



# Investigating the protective role of *Foeniculum Vulgare* against reproductive toxicity induced by Zinc Oxide Nanoparticles: A study on Oxidative stress and Hormonal disruption in Male Rats

Vijender Singh<sup>1\*</sup>, Dr. Priyanka Sharma<sup>2</sup>

1. Research Scholar, Department of Zoology, Lords School of Sciences, Lords University, Alwar, Rajasthan, India  
vijenderpayal2010@gmail.com ,
2. Professor, Department of Zoology, Lords School of Sciences, Lords University, Alwar, Rajasthan, India

**Abstract:** Zinc oxide (ZnO) nanoparticles are widely employed in various industries due to their unique physicochemical properties; however, their increasing environmental and biological exposure has raised significant health concerns the scientific community as a whole is becoming more interested in nanotechnology. biomedical applications are just a few of the many uses for zinc oxide nanoparticles (ZnO NPs). The injection of ZnO nanoparticles in the present investigation increased serum MDA levels and decreased serum CAT levels relative to the control group.

**Keywords:** Nanoparticles, Zinc, Oxide, physicochemical , biological

----- X -----

## INTRODUCTION

A primary worry is reproductive toxicity, especially regarding male fertility, where oxidative damage may disrupt spermatogenesis and hormonal equilibrium. ZnO nanoparticles are recognized for producing reactive oxygen species (ROS), resulting in oxidative stress, lipid peroxidation, DNA damage, and death in testicular tissues. Moreover, they may interfere with the hypothalamic-pituitary-gonadal (HPG) axis, modifying hormone levels and sperm characteristics, including motility, morphology, and viability.

ZnO NPs have the ability to combat major uncontrolled diseases like cancer. It has been shown that the plant can lower free radical concentration, which means it may protect against oxidative reaction-related disorders. Publications have shown that zinc oxide nanoparticles possess antibacterial, antioxidant, and anticancer capabilities. If these NPs are successful, they will hopefully allow for the growth of cutting-edge treatments for a wide range of devastating illnesses.

The calming effects of fennel on nerves and stress levels Potential for memory enhancement was also mentioned by them. There are a number of natural compounds, including vital fatty acids, that make this plant nutritionally useful. Extracts of *F. vulgare* have also yielded phenolic chemicals including flavonoids. The toxicity of ZnO Nps is lower than that of other metallic nanoparticles. These NPs have antibacterial characteristics, according to the research.

The scientific community as a whole is becoming more interested in nanotechnology. It is one of the many exciting and ever-changing areas of study centered on materials at the nanoscale. It is possible to create nanoparticles (NPs) chemically, physically, or biologically; these NPs serve as the fundamental units of nanotechnology.

They are widely used in cosmetics, food packaging, pharmaceutical delivery systems, and electrical equipment. The rapid rise in the use of ZnO nanoparticles has elicited much apprehension over their possible toxicity in biological systems. Zinc oxide (ZnO) nanoparticles, widely utilized in several sectors, have been documented to cause reproductive damage in male rats through the induction of oxidative stress and the disruption of hormonal equilibrium. This research examines the preventive effects of phytochemicals derived from *Foeniculum vulgare* against reproductive toxicity induced by ZnO nanoparticles.

Zinc oxide nanoparticles are extensively utilized in cosmetics, electronics, and biological fields. Nonetheless, their diminutive size facilitates effortless infiltration into biological systems, where they can induce oxidative stress, inflammation, and cellular damage. In male rats, ZnO nanoparticles have been demonstrated to disturb hormonal equilibrium by influencing testosterone and luteinizing hormone concentrations, cause testicular tissue damage resulting in compromised spermatogenesis, and elevate lipid peroxidation while diminishing antioxidant enzyme activity.

## LITREATURE REVIEW

**Borotová, Petra et.al. (2021).** A medicinal herb, one other flavoring component is *Foeniculum vulgare* Mill. The food sector may use the essential oil of *F. vulgare*, which may kill bugs and germs —to keep pests and pathogens from contaminating food supplies and food products. utilizing gas chromatography/mass spectrometry (GC/MS) and gas chromatography (GC-FID), the research intended to characterize the volatile components of essential oil from *F. vulgare*, as well as analyze the antibacterial activity in the vapor phase and utilizing the disc diffusion method. Evidence of insecticidal action in the vapor phase of Essential oil from *F. vulgare* was also discovered. The main ingredients of the perfume made from the leaves of *F.*

**Ke, Weiwei e.al. (2021).** One of the most prevalent types of cancer is hepatocellular carcinoma, or HCC. Due to the lack of effective therapies and the absence of symptoms, the prognosis of HCC is quite dismal. It is still unclear how *Foeniculum vulgare* inhibits HCC via its molecular modes of action. Our findings establish that a seed extract of *Foeniculum vulgare* (75% ethanol) decreased cell viability, caused cell death, and successfully blocked the migration of HCC cells in laboratory tests. FVE considerably reduced Surviving levels, which in turn suppressed HCC development in vivo, according to HCC xenograft experiments in nude mice.

**Mehra, Nisha et.al. (2021).** One spice that Worldwide, people in both tropical and temperate regions love the *Foeniculum vulgare* plant, which is a member of the Apiaceae family. The fennel grown in India is mostly seeds come from arid and semiarid climates. The pharmacological features, such as antibacterial, antidiabetic, antioxidant, and anticancer actions, have just recently been discovered. A number of phytoconstituents, including flavonoids, glycosides, and others, are contained in fennel and have

therapeutic uses. Fennel has phenolic chemicals that are good for people's health. Isolated bioactive chemicals from this plant include trans-anethole, estragole, fenchone, kaempferol, quercetin, and rosmarinic acid, some of which have associations with potential human body systems.

**P. Aydınlik, Nur et.al. (2021).** As fragrant plants, parsley (*Petroselinum crispum*) and fennel (*Foeniculum vulgare*) are members of the Apiaceae and Lamiaceae families, respectively. Phytochemical testing identified the presence of physiologically active chemicals including components such as glycosides, alkaloids, terpenoids, steroid, tannin, and flavonoids in the two plants' leaves that were separated using ethanol in a sequential fashion. According to GC-MS analysis, six bioactive compounds were found in *Petroselinum crispum*: cineole, l-limonene, cyclohexane, phenol, neophytadiene, and 9,12,15 octadecatrienoic. On the other hand, *Foeniculum vulgare* included two bioactive compounds, 1,4 cyclohexadiene and metronidazole.

**Wazir, Muhammad Asif. (2021).** Objectives and Background: Traditional remedies and natural goods are very important. In some parts of the globe, traditional medical practices including Unani, traditional Korean medicine, Kampo, Ayurveda, and traditional Chinese medicine have developed into well-organized medical systems. Herbal medicine made from the garlic plant (*Allium sativum* L.), a crude medication belonging hyperglycemia, cancer, cardiovascular disease, hypertension, infections, and stress are some of the conditions that this plant from the Liliaceae family can alleviate. The fruit of the Rosaceae plant, *Malus domestica* B., is used for the management and prevention of hyperlipidemia, diabetes, and other related conditions because to its antioxidant, antiproliferative, antibacterial, and anti-inflammatory characteristics, asthma, and diabetes properties.

## ZINC OXIDE (ZnO) NANOPARTICLES

Zinc Oxide (ZnO) nanoparticles (NPs) have attracted considerable interest recently because of their distinctive physicochemical characteristics and many commercial uses. These nanoparticles are extensively used in sectors including cosmetics, medicines, electronics, food packaging, and environmental cleanup. Their exceptional antibacterial, UV-blocking, and catalytic qualities render them highly sought after in diverse commercial items, such as sunscreens, textiles, and coatings. A primary benefit of ZnO nanoparticles is their efficacy as antibacterial and antifungal agents. The increased surface area to volume ratio improves their contact with microbial membranes, resulting in membrane rupture and consequent cellular demise.

ZnO nanoparticles have been shown to influence reproductive health. Investigations using animal models have shown that exposure to ZnO nanoparticles may result in testicular toxicity, hormonal dysregulation, and compromised spermatogenesis. The deleterious consequences are thought to arise from oxidative stress-related damage, inflammation, and the disturbance of the hypothalamic-pituitary-gonadal (HPG) axis. The toxicity of ZnO nanoparticles is affected by several parameters, such as particle size, surface charge, aggregation state, and exposure concentration. Smaller nanoparticles with a high surface charge exhibit increased reactivity and possible toxicity.

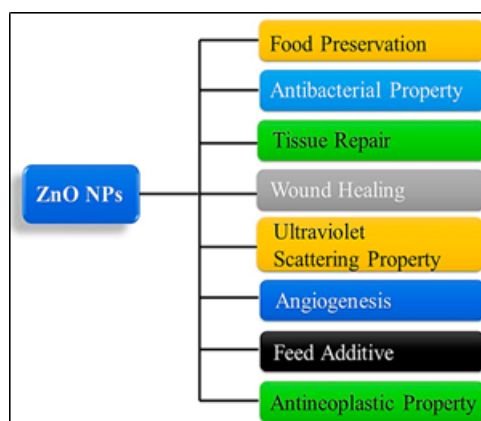
To alleviate possible health hazards, current research is concentrating on surface modifications and coatings to improve biocompatibility and diminish toxicity. Surface functionalization with

biocompatible polymers, proteins, or other stabilizing agents has shown potential in reducing ROS production and enhancing cellular tolerance. Although ZnO nanoparticles have several technical benefits, their capacity to interfere with biological systems requires a comprehensive study of their toxicological characteristics. Additional research is necessary to create safer formulations and to establish regulatory rules that assure their responsible use in consumer and healthcare products.

## **TYPICAL USES FOR NANOPARTICLES ZINC OXIDE**

A wide range of commercial items using nanomaterials have had significant influence on people's lives in recent years. These products include those in the medical field, food sector, cosmetics, animal husbandry, agriculture, and environmental protection. Sunscreens, textile coatings, antibacterial agents for food packaging, fertilizers, and even some biomedical applications are just a few of the many uses for zinc oxide nanoparticles (ZnO NPs). These NPs are also known for their physical distinctiveness and their ability to fight off fungi and bacteria. In addition, the release of ZnO NPs into the environment is an inevitable consequence of using commercial items.

These factors may have contributed to higher exposure levels to ZnO NPs, which has raised concerns about their safety. Male rats treated intraperitoneally with ZnO NPs had a decreased sperm count, slower motility, and more abnormalities, according to research by Abbasalipourkabir et al. Talebi et al. found that spermatogenic parameters such as sperm count, motility, abnormality percentage, seminiferous tubule diameter, seminiferous epithelium height, and maturation arrest were significantly altered when administered 50 and 300 mg/kg of ZnO NPs. This suggested that ZnO NPs had a negative effect on spermatogenesis and testicular toxicity.



**Figure 1: Visual representations of typical uses for ZnO NP. nanoparticles; zinc oxide**

Many different approaches, broadly categorized as either top-down or bottom-up procedures, may be used to synthesize NPs. In a bottom-up strategy, atom-sized particles are combined to form nanoparticles; in a top-down approach, bigger molecules are first broken down into smaller ones, and then these smaller ones are transformed into appropriate nanoparticles.



Figure 2: Standard procedures for the synthesis of nanoparticles of zinc oxide.

## NANOPARTICLE TOXICOLOGY AND ITS MECHANISM

There are primarily three entry points for nanoparticles into living organisms. The physicochemical properties of NPs determine the most common entry point, which is inhalation, followed by the skin, and finally, digestion, which is the rarest route. Particle size, form, surface charge, dispersion, hydrophobic and hydrophilic characteristics, and so on. Depending on their physicochemical qualities, nanoparticles have the potential to create ROS in a range of concentrations. In NPs based on transition metals, the existence of prooxidant functional groups, interactions between particles and cells, and active redox cycling on NP surfaces are the primary characteristics of NPs that enhance ROS generation.

In NPs based on transition metals, the existence of prooxidant functional groups, interactions between particles and cells, and active redox cycling on NP surfaces are the primary characteristics of NPs that enhance ROS generation. It has not been shown in all circumstances that oxidative stress plays a pivotal role in the harmful effects caused by NPs. This is because certain NPs might cause toxicity without inducing oxidative stress, for example, if they have an inactive surface or are poorly soluble.

## NANOPARTICLE-MEDIATED FREE RADICAL DAMAGE

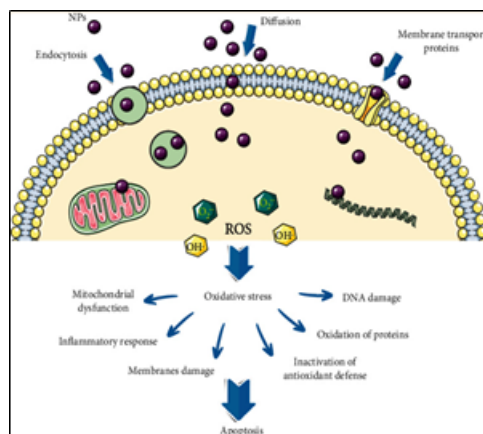
Oxidative stress is often produced by the interaction with nanoparticles leads to an excess of reactive oxygen species (ROS). Hypersensitivity to some types of oxygen-consuming molecules formation and their removal in interaction with antioxidant defense mechanisms is what term "oxidative stress" describes.

Cells and the body as a whole may be damaged when there is an imbalance between the prooxidant and antioxidant relations, which can lead to in-depth protein deterioration, lipids, and nucleic acids, among other biomolecules. While ROS creation during mitochondrial respiration or phagocytosis is a typical biological activity, there are a number of factors that might lead to an overproduction of these compounds. An organism may acquire serious illnesses if its antioxidant defense mechanisms, which include enzymes like glutathione (GSH), superoxide dismutase (SOD), and catalase (CAT), are unable to counteract the elevated levels of reactive oxygen species (ROS).

Nuclear particles (NPs) disrupted cellular processes by directly destroying cell and organelle membranes and by attaching to biomacromolecules, altering their shape and function. Reactive oxygen species generated inside cells (ROS) by NPs also damages cell organelles and membranes while influencing the

structure and function of the cell's primary components—lipids, DNA, proteins, and carbohydrates. The complicated processes of NP-induced toxicity include the critical roles played by mitochondria, lysosomes, and the endoplasmic reticulum (ER).

If the antioxidant defense is unable to restore the redox equilibrium or if NPS-induced mitochondrial dysfunction disrupts mitochondrial function, additional oxidative stress may develop via indirect mechanisms. Although NPs do not cause oxidative stress per se, they do increase ROS levels in cells via their effects on mitochondria and phagocytes. One example is the interaction between NPs and phagocytes, which are white blood cells and macrophages that are meant to digest them. However, since NPs often include inorganic components, the phagocytes become injured because they can't neutralize the inorganic compounds. The end consequence is a rise in reactive oxygen species (ROS) levels inside the cell, which in turn causes oxidative stress. The quantities of inflammatory factors such as  $\text{TNF-}\alpha$  and interleukins may be influenced by NPs, leading to mitochondrial dysfunction, ER stress, and DNA damage.



**Figure 3: Cell oxidative stress caused by nanoparticles: a synopsis of key developments**

Nuclear particles (NPs) disrupted cellular processes by directly destroying cell and organelle membranes and by attaching to biomacromolecules, altering their shape and function. Reactive oxygen species generated inside cells (ROS) by NPs also damages cell organelles and membranes while influencing the structure and function of the cell's primary components—lipids, DNA, proteins, and carbohydrates. The complicated processes of NP-induced toxicity include the critical roles played by mitochondria, lysosomes, and the endoplasmic reticulum (ER).

## **IMPAIRMENT OF SPERMATOGENESIS AND HORMONAL BALANCE**

ZnO nanoparticles negatively impact spermatogenesis by impairing Sertoli and Leydig cells, which are essential for sperm generation and hormone synthesis. Sertoli cells provide structural and nutritional support to growing germ cells, while Leydig cells are tasked with testosterone synthesis, crucial for sustaining spermatogenesis and male rat reproductive health.

The synergistic effects of cellular damage and hormone disturbance underscore the complex influence of ZnO nanoparticles on male reproductive health. Ongoing research is essential to investigate possible preventative methods and to formulate safe exposure recommendations to alleviate these detrimental consequences.



Exposure to ZnO nanoparticles produces oxidative stress and inflammation in testicular tissue, resulting in cellular damage and compromised function of these essential cells. The synergistic effects of cellular damage and hormone disturbance underscore the complex influence of ZnO nanoparticles on male reproductive health.

## **MECHANISM OF REPRODUCTIVE TOXICITY**

The reproductive toxicity caused by Zinc Oxide (ZnO) nanoparticles is a complex process involving many interconnected pathways. These pathways result in adverse consequences on male reproductive health, as shown by many in vivo and in vitro investigations. The principal mechanisms by which ZnO nanoparticles.

ZnO nanoparticles are recognized for producing excessive reactive oxygen species following cellular infiltration. Reactive oxygen species (ROS) are chemically active molecules that include oxygen, including free radicals such as superoxide anions ( $O_2^-$ ) and hydroxyl radicals ( $OH^-$ ), in addition to non-radical entities like hydrogen peroxide ( $H_2O_2$ ). These reactive compounds are generally generated in mitochondria as by-products of normal cellular metabolism. Nonetheless, ZnO nanoparticles may markedly increase ROS levels above physiological limits.

ZnO nanoparticles are recognized for producing excessive reactive oxygen species following cellular infiltration. Reactive oxygen species (ROS) are chemically active molecules that include oxygen, including free radicals such as superoxide anions ( $O_2^-$ ) and hydroxyl radicals ( $OH^-$ ), in addition to non-radical entities like hydrogen peroxide ( $H_2O_2$ ).

## **CONCLUSION**

The injection of ZnO nanoparticles in the present investigation increased serum MDA levels and decreased serum CAT levels relative to the control group. ZnO nanoparticles in the liver is attributable to their significant antioxidant activity. The robust antioxidant properties of fennel and anise have been previously shown. A wide range of commercial items using nanomaterials have had significant influence on people's lives in recent years. These products include those in the medical field, food sector, cosmetics, animal husbandry, agriculture, and environmental protection. The scientific community as a whole is becoming more interested in nanotechnology.

---

## **References**

1. Borotová, Petra & Galovičová, Lucia & Šimora, Veronika & Ďúranová, Hana & Vukovic, Nenad & Vukić, Milena & Babošová, Mária & Kacaniova, Miroslava. (2021). Biological activity of essential oil from *Foeniculum vulgare*. *Acta Horticulturae et Regiotecturae*. 24. 148-152. 10.2478/ahr-2021-0037.
2. Ke, Weiwei & Wang, Hongbo & Zhao, Xiangxuan & Lu, Zaiming. (2021). *Foeniculum Vulgare* Seed Extract Exerts Anti-Cancer Effects by targeting Survivin in Hepatocellular Carcinoma. *Food & Function*. 12. 10.1039/D0FO02243H.
3. Mehra, Nisha & Tamta, Garima & Nand, Vivekanand. (2022). Phytochemical screening and in vitro antioxidant assays in *Foeniculum vulgare* Mill. (Fennel) seeds collected from Tarai region in the Uttarakhand. *Indian Journal of Natural Products and Resources*. 13. 213-222. 10.56042/ijnpr.

v13i2.51347.

4. P. Aydınlik, Nur & ABUBAKAR, Jamaluddeen & Edo, Great. (2021). Phytochemical and GCMS analysis on the ethanol extract of *Foeniculum Vulgare* and *Petroselinum crispum* leaves. *International Journal of Chemistry and Technology*. 5. 10.32571/ijct.911711.
5. Wazir, Muhammad Asif. (2021). Phytochemical, Fluorescence and Antimicrobial evaluation of Poly herbal extract. *International Journal of Integrated Health Sciences*. 01. 41-57.
6. West LA, Horvat RD, Roess DA, Barisas BG, Juengel JL, Niswender GD. Steroidogenic acute regulatory protein and peripheral-type benzodiazepine receptor associate at the mitochondrial membrane. *Endocrinology*. 2001;142(1):502–505. doi:10.1210/endo.142.1.8052
7. Y. Yin, Q. Lin, H. Sun, D. Chen, Q. Wu, X. Chen, et al., Cytotoxic effects of ZnO hierarchical architectures on RSC96 Schwann cells, *Nanoscale. Res. Lett.*,7 (2012), 439-446. <http://dx.doi.org/10.1186/1556-276x-7-439>
8. Sadegh-Nejadi S., Aberomand M., Ghaffari M.A., Mohammadzadeh G., Siahpoosh A., Afrisham R.: Inhibitory effect of *Ziziphus Jujuba* and *Heracleum Persicum* on the activity of partial purified rat intestinal alpha glucosidaseenzyme. *J Mazandaran Univ Med Sci* 2016, 25, 135–146.
9. Salami, Maryam & Rahimmalek, Mehdi & Ehtemam, Mohammad. (2016). Inhibitory effect of different fennel (*Foeniculum vulgare*) samples and their phenolic compounds on formation of advanced glycation products and comparison of antimicrobial and antioxidant activities. *Food Chemistry*. 213. 10.1016/j.foodchem.2016.06.070.
10. Saliyani M, Jalal R, Goharshadi E. Mechanism of oxidative stress involved in the toxicity of ZnO nanoparticles against eukaryotic cells. *Nanomed J*. 2016;3(1):1–14.
11. Seid, Mohammed & Dekebo, Aman & Babu G., Neelaiah. (2018). Phytochemical Investigation and Antimicrobial Evaluation of *Foeniculum vulgare* Leaves Extract Ingredient of Ethiopian Local Liquor. *Journal of Pharmacy and Nutrition Sciences*. 8. 20-28. 10.6000/1927-5951.2018.08.01.4.
12. Senatore, Felice & Oliviero, Filomena & Scandolera, Elia & Taglialatela-Scafati, Orazio & Roscigno, Graziana & Zaccardelli, Massimo & Falco, Enrica. (2013). Chemical composition, antimicrobial and antioxidant activities of anethole-rich oil from leaves of selected varieties of fennel [*Foeniculum vulgare* Mill. ssp *vulgare* var. *azoricum* (Mill.) Thell]. *Fitoterapia*. 90. 10.1016/j.fitote.2013.07.021.
13. Sfar, Manel & Souid, Ghada & Zeineb, Mzoughi & Le Cerf, Didier & Majdoub, Hatem. (2023). Hepatoprotective Effect Against Cadmium-Induced Liver Toxicity in Rats of *Foeniculum vulgare* Seed Polysaccharides. *Chemistry Africa*. 6. 3. 10.1007/s42250-023-00621-7.
14. Shaffie M.N., Morsy A.F., Ali G.A. and Sharaf A.H. Effect of Caway, Coriander and Fennel on the structure of kidney and islets of Langerhans in alloxan-Induced diabetic rats: Histological and histochemical study, *Researcher*. 2010; 2 (7): 27-40.
15. Shang L, Nienhaus K, Nienhaus GU. Engineered nanoparticles interacting with cells: size matters. *J*



Nanobiotechnol. 2014;12(1):5. doi:10.1186/1477-3155-12-5