the goat. *Int J Fertil Sterility.* 7 (2):122–129.
- Mahdi, M.H. & Khalil, Y.I. (2022). The effect of bacillus licheniformis on weight gain, blood picture and lipid profiles in rats feed high cholesterol diet. *Tikrit Journal for Agricultural sciences.* 22(2):36-43
- Mani, A., Kushwaha, K., Khurana, N. and Gupta, J. (2022). 'p-Coumaric acid attenuates high-fat diet-induced oxidative stress and nephropathy in diabetic rats', *J Anim Physiol Anim Nutr (Berl)*, 106: 872-80.
- Panchal, S. K., & Brown, L. (2010). Rodent models for metabolic syndrome research. *Journal of Biomedicine and Biotechnology*, 1-14
- Phillips K., Tanphaichitr N. (2010). Mechanisms of obesity-induced male infertility. *Expert Rev Endocrinol Metab.* 5(2): 229-251
- Purabiya, P., Kaushik, S., Satapathy, A., Jain, P. and Satapathy, T. (2023). Pre-Clinical Evaluation for Anti-Inflammatory, Antioxidant and Antibacterial Potential of *Boswellia serrata*. *Adv in Phar & Clin Tria.* 8(4) : 000225.
- Ruiz-Valderrama, L., Mendoza-Sánchez, J.E., Rodríguez-Tobón, E., Arrieta-Cruz, I., González-Márquez, H., Salame-Méndez, P.A., Tarragó-Castellanos, R., Cortés-Barberena, E., Rodríguez-Tobón, A. and Arenas-Ríos, E. (2025). High-Fat Diets Disturb Rat Epididymal Sperm Maturation. *Int. J. Mol. Sci.* 26:1850.
- Sakamoto, J., Hashimoto, K. (1986). Reproductive toxicity of crylamide and related compounds in mice: Effect on Fertility and sperm morphology. *Arch. Toxicol.* 59: 201-205
- Schmiech, M., Lang, S.J., Ulrich, J., Werner, K., Rashan, L.J., Syrovets, T., Simmet, T. (2019). Comparative investigation of frankincense nutraceuticals: Correlation of boswellic and lupeolic acid contents with cytokine release inhibition and toxicity against triple-negative breast cancer cells. *Nutrients.* 11(10): E2341.
- Shen, K.P., Hao, C.L., Yen, H.W. (2016 ). Pre-germinated brown rice prevented high fat diet induced hyperlipidemia through ameliorating lipid synthesis and metabolism in C57BL/6J mice. *J Clin Biochem Nutr.* 15(117):1–6

- Sherif ,M., Kamel,R., Elmorsy,E. and Ahmed, S .K. (2019). Role of Boswellic acid in the treatment of peptic ulcer. *Az. J. Pharm Sci.* 60:122-145
- Shirwaikar, A., Rajendran, K., Dinesh Kumar, C., Bodla, R.(2004). Antidiabetic activity of aqueous leaf extract of *Annona squamosa* in streptozotocin– nicotinamide type 2 diabetic rats. *J Ethnopharmacol* .91:171–175.
- Trivedi, V.L., Soni, R., Dhyani, P., Sati, P., Tejada, S., Sureda, A., Setzer, W.N., Faizal Abdull Razis, A., Modu, B., Butnariu, M. and Sharifi-Rad, J. (2023). Anti-cancer properties of boswellic acids: mechanism of action as anti-cancerous agent. *Front. Pharmacol.* 14:1187181. doi: 10.3389/fphar.2023.118718
- Wagner, H., Cheng, J.W., Ko, E.Y. (2019). Role of reactive oxygen species in male infertility : an updated review of literature. *Arab J Urol.* 16(1):35–43
- World Health Organization (WHO)(2012). WHO laboratory manual for the examination and processing of human semen. 6th ed.
- Yokoi, K. & Mayi, Z. K. (2004). Organ apoptosis with cytotoxic drugs. *Toxicology*, 290,8- 85