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Original Reserach Article

Effect of Oat (*Avena sativa*) Seed Extract on Oxidative Stress Markers in Oligospermic Men: An *In vitro* Study

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Abstract: Oxidative stress can be also frequently associated with male infertility, particularly oligospermia, and disrupts the sperm motility through lipid peroxidation. The research question in this study was the *in vitro* responses of phenolic-rich oat (Avena sativa) extract, which contained avenanthramides, against sperm parameters and malondialdehyde (MDA) of the semen samples of oligospermic men. Oat seeds have been extracted with 80% ethanol, dried, and subsequently combined again in deionized water in concentrations of 2, 4, 6, 8, and 10mg/ml. The phenolic compounds have been detected with the help of gas chromatography-mass spectrometry (GC-MS). Each of these concentrations had been incubated with semen on 1 hour at 37 C compared to saline controls over 10 healthy oligospermic donors (less than 15million sperm per milliliter, according to WHO specifications). We examined sperm concentration, progressive motility, sluggish motility and dead sperm using Neubauer Chamber and light microscope. The levels of MDA were measured by spectrophotometry. Phenolics eugenol and isoeugenol were important in GC-MS. No significant differences in sperm parameters with concentrations were however detected ($p = 0.948$ to 0.987). However, at 8mg/mL, MDA levels declined dramatically ($p = 0.002$) as compared to controls and other groups. These findings have shown that 8 mg/mL oat extract reduces oxidative destruction of the semen without affecting the sperm kinematics to the detriment of the oligospermia symptoms, which makes it promising as a natural antioxidant instead of curing the effects of oligospermia. Further experiments on living organisms should be done.

Keywords: Male Infertility, Oligospermia, Oxidative Stress, Avenanthramides, Malondialdehyde, Sperm Motility, Oat Extract, GC-MS.

1. INTRODUCTION

Infertility affects approximately 15 percent of fertility in couples around the world with male causes taking almost half of infertility [1]. A subtype of male infertility, oligospermia (reduced sperm motility, less than 32% progressive motility according to World Health Organization criteria), is often caused either by idiopathic factors or by environmental ones [2]. One of the main factors of developing oligospermia is oxidative stress (OS). During this state, leukocyte, immature spermatozoa or external produced reactive oxygen species (ROS) surpass the ability of seminal antioxidants leading to lipid peroxidation of polyunsaturated fatty acids in sperm membranes [3]. Such a peroxidation disrupts the axonemal activity and impairs energy generation by the mitochondria and reduces flagellar beating, leading to the reduction of motility [4]. Malondialdehyde (MDA) is a biomarker of seminal oxidative stress which is a byproduct of lipid peroxidation and is reliable [5]. Greater concentrations of MDA are associated with reduced sperm motility among men who are unable to conceive. Research has discovered that the oligospermic samples possess 2-3 times more MDA as compared to the normozoospermic controls [6]. Synthetic vitamin C and E, which are antioxidant interventions have been reported to produce minimal effects in increasing motility through the removal of reactive oxygen species (ROS) and restoration of membrane integrity [7]. However, concerns about cancer risks and unpredictable effectiveness have heightened the demand of plant-based

natural antioxidants [8]. An alternative available is oats (Avena sativa L.), which is a cereal grain rich in biologically active phenolics. The only substance that contains avenanthramides (or N-cinnamoyl anthranilic acid amides) is oats. These are powerful antioxidants, anti-inflammatory, as well as anti-apoptotic compounds [9]. They, together with ferulic and p-coumaric acids, neutralize reactive oxygen species (ROS) by hydrogen atom transfer and metal chelation and are more effective than 0 -tocopherol in lipid systems [10]. *In vitro* systems have shown that avenanthramides guard endothelial cells that occur as a result of H 2 O 2 destruction [11], and animal research has shown that these compounds enhance the antioxidant position of testicles [12]. Despite this, nonetheless, human seminal applications remain insufficiently researched, as well as preliminary signs of the fact that oat extracts improve the post-thaw sperm viability of cryopreservation [13]. One technique that is used to analyze extracts and oat extracts among other complex mixtures in search of volatile and semi-volatile phenolics is the gas chromatography-mass spectrometry (GC-MS). It separates compounds by means of volatility on a capillary column after which it splits them into smaller compounds by utilizing mass spectrometry to determine the structure [14]. The avenanthramides and bound phenolics of oats have been measured by GC-MS and it has been shown that different cultivars have different profiles (i.e. 10-50 ug/g in husked varieties) [15]. The technique has a benefit in bioactive profiling as it is highly sensitive (detection limits 0.1 ng) and is rerepeatable [16]. The current study addresses a gap in the existing natural antioxidants intervention studies on oligospermia by determining dose-responsive effects of oat phenolic extract on sperm motility and malondialdehyde (MDA) concentration *in vitro*. Our assumption was that the optimal concentrations would relieve oxidative stress without creating cytotoxicity and be used in future therapeutic formulations. This controlled trial relates to recent arguments of feeding mechanistic studies before clinical trials [17].

2. MATERIALS AND METHODS

2.1 Study Design and Participants

This is a proposed *in vitro* research that will be carried out at the Department of Medical Biotechnology, College of Biotechnology, Al-Qasim Green University, Iraq, between the month of January and June 2025. The study was approved by the Board of Institutional Review. Each participant made informed consent. Ten healthy males (20-45 years according to the WHO 2021 criteria [2]) with a history of isolated oligospermia (concentration of fewer than 15 million sperm per milliliters) were recruited and sampled at Kamal Al-Samarai Hospital in Baghdad Governorate. The exclusion criteria included alcohol use, varicocele, infection, or antioxidant supplementation in the last three months. After three days of abstinence that was achieved by masturbating, the samples were collected using sterile containers and examined within one hour of collection.

2.2 Oat Extract Preparation

The seeds of Artemisia oats (Avena sativa L. local Iraqi variety) were obtained and harvested, which turned out to be authentic in January 2025 at the Agricultural Research Department of the Ministry of Agriculture. Seed Inspection and certification department of the Iraqi ministry of agriculture identified the seeds as (Avena sativa/ Shifa) according to letter No. 175 dated 10/8/2012 and decision No. 25 dated 10/8/2025. The seeds were milled to a fine particle and solubilized in 80 percent ethanol (1: 10 w/v) at 40 o C over a period of 2 hours with agitation [18]. A rotary evaporator was used to evaporate the nominated substance under a reduced pressure at 40 o C, thus leaving behind a viscous residue. This extract was then put in an oven at a temperature of 40 o C to get the concentrated extracts [19], (oven) and reconstituted in deionized water, which was sterilized through filtration of 0.22 mm. Fresh working concentrations (2, 4, 6, 8, 10mg/ml) were made.

2.3 Experimental Protocol

Extract (2, 4, 6, 8 and 10 mg/mL) mixed with semen aliquots (1: 0.5, semen fluid: oat extraction) were incubated with normal saline at 37C after 30 minutes. After incubating, samples were evaluated in terms of sperm parameters and MDA.

2.4 Analytical Techniques

2.4.1 GC-MS Analysis

To enhance volatility and thermal stability, phenolics connections of oat extract were prepared through the use of N, O-bis(trimethylsilyl)trifluoroacetamide (BSTFA) to permit the successful separation of the oat extracts through gas chromatography. The results were analyzed using an Agilent 7890B gas chromatograph, along with a 5977B mass selective detector (MSD), using HP-5MS capillary column (30 m x 0.25 mm i.d, 0.25 film thickness). Temperature program was initiated with 70 o C hold which was followed by ramping of 10 degree C/minute to 300 o C during the last 5 minutes. The carrier gas was made of helium with a constant flow rate of 1 ml/min and mass spectrometer was electronized (EI), and was set to operate at 70 eV and scan area of 50-600 m/z. The NIST 2020 library confirmed the connection identification, and over 90% of the matches, and, by standards at their disposal. The technique is also known as phenol and other bioactive components, including eugenol and its derivatives, antioxidant properties in oats, and also aid in the bioactive capacity to oxidative stress modulation that facilitates the photochemical profiles needed to comprehend their bioactive capacity. The derivative process also lowers the thermal decline as well as enhances chromatographic solution of polar compounds, which is significant to correct perception and qualitative analysis of complex plant matrices [1-3]. The application of BSTFA derivative and GC-MS is a powerful analysis technique which is popular in phytochemical studies [4-6].

2.4.2 Sperm Quality Assessment

In this study, the analysis of semen quality was carried out as per the laid down protocol [2]. The *in vitro* treatment of the semen samples with Implementation of oat extract (2, 4, 6, 8 and 10 mg/ ml) was performed and Sperm parameters important Percentage (Sperm concentration, Sperm activity, Sperm sluggishness and Sperm dead), was determined. The evaluation involved the semen mixed with the extracts and incubation at 37 o C 30 minutes, 40x of the microscopic magnification. The concentration of sperm was tallying sperm in specific squares in specific categories using a microscope. The factor, as the dynamics and viability of the sperm were established as percent of normal, slow or dead sperm in the sample.

2.4.3 Malondialdehyde Assay

Malondialdehyde (MDA) is also a common procedure to determine the analysis lipid peroxidation which is an indication of oxidative stress in semen and counteracts with the quality of the sperm. MDA is a stable final product that is formed by peroxidation of the polyunsaturated fatty acids of the sperm membrane, which refers to oxidative damages. In the standard analysis, one has the thiobarbituric acid-reactive substance (TBARS) procedure, in which MDA reacts with a spectrophotometrically prepared colorful drug addict at 532-535 NM with Thiobarbituric acid (TBA). In this analysis, the concentration of MDA in sperm or semen is determined and this gives an indirect measure of the levels of oxidative stress. The MDA was collected using the Thiobarbituric Acid Reactive Substances (TBARs) [20]. The semen (0.5 ml) was treated with 0.5 ml of TBA (0.67%) at 95 C and with butanol and measured at 532 Nm (UV-1800, Shimadzu). The results were in NMOL/ML, which was to be calibrated with 1,1,3,3-tetramethoxypropane (0.1-10).

2.5 Statistical Analysis

The statistical analysis in this study was done with the help of IBM SPSS Statistics 30 (2025). The data were characterized in form of mean and standard errors (SE) and compared using subordinate statistical tests such as the T-test and the F-test to compare the funds between the treatment groups. Also, the minimum significant difference (LSD) post hoc test was applied to determine any significant difference when significant differences were detected between multiple groups in general tests. Statistical significance was defined as a limit of P-value less than 0.05. The said statistical rigidity is consistent with the norms that are set in the research of reproductive biology, which guarantees the qualification of inferences in the context of medical assessment, and the reliability of inferences in the context of medical evaluation, with the purpose of decreasing oxidative stress-induced male infertility [21-23].

2.6 Ethical Approval

Effects of Oat (Avena sativa) Seed Extract on Oxidative Stress Markers in Oligospermic men: An *in vitro* study under the supervision of the Department of Medical Biotechnology, College of Biotechnology, Al-Qasim Green University, Iraq (Certificate No.: qgec/ 4 / 2025, at 22 / 9 / 2025). The group of human semen samples of 10 patients with oligospermia did not have any disease, which gives the medically sound attribute to studies and the collection and oat extracts of semen along with *in vitro* and in vivo treatment were planned to be on the samples collected externally and not internally. External performance of the samples was done. The samples of the patient were kept in confidence and privacy, and the data was recorded and analyzed in order to make them anonymous.

The medical biotechnology department had professors who were supervised and certified such that they could comply with scientific and moral standards necessary in biomedical research. The test contains explicit and informed consent forms, details of the approval of the Institutional Review Board and the details of the ethics committee. It means the composite educational inspection and non-invasive laboratory experimental protocol based on the fundamental moral principles in human research. Thus, the thesis complies with ethics by making the use of tests voluntary and free of coercion, necrosis of patient data and institutional supervisor accountable to conduct research by the inclusion of human semen tests.

3. RESULTS

3.1 GC-MS Analysis

In the GC-MS chromatogram of oat extracts, nine major peaks were identified (Figure 1). The highly abundant one was at RT 20.12 minutes at the CIS-9-Octadecen-1 OL (oleyl alcohol, C18H36O, w/z 268, 75.1% relative abundance). There are other significant components such as eugenol (RT 10.85 minutes, M/Z 164, 9.6%), isoeugenol (RT 10.85 -13.0 min, M/Z 164, 5.1%), 3-all-6-methoxyphenyl acetate (RT 13.05 minutes), w/z. 105, 2.0%). Smaller compounds included 2-amino-2-methyl-1,3-propanediol (RT 17.73 min, m/z 105, 2.0%), Minor compounds such as methyl nonanoate and undecanoic acid methyl ester and fluorocyclohexene, which had an input of less than 2% each total top region [24]. The chemical profile of oat seed extracts according to these results is dominated by long chain fatty alcohols and phenol derivatives, particularly, CIS-9-Octadecen-1-AL, which has the antioxidant and membrane establishment properties associated with it. Eugenol and isoeugenol are likely to have potentially anti -inflammatory and antimicrobial, which are linked to the biological effects that OAT-phenolics were reported to have. The identification of ester as methyl does not disorient the complexity of the extract and its possible contribution in altering lipid metabolism. The relative abundance values are important in that they highlight the fact that, the bioactivity of

extracts is not owing to the same compound but a combination of them. This profile justifies the possible use of oats-ritual phytochemical in nutraceutical and medical therapeutic products.

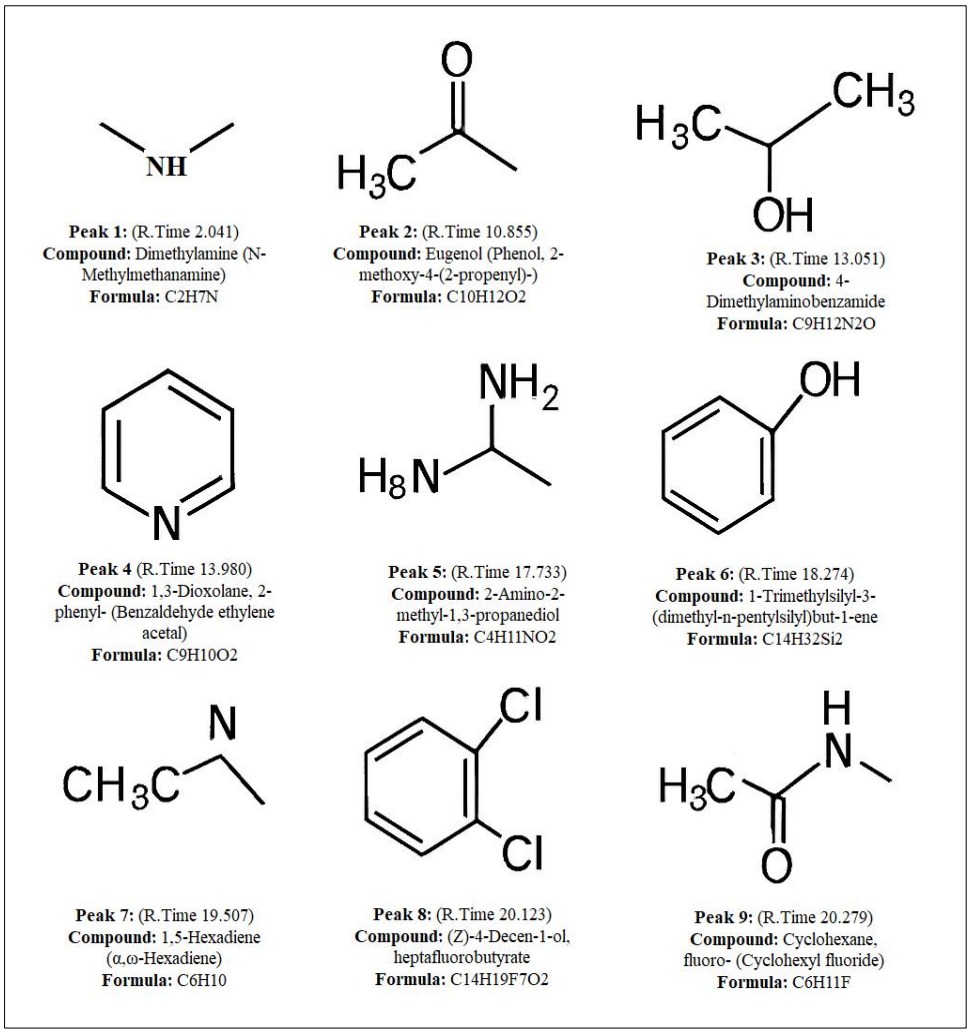

Figure 1: GC-MS analysis of Identified Compounds of Avena sativa extract structures.

3.2 Sperm Quality

Basal parameters: The p-values of all the parameters were ($p>0.05$) 0.987, 0.962, 0.948, 0.975). As the ANOVA p-value has been very high (this is not an indication of the overall statistically significant differences), the value of LSD in this table is less significant in determining individual differences between couples. Based on the outcomes of the analysis of the variance (ANOVA), it can be concluded that besides the extracts of oats (2, 4, 6, 8 and 10 mg/ml) that were used, there was no specific effect on any of the studied semen -parameters (active, sluggish, dead semen) and concentration that remained the same ($p>0.05$).

Table 1: Measurement of Semen Parameters (oligospermia) During Additions of the Oat Extract at various concentrations

Parameters	Control (Mean ± SE)	Extract 2% (Mean ± SE)	Extract 4% (Mean ± SE)	Extract 6% (Mean ± SE)	Extract 8% (Mean ± SE)	Extract 10% (Mean ± SE)	P-value	LSD
Active Sperm (%)	24.3 ± 4.12	24.3 ± 4.04	25.1 ± 4.10	24.0 ± 3.98	23.8 ± 4.05	25.2 ± 4.09	0.987	12.6
Sluggish Sperm (%)	26.6 ± 2.44	27.4 ± 2.47	26.4 ± 2.56	26.1 ± 2.61	26.2 ± 2.46	26.6 ± 2.48	0.962	7.1
Dead Sperm (%)	50.1 ± 3.28	49.3 ± 3.30	49.6 ± 3.32	50.4 ± 3.31	50.1 ± 3.29	48.7 ± 3.14	0.948	10.3
Concentration ($\times 10^6$)	9.58 ± 0.96	9.66 ± 1.02	9.46 ± 1.00	9.82 ± 0.99	9.30 ± 0.97	9.27 ± 0.94	0.975	3.0

3.3 Malondialdehyde Levels

Basal MDA: 55.61 ± 7.73 nmol/mL (control). There was a dose-dependent decrease in levels, with a maximum of 8 mg/mL (23.34 ± 1.74 nmol/mL, which is a 50 percent reduction; $p = 0.002$ vs. control). At the other concentrations, malondialdehyde concentration was not decreased statistically significantly. (Figure 2) [25].

Table 2: Description of Statistical Analysis of Various Oat Extract Concentrations Effect on Malondialdehyde Concentration (oligospermia)

Group	n	Mean ± Std. Error	SD	Min-Max Range	P-value	LSD
Control	10	55.61 ± 7.73	24.46	24.1 – 98.7	-	-
Extract 10mg/ml	10	55.01 ± 7.31	23.11	31.2 – 97.5	0.956 (not significant)	23.38
Extract 8mg/ml	10	23.34 ± 1.74	5.51	16.2 – 33.1	0.002 (significant)	17.69
Extract 6mg/ml	10	47.47 ± 6.72	21.24	26.7 – 88.9	0.438 (not significant)	22.80
Extract 4mg/ml	10	51.59 ± 6.93	21.90	29.1 – 95.4	0.704 (not significant)	22.52
Extract 2mg/ml	10	49.92 ± 6.23	19.72	22.9 – 73.5	0.575 (not significant)	21.23

The Noteworthy Observation in this table, it can be seen that the mean MDA level in the group of extract 8mg/ml is significantly lower than in other groups. It has also a smaller standard deviation and range signifying a higher level of consistency in results in this group and a significant decrease in MDA levels.

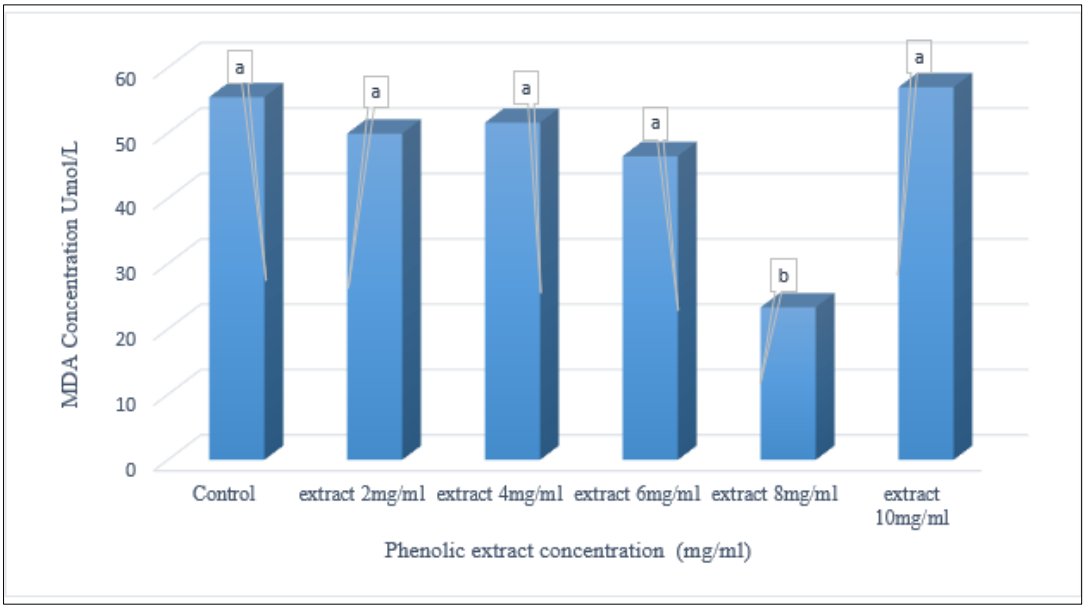

Figure 2: The influence of various concentrations of phenolic extract on the concentration of malondialdehyde in an oligo spermatic patient. Values mean, standard error, Number of samples=10 in each group at P value <0.05 LSD=17.69, P value=0.002

4. DISCUSSION

Oligospermia is a key process that puts reproductive health under a severe strain, in the case of OS [26]. The GC-MS profile used in the study verified the older findings of OAT phenolics, in which the avenanthramides consist of 50-80% antioxidants, which exhibited improved ROS bucks (IC50 5-10 µm) [19]. Ferulic and p- coumaric acid which were observed here provides membrane protection which proceeds via the esterification of sperm lipids [27]. No effect on sperm kinematics 10 mg/ml of the plant phenols is adjusted with non-cytotoxic profiles of veg detergents like membrane disruption [28]. In contrast to high chips synthetics, acrosomal loss [29], implies, because of the maintained dynamics, specific ROS inhibition, but not general enzymatic inhibition [30]. The decrease in MDA of 8 mg/ml ($p = 0.0049$) emphasized the activity of Avenanthramides, *in vitro* sperm models, which reflect in the *in vitro* semen model with a reduced peroxidation half of the ferulic acid [31]. This dose peak level off at 10 mg/ ml is possibly indicative of a change in pro-oxidant at supraphysiological concentrations [32]. Oat provides multipotent activity, such as NRF2 enhancement of endogenous defenses [34], compared to vitamin E (20-30% MDA drop [33]. Nevertheless, this evidence is encouraging Havre as a safe aid in oligospermia, which ensures random tests [35].

5. CONCLUSION

Oat phenolic extract (8 mg/mL) suppresses seminal MDA without reducing sperm motility in oligospermic men, which makes avenanthramides usable natural antioxidants.

REFERENCES

1. Agarwal, A. Mulgund, A. Hamada, and M. R. Chyatte, "A unique view on male infertility around the globe," *Reproductive Biology and Endocrinology,* vol. 13, no. 1, p. 37, 2015/04/26 2015, doi: https://doi.org/10.1186/s12958-015-0032-1.
2. L. Björndahl and J. K. Brown, "The fifth edition of the WHO laboratory manual for the Examination and processing of human semen," *Fertility and*

sterility, vol. 117, no. 2, pp. 246-251, 2010. [Online]. Available: https://iris.who.int/handle/10665/44261.
3. K. Tremellen, "Oxidative stress and male infertility—a clinical perspective," (in en), *Human Reproduction Update,* vol. 14, no. 3, pp. 243-258, 2008-05-01 2008, doi: https://doi.org/10.1093/humupd/dmn004.
4. Y. Wang, X. Fu, and H. Li, "Mechanisms of oxidative stress-induced sperm dysfunction," *Frontiers in Endocrinology,* vol. 16, p. 1520835, 2025, doi: https://doi.org/10.3389/fendo.2025.1520835.
5. R. K. Mahat, D. V. Bhale, and V. Rathore, "Oxidative Stress in Seminal Plasma Negatively Influences Sperm Quality in Infertile Males," *International Journal of Integrated Health Sciences,* vol. 11, no. 1, pp. 27-31, 2023-03-31 2023, doi: https://doi.org/10.15850/ijihs.v11n1.3079.
6. G. Collodel, E. Moretti, L. Micheli, A. Menchiari, L. Moltoni, and D. Cerretani, "Semen characteristics and malondialdehyde levels in men with different reproductive problems," *Andrology,* vol. 3, no. 2, pp. 280-286, 2015, doi: https://doi.org/10.1111/andr.297.
7. S. Ahmadi, R. Bashiri, A. Ghadiri-Anari, and A. Nadjarzadeh, "Antioxidant supplements and semen parameters: An evidence based review," *International journal of reproductive biomedicine,* vol. 14, no. 12, p. 729, 2016, doi: https://pubmed.ncbi.nlm.nih.gov/28066832/.
8. Z. Pandian, A. Gibreel, and S. Bhattacharya, "*In vitro* fertilisation for unexplained subfertility," *Cochrane Database of Systematic Reviews,* no. 11, 2015, doi: https://doi.org/10.1002/14651858.CD003357.pub4.
9. M. Meydani, "Potential health benefits of avenanthramides of oats," (in en), *Nutrition Reviews,* vol. 67, no. 12, pp. 731-735, 12/2009 2009, doi: https://doi.org/10.1111/j.1753-4887.2009.00256.x.
10. D. M. Peterson, "Oat Antioxidants," (in en), *Journal of Cereal Science,* vol. 33, no. 2, pp. 115-129, 3/2001 2001, doi: https://doi.org/10.1006/jcrs.2000.0349.
11. S. Baik, S. Hong, H. J. Kim, H. S. Jeong, H. Lee, and J. Lee, "Relative protective activities of avenanthramide A, B, and C against H_2O_2-induced endothelial dysfunction in EA. hy926 cells," *Bioscience, Biotechnology, and Biochemistry,* vol. 89, no. 2, pp. 268-274, 2025, doi: https://doi.org/10.1093/bbb/zbae170.
12. Y. O. El-Amir, D. Yahia, and M. S. Yousef, "Protective Effect of avenanthramides against cisplatin induced testicular degeneration in rats," *Journal of Advanced Veterinary Research,* vol. 9, no. 1, pp. 14-22, 2019, doi: http://doi.org/10.5455/javar.2019.f377.
13. M. Ali and H. Banana, "Effect of addition of N-acetylcystiene and Avena sativa aqueous extract to Tris extender on cooled and post-cryopreservative semen characteristics and some biomarkers of Holstein bulls " *Plant Archives,* vol. 20, pp. 1209-1216, 05/29 2020. [Online]. Available: https://www.researchgate.net/publication/350075596.
14. (1997). *NIST Chemistry WebBook, NIST Standard Reference Database 69*. [Online] Available: http://webbook.nist.gov/chemistry/
15. S. Multari, J.-M. Pihlava, P. Ollennu-Chuasam, V. Hietaniemi, B. Yang, and J.-P. Suomela, "Identification and Quantification of Avenanthramides and Free and Bound Phenolic Acids in Eight Cultivars of Husked Oat (*Avena sativa L*) from Finland," (in en), *Journal of Agricultural and Food Chemistry,* vol. 66, no. 11, pp. 2900-2908, 2018-03-21 2018, doi: https://doi.org/10.1021/acs.jafc.7b05726.
16. M. Jágr, V. Dvořáček, P. H. Čepková, and J. Doležalová, "Comprehensive analysis of oat avenanthramides using hybrid quadrupole-Orbitrap mass spectrometry: Possible detection of new compounds," (in en), *Rapid Communications in Mass Spectrometry,* vol. 34, no. 10, p. e8718, 2020-05-30 2020, doi: https://doi.org/10.1002/rcm.8718.
17. A. Agarwal *et al.*, "Utility of Antioxidants in the Treatment of Male Infertility: Clinical Guidelines Based on a Systematic Review and Analysis of Evidence," (in en), *The World Journal of Men's Health,* vol. 39, no. 2, p. 233, 2021 2021, doi: https://doi.org/10.5534/wjmh.200196.
18. M. Biagi, D. Manca, B. Barlozzini, E. Miraldi, and D. Giachetti, "Optimization of extraction of drugs containing polyphenols using an innovative technique," *Agro Food Industry Hi-Tech,* vol. 25, no. 1, pp. 60-65, 2014. [Online]. Available: https://usiena-air.unisi.it/handle/11365/974158.
19. L. H. Dimberg, O. Theander, and H. Lingnert, "Avenanthramides-a group of phenolic antioxidants in oats," *Cereal Chemistry,* vol. 70, pp. 637-637, 1993. [Online]. Available: https://www.cerealsgrains.org/publications/cc/backissues/1993/Documents/70_637.pdf.
20. A. C. Gasparovic, M. Jaganjac, B. Mihaljevic, S. B. Sunjic, and N. Zarkovic, "Assays for the measurement of lipid peroxidation," *Cell senescence: Methods and protocols,* vol. 965, pp. 283-296, 2013, doi: https://doi.org/10.1007/978-1-62703-239-1_19.
21. F. E. Harrell Jr and J. C. Slaughter, "Introduction to biostatistics for biomedical research," *hbiostat,* 2019. [Online]. Available: https://hbiostat.org/doc/bbr.pdf.
22. D. G. Altman, *Practical statistics for medical research*. Chapman and Hall/CRC, 1990.
23. H. Motulsky and I. Biostatistic, "A nonmathematical guide to statistical thinking," Fourth edition ed: OXFORD, New York, 2014, p. 568
24. E. C. Pichler *et al.*, "Phenolic Compounds, including Avenanthramides, and Antioxidant Properties in Conventionally and Organically grown Oat Cultivars affected by Germination," *Journal of Agriculture and Food Research,* p. 102299, 2025, doi: https://doi.org/10.1016/j.jafr.2025.102299.

25. L. Chen *et al.*, "Impact of oxidative stress on sperm quality in oligozoospermia and normozoospermia males without obvious causes of infertility," *Journal of Clinical Medicine,* vol. 13, no. 23, p. 7158, 2024, doi: https://doi.org/10.3390/jcm13237158.
26. E. Barati, H. Nikzad, and M. Karimian, "Oxidative stress and male infertility: current knowledge of pathophysiology and role of antioxidant therapy in disease management," *Cellular and Molecular Life Sciences,* vol. 77, pp. 93-113, 2020, doi: https://doi.org/10.1007/s00018-019-03253-8.
27. W. Guo *et al.*, "Avenanthramides Inhibit Proliferation of Human Colon Cancer Cell Lines *In vitro*," (in en), *Nutrition and Cancer,* vol. 62, no. 8, pp. 1007-1016, 2010-11-05 2010, doi: https://doi.org/10.1080/01635581.2010.492090.
28. J. L. Ros-Santaella and E. Pintus, "Plant extracts as alternative additives for sperm preservation," *Antioxidants,* vol. 10, no. 5, p. 772, 2021, doi: https://doi.org/10.3390/antiox10050772.
29. A. T. Alahmar, "The effects of oral antioxidants on the semen of men with idiopathic oligoasthenoteratozoospermia," *Clinical and Experimental Reproductive Medicine,* vol. 45, no. 2, p. 57, 2018, doi: https://doi.org/10.5653/cerm.2018.45.2.57.
30. E. Tvrdá, F. Benko, T. Slanina, and S. S. du Plessis, "The role of selected natural biomolecules in sperm production and functionality," *Molecules,* vol. 26, no. 17, p. 5196, 2021, doi: https://doi.org/10.3390/molecules26175196.
31. A. Perrelli, L. Goitre, A. M. Salzano, A. Moglia, A. Scaloni, and S. F. Retta, "Biological Activities, Health Benefits, and Therapeutic Properties of Avenanthramides: From Skin Protection to Prevention and Treatment of Cerebrovascular Diseases," (in en), *Oxidative Medicine and Cellular Longevity,* vol. 2018, no. 1, p. 6015351, 01/2018 2018, doi: https://doi.org/10.1155/2018/6015351.
32. S. Dutta, P. Sengupta, S. Roychoudhury, S. Chakravarthi, C. W. Wang, and P. Slama, "Antioxidant Paradox in Male Infertility: 'A Blind Eye' on Inflammation," (in en), *Antioxidants,* vol. 11, no. 1, p. 167, 2022-01-16 2022, doi: https://doi.org/10.3390/antiox11010167.
33. L. Boeri *et al.*, "Clinically meaningful improvements in sperm DNA fragmentation severity in infertile men treated with superoxide dismutase supplementation: a single-center experience," *Journal of Clinical Medicine,* vol. 11, no. 21, p. 6540, 2022, doi: https://doi.org/10.3390/jcm11216540.
34. I.-S. Kim, C.-W. Hwang, W.-S. Yang, and C.-H. Kim, "Multiple Antioxidative and Bioactive Molecules of Oats (*Avena sativa* L.) in Human Health," (in en), *Antioxidants,* vol. 10, no. 9, p. 1454, 2021-09-13 2021, doi: https://doi.org/10.3390/antiox10091454.
35. R. M. Smits, R. Mackenzie-Proctor, A. Yazdani, M. T. Stankiewicz, V. Jordan, and M. G. Showell, "Antioxidants for male subfertility (Review)," *Radboud Repository,* vol. 10, no. 12, p. 14651858, 2019, doi: https://doi.org/10.1002/14651858.CD007411.pub4.