J Nanostruct 17(1): 238-244, Winter 2027

RESEARCH PAPER

Evaluation of the Effectiveness of the Nanoparticle Compound of Carpesium Cernuum Leaves on Physiological and Histological Changes in the Brain of Male Albino Rats

Sarah Farouk Majeed Al-Taweel [1]*, ***Heba A. Abd-Alsalam Alsalame*** [2,3]*

[1] *College of Education for Pure Science, Kerbala University, Karbala, Iraq*

[2] *College of Education for Pure Science, Kerbala University, Karbala, Iraq*

[3] *College of Health and Medical Techniques, Al-Zahrawi University, Karbala, Iraq*

ARTICLE INFO

Article History:
Received 07 August 2026
Accepted 25 October 2026
Published 01 January 2027

Keywords:
Carpesium cernuum L.
Green Synthesis
Histopathology
Oxidative Stress
ZnO NPs

ABSTRACT

This study aimed to evaluate the effectiveness of zinc oxide nanoparticles (ZnO NPs) biosynthesized through green method using the alcoholic leaf extract of *Carpesium cernuum L.* in reducing the physiological and histopathological brain alterations induced by N-Methyl-N-nitrosourea (MNU) in male albino rats. ZnO NPs were synthesized using the plant extract and characterized by UV–Visible spectroscopy (UV–Vis), Fourier Transform Infrared Spectroscopy (FTIR), Scanning Electron Microscopy (SEM), and X-ray Diffraction (XRD). The characterization results confirmed the successful green synthesis of stable ZnO NPs with good crystallinity and particle size range from 23.26 to 67.27 nm. The median effective dose (ED_{50}) was determined based on oxidative stress biomarkers and selected biochemical parameters. The neuroprotective action of the fabricated nanoparticles was assessed based on experiments conducted on four groups of animals including controls, MNU-treated, nanoparticle treated, and protective groups. The results indicated that nanoparticle treatment significantly lowered the level of MDA and raised the level of GSH, as well as improved the parameters of the lipid profile in terms of decreased TC and increased HDL, which signifies the attenuation of oxidative stress. Moreover, the nanoparticles decreased MAPK expression and normalized AChE activity, showing improved neuronal functioning. According to histopathology, there were significant reduction in MNU-induced pathologies such as neuronal degeneration, apoptosis, neuronal shrinkage, perineuronal halo formation, and others. Based on immune-histochemical analysis, there was better expression of MARK-1 and BCL-2 proteins than in MNU-treated group. In conclusion, *Carpesium cernuum L.*-assisted synthesized ZnO NPs possessed potent antioxidant and anti-inflammatory activities and provided significant neuroprotective effects against MNU-induced brain damage. These findings suggest that the synthesized NPs represent a promising candidate for future biomedical applications in the prevention and treatment of neurodegenerative disorders.

How to cite this article
Majeed Al-Taweel S., Abd-Alsalam Alsalame H. Evaluation of the Effectiveness of the Nanoparticle Compound of Carpesium Cernuum Leaves on Physiological and Histological Changes in the Brain of Male Albino Rats. J Nanostruct, 2027; 17(1):238-244. DOI: 10.22052/JNS.2027.01.022

* *Corresponding Author Email: sarah.f@s.uokerbala.edu.iq*

This work is licensed under the Creative Commons Attribution 4.0 International License.
To view a copy of this license, visit http://creativecommons.org/licenses/by/4.0/.

INTRODUCTION

The *Carpesium cernuum L*. is an herbal plant that belongs to the genus Carpesium and Asteraceae family. The plant belongs to the plant kingdom, division Streptophyta, and Asterales order. The species is native to wide geographic distribution from Europe to East Asia that includes Japan and Borneo and mainly grows in temperate climates. The genus *Carpesium* includes 24 species distributed in Asia and Eurasia, wherein 18 species are widespread in China and mainly found in the southwestern mountainous areas of the country. The seeds, roots, leaves, and stem of some species from the genus *Carpesium* are traditionally used in Chinese medicine to treat bleeding, showing an anthelmintic, anti-inflammatory, and detoxification therapy [1]. Besides, some species from the genus *Carpesium* are well-known for the production of various bioactive substances such as sesquiterpenes, monoterpenes, diterpenoids, phenolic compounds, and others. The sesquiterpene lactones can be regarded as unique metabolites of the genus and might be studied further as a potential drug source [2]. Plants used as traditional medicine, like *Carpesium cernuum L*., having bioactive molecules exhibit significant antioxidant and anti-inflammatory properties for protecting the cells and their neurological functions. ZnO NPs in nanotechnology have proven to be effective and efficient in increasing the bioavailability of these extracts. This study highlights the impact of green synthesized ZnO NPs by *Carpesium cernuum L.* to adjust the physiological and histopathological brain alterations in male albino rats.

MATERIALS AND METHODS

Male albino rats were randomly divided into control and experimental groups and orally administered different doses of *Carpesium cernuum L*. extract or reference compounds daily for one month. While the MNU injection was accomplished under preton one time weekly. At the end of the experimental period, blood samples were collected after overnight fasting, and serum was separated using centrifugation. Biochemical and molecular analysis were conducted using ELISA and spectroscopy techniques to evaluate the status of various enzymes, hormones, and markers of inflammation. Brain samples were collected and fixed in 10% formalin solution before undergoing standard tissue preparation procedures, which included dehydration, clearing, embedding, slicing, and staining. The stained slides were observed under light microscopy to note the changes. All collected data was subjected to statistical analysis using the SPSS program and LSD test.

RESULTS AND DISCUSSION

Characterization

Identification of the products revealed the successful eco-friendly synthesized ZnO NPs through the ethanoic extract of *Carpesium cernuum L*. Based on the reported XRD analysis, there are definite peaks observed at 2θ angles of 31.82°, 34.46°, 36.26°, 47.54°, 56.62°, 62.94°, and 67.94°.

The FTIR analysis proved the presence of ZnO nanoparticles, with specific absorption peaks. These peaks have been found at 2346 cm^{-1} and 1120 cm^{-1} which represent O=C=O and C–O stretches, respectively. The band at 668 cm^{-1} corresponded to C–H bending and C–O vibrations, while the peak at 403 cm^{-1} indicated Zn–O bonds, confirming the formation of ZnO nanoparticles.

According to the results obtained through the UV-visible spectroscopy, there was an absorption peak detected at 350 nm, which confirms the successful synthesis of ZnO NPs through the ethanolic extract of *Carpesium cernuum L*. The absorption peak detected in the range of 300-380 nm is typical for the synthesis of ZnO NPs and relates to the electronic band gap transitions, which implies the successful synthesis of nanoparticles by plant bioactive compounds [3-5]. The peak detected proves that stable nanoparticles have been produced due to the activity of the extract [6, 7]. SEM analysis further confirmed successful biosynthesis, showing nanoparticles with varied morphologies (spherical, cubic, and irregular) and slight aggregation, which is commonly observed due to high surface energy. Particle size ranged from 23.26 to 67.27 nm, confirming nanoscale formation (1–100 nm) and indicating a high surface area-to-volume ratio that enhances biological activity. Phytochemicals like flavonoids, terpenoids, and phenolics present in *Carpesium cernuum L*. possibly facilitated nanoparticle stability and enhanced the efficacy of the particles's antioxidant and anti-inflammatory properties [1, 8, 9]. This demonstrates that the efficacy of the plant extract as a reducing and stabilizing agent leading to the production of nanoparticles with

(cc) BY

appropriate physicochemical characteristics for biomedical applications. Therefore, in the current study, the nanoformulation prepared through the plant extract was employed to evaluate the neuroprotective effect of the nanoformulation against the brain injury induced by MNU through the regulation of apoptosis, oxidative stress, and inflammation.

Determination of the median effective dose (ED_{50}) of the Carpesium cernuum leaf extract-based nanoparticle

This experiment was conducted to determine the median effective dose (ED_{50}) of the nanoparticle prepared from the extract of *Carpesium cernuum* leaves. A dose–response curve was established using a series of increased doses, while monitoring several biochemical markers, including malondialdehyde (MDA), reduced glutathione (GSH), total cholesterol (TC), and high-density lipoprotein (HDL). The purpose was to identify the most effective dose for use in the evaluating the protective effect of the nanoparticle formulation against MNU-induced brain injury. The results demonstrated clear dose-dependent variations in the investigated biochemical parameters, indicating differences in the biological response to the administered doses. The results of the dose–response experiment demonstrated that increasing doses of the *Carpesium cernuum* nano-extract significantly improved the investigated biochemical parameters. Serum malondialdehyde (MDA) and total cholesterol (TC) levels decreased progressively, whereas reduced glutathione (GSH) and high-density lipoprotein (HDL) levels increased in a dose-dependent manner. It can be observed from the findings that the extract has strong antioxidant activity and hypolipidemic activity through decreasing oxidative stress and increasing antioxidant activity/ lipid metabolism. The dose–response results showed a dose-dependent reduction in MDA, indicating reduced lipid peroxidation and oxidative stress [10, 11]. In contrast, GSH levels increased with higher doses, reflecting enhanced antioxidant defense and improved redox balance [12, 13]. Additionally, TC decreased while HDL increased, suggesting improved lipid metabolism and cellular protection [14, 15]. These biochemical improvements are consistent with the antioxidant and anti-inflammatory properties of plant-derived compounds and biosynthesized nanoparticles, which reduce reactive oxygen species and enhance endogenous defense systems [6, 16]. Overall, the findings demonstrate a clear protective and dose-dependent effect of the nano-extract of *Carpesium cernuum*, supporting the selection of the ED_{50} as the optimal dose for further neuroprotective studies against MNU-induced brain injury [17].

Effect of different treatments on physiological parameters

Table 1 shows that there are considerable differences in MAPK and AChE in terms of the experimental groups, where MNU caused a substantial enhancement in MAPK, while the two protected groups had lower levels than MNU. The activity of AChE had some differences without any drastic changes, which indicates that the nano-extract has a regulating effect only on MAPK but not on AChE. The results demonstrated a significant increase in MAPK levels in the MNU group, indicating activation of oxidative stress, inflammation, and cellular damage pathways [18, 19]. In contrast, the protective group showed a reduction in MAPK levels, suggesting that the nanoparticle extract attenuated cellular injury through its antioxidant and anti-inflammatory properties [9, 20]. Regarding AChE, its activity in the protective group approached that of the control group, indicating partial restoration of neurological function. This is as a result of the

Table 1. Effect of various treatments on the level of MAPK and AChE physiological parameters in various experimental groups.

Mean ± Standard Error (SE)		Group
ACHE	MARK	
0.07 ± 0.165B	0.71 ± 121.614D	Control
0.12 ± 0.268AB	1.31 ± 188.214A	MNU
0.25 ± 0.610 A	1.16 ± 134.917 B	Carpesium cerenuum extract
0.147 ± 0.05 B	134.917 ± 1.16 B	Extract + MNU
0.4387	3.3305	L.S.D 0.05

CC BY

capacity of the nanoparticle extract to lower oxidative stress and sustain cholinergic equilibrium [21, 22]. In all, this indicates that the extract could be neuroprotective in MNU-induced neurotoxicity.

Histopathological results of the secondary marker

Effect of the nano-compound of *Carpesium cernuum* on the immunoexpression level of MARK-1 in the brain tissues of male albino rats exposed to MNU has been listed in Table 2. The control group was associated with the least immunoexpression level of MARK-1 (121.614 ± 0.71; D), representing normal brain tissue structure, free from any toxin or oxidation factors. This denotes the stability of cellular balance, structural integrity of neurons and glia, and lack of inflammation, oxidation, or pathology. Hence, the control group is the physiological standard for the expression of MARK-1 in healthy brain tissues. Any elevation of MARK-1 in any experimental group is regarded as an answer of MNU induced toxicity, while its decline to that of the control group is seen as protection in terms of neuronal preservation, histopathological damages, and oxidation alteration in male albino rats.

Results of the MNU group (BCL-2 immunoexpression)

Table 3 shows mean immunoexpression levels of BCL-2 protein in the brain tissues of male albino rats treated with the nano-compound of *Carpesium cernuum* and MNU. The group MNU had a statistically significant reduction in the immunoexpression of BCL-2 (53.026 ± 0.70; D), representing the minimum value of all experimental groups under $P \leq 0.05$. This illustrated the neurotoxic effect of MNU in relation to the brain due to the suppression of the anti-apoptotic protein BCL-2. It should be noted that the cause of its reduction is mostly due to MNU-induced oxidative stress, DNA damage, and the formation of free radicals, which unbalance the regulation of pro- and anti-apoptosis mechanisms. Thus, neuronal cells become more prone to apoptosis with associated cell degradation and damage. Therefore, these results clearly show the

Table 2. Effect of the nano-compound of Carpesium cernuum on the immunoexpression level of MARK-1 in the brain tissues of male albino rats exposed to MNU.

MARK-1	Group
0.71 ± 121.614 D	Control
1.31 ± 188.214 A	MNU
1.33 ±126.768 C	Carpesium cerenuum Extract
1.16 ±134.917 B	Extract+MNU
3.3305	L.S.D 0.05

Table 3. Mean immunoexpression levels of BCL-2 protein in the brain tissues of male albino rats treated with the nano-compound of Carpesium cernuum and MNU.

BCL-2	Group
0.64 ± 84.391 B	Control
0.70 ± 53.026 D	MNU
1.07 ± 87.210 A	Carpesium cerernuum extract
0.79 ± 77.723 B	Extract+ MNU
2.3604	L.S.D 0.05

CC BY

high neurotoxicity of MNU and can be used as an appropriate experimental model of brain trauma to assess the protective potential of nanoextract of *Carpesium cernuum* in the groups of treatment.

Histological findings of brain tissue

The section displays widespread early signs of cellular necrosis characterised by eosinophilic (pink), finely fibrillary network composed of unmyelinated axons, dendrites, and glial cell depletion. There is noticeable evidence of acute neuronal cellular injury. Several neurons demonstrate classic features of nuclear pyknosis and cytoplasmic hypereosinophilia. This morphology is highly indicative of acute cell death, which can occur via apoptotic or ischemic necrotic pathways (Red arrows). Glial cells as scattered small, round, dark nuclei are visible throughout the neuropil (Green arrows, Fig. 1).

Generally, red neurons with a widespread several pyramidal neurons show classic signs of acute, irreversible cell injury (necrosis). They display angular, shrunken cell bodies, intense cytoplasmic eosinophilia. There is prominent clear space (halos) around multiple cellular elements. In Fig. 2, there is a large vacuole (Black arrow) containing a severely shrunken, degenerated cellular remnant (Red arrow). The degree of cellular shrinkage here points to true edema and cellular regression, Glial cells, scattered throughout the background are normal-appearing glial nuclei (astrocytes with open, vesicular nuclei and oligodendrocytes with small, round, dark nuclei) (Green arrows). There is no significant inflammatory infiltrate (such as

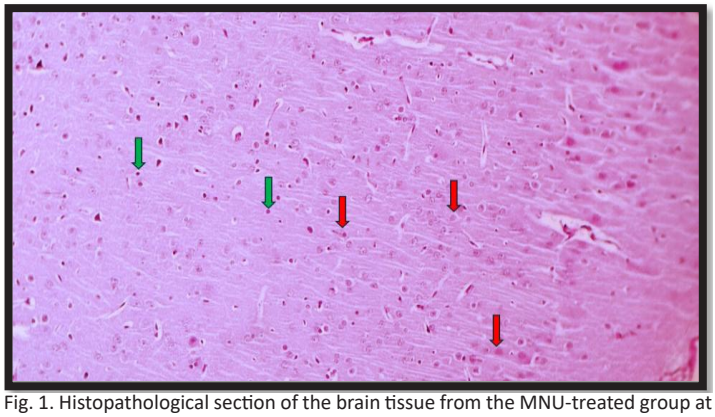


Fig. 1. Histopathological section of the brain tissue from the MNU-treated group at ×100 magnification, showing early neuronal necrosis and glial cells.

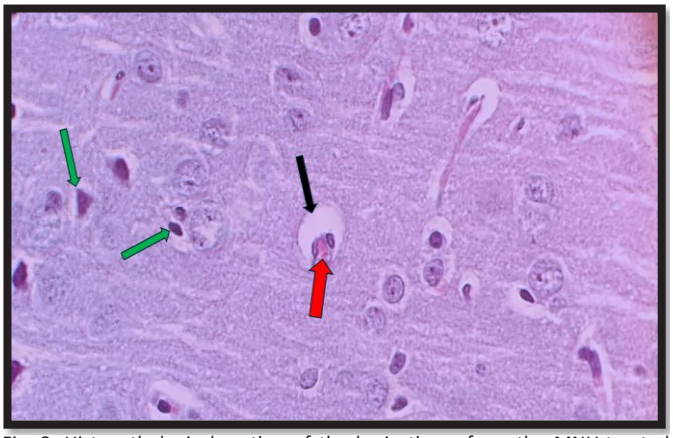


Fig. 2. Histopathological section of the brain tissue from the MNU-treated group at ×400 magnification, showing degenerating (red) neurons, cytoplasmic vacillation, and perineuronal halos.

CC BY

neutrophils or prominent microglial rods) in this field.

In Fig. 3, no large-scale tissue necrosis. The damage is primarily at the individual cell level.

The section shows severe neuronal injury characterized by shrunken pyramidal neurons with angular borders, intense cytoplasmic eosinophilia, and nuclear pyknosis (red neurons). There is also perineuronal halo formation with apoptotic features, signifying acute neurodegeneration, with the neuropil being dense in appearance, and the glial cell layer being intact.

In Fig. 4, no widespread necrosis of tissues. The injury presented here occurs mainly on a cellular scale, as below:

The sample has severe damage of the neurons marked by the presence of pyramidal neurons that are shrunken with sharp borders, hyper-eosinophilia of the cytoplasm and pyknotic nuclei (red neurons) as well as haloing around neurons due to shrinkage of the cells and edema. The apoptotic changes are present, whereas the

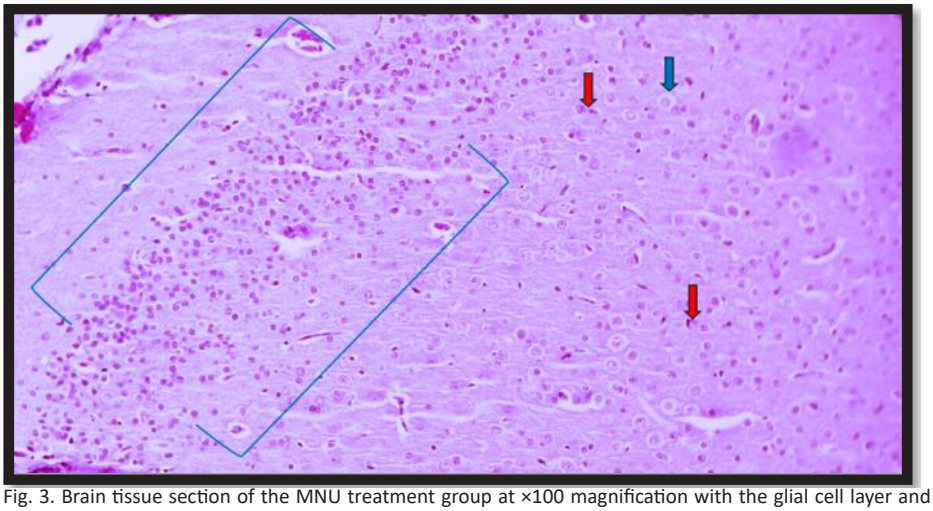


Fig. 3. Brain tissue section of the MNU treatment group at ×100 magnification with the glial cell layer and severely damaged neurons.

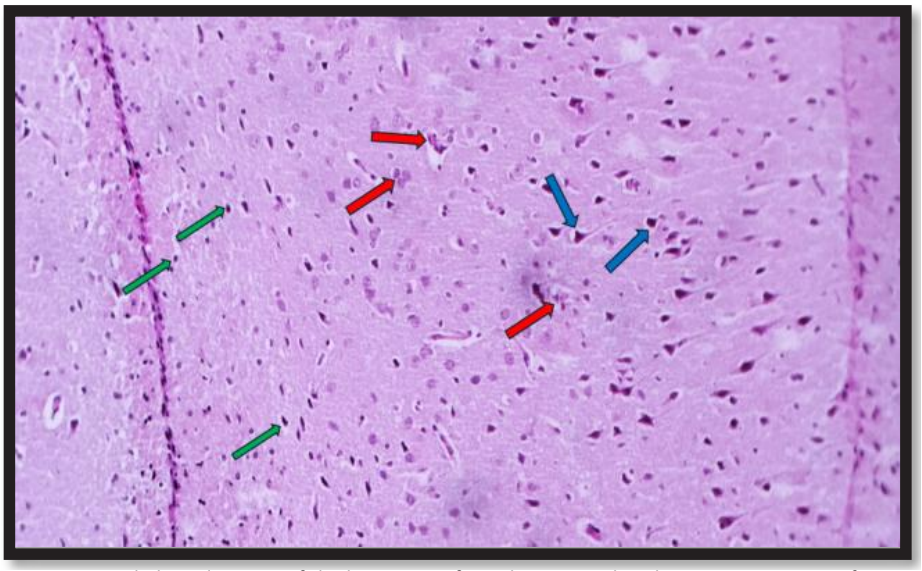


Fig. 4. Histopathological section of the brain tissue from the MNU-induced group at ×100 magnification, demonstrating the layer of microglial cells with neuronal and apoptotic changes.

CC BY

neuropil is dense and the microglia cells are intact.

CONCLUSION

It was evident that ZnO nanoparticles had been successfully prepared through green chemistry using alcoholic extracts of *Carpesium cernuum*, with evidence provided by UV-Visible spectroscopy, SEM, FTIR, and XRD for the stability and crystallization of nanoparticles in nanoscale range. Nano-extracts were characterized by antioxidant activities due to reduction in MDA level, enhancement in GSH level, and improvement in lipid profile marker by reduction in TC and enhancement in HDL, which led to estimation of ED_{50} dose. These nanoparticles also showed positive influence on the physiological functioning of brain due to reduction in MAPK levels and normalization of AChE activity level, reflecting neuroprotective effects. The histological and immunological data revealed reduced neuronal damages and enhanced expression of MARK-1 and BCL-2 in treated groups.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this manuscript.

REFERENCES

1. Shi N-N, Hou C-C, Liu Y, Li K-Y, Mi S-D, Tong B-L, et al. Chemical constituents of plants from the genus Carpesium. Heterocycl Commun. 2022;28(1):95-123.
2. Zhang T, Zhang Q-B, Fu L, Li L-Y, Ma L-Y, Si J-G, et al. New antiproliferative germacranolides from Carpesium divaricatum. RSC Advances. 2019;9(20):11493-11502.
3. Agarwal H, Venkat Kumar S, Rajeshkumar S. A review on green synthesis of zinc oxide nanoparticles – An eco-friendly approach. Resource-Efficient Technologies. 2017;3(4):406-413.
4. Alhujaily M, Albukhaty S, Yusuf M, Mohammed MKA, Sulaiman GM, Al-Karagoly H, et al. Recent Advances in Plant-Mediated Zinc Oxide Nanoparticles with Their Significant Biomedical Properties. Bioengineering. 2022;9(10):541.
5. Hamed R, Obeid RZ, Abu-Huwaij R. Plant mediated-green synthesis of zinc oxide nanoparticles: An insight into biomedical applications. Nanotechnology Reviews. 2023;12(1).
6. Fatima K, Asif M, Farooq U, Gilani SJ, Bin Jumah MN, Ahmed MM. Antioxidant and Anti-inflammatory Applications of Aerva persica Aqueous-Root Extract-Mediated Synthesis of ZnO Nanoparticles. ACS Omega. 2024;9(14):15882-15892.
7. Elhabal S, Abdelaal N, Saeed Al-Zuhairy S, Elrefai M, Elsaid Hamdan A, Khalifa M, et al. Green Synthesis of Zinc Oxide Nanoparticles from Althaea officinalis Flower Extract Coated with Chitosan for Potential Healing Effects on Diabetic Wounds by Inhibiting TNF-α and IL-6/IL-1β Signaling Pathways. International Journal of Nanomedicine. 2024;Volume 19:3045-3070.
8. Dang H, Li H, Ma C, Wang Y, Tian J, Deng L, et al. Identification of Carpesium cernuum extract as a tumor migration inhibitor based on its biological response profiling in breast cancer cells. Phytomedicine. 2019;64:153072.
9. Huang Y, Mao J, Zhang L, Guo H, Yan C, Chen M. Incaspitolide A isolated from Carpesium cernuum L. inhibits the growth of prostate cancer cells and induces apoptosis via regulation of the PI3K/Akt/xIAP pathway. Oncol Lett. 2021;21(6).
10. Chen B, Dong X, Zhang J, Wang W, Song Y, Sun X, et al. Effects of oxidative stress regulation in inflammation-associated gastric cancer progression treated using traditional Chinese medicines: A review. Medicine. 2023;102(46):e36157.
11. Sies H, Jones DP. Reactive oxygen species (ROS) as pleiotropic physiological signalling agents. Nature Reviews Molecular Cell Biology. 2020;21(7):363-383.
12. Liu J, Huang Z, Yin S, Jiang Y, Shao L. Protective effect of zinc oxide nanoparticles on spinal cord injury. Front Pharmacol. 2022;13.
13. Gultekin M, Eren M, Koca FD, Bozbek CK, Develi N. The effects of zinc oxide nanoparticles on the oxidative stress, caspase-3, cytokine and immunity in rats. Vet Med (Praha). 2025;70(10):379-392.
14. Gunes-Bayir A, Guler EM, Bilgin MG, Ergun IS, Kocyigit A, Dadak A. Anti-Inflammatory and Antioxidant Effects of Carvacrol on N-Methyl-N′-Nitro-N-Nitrosoguanidine (MNNG) Induced Gastric Carcinogenesis in Wistar Rats. Nutrients. 2022;14(14):2848.
15. Yang J, Xiong D, Long M. Zinc Oxide Nanoparticles as Next-Generation Feed Additives: Bridging Antimicrobial Efficacy, Growth Promotion, and Sustainable Strategies in Animal Nutrition. Nanomaterials. 2025;15(13):1030.
16. Forman HJ, Zhang H. Targeting oxidative stress in disease: promise and limitations of antioxidant therapy. Nature Reviews Drug Discovery. 2021;20(9):689-709.
17. Zou X, Watanabe Y, Ozawa S, Ebizuka Y, Shobudani M, Sakamaki Y, et al. Gene expression profiles of neuroinflammatory responses in broad brain regions in rats repeatedly administered with N -methyl- N -nitrosourea for 28 days. The Journal of Toxicological Sciences. 2024;49(11):481-495.
18. Behl T, Rana T, Alotaibi GH, Shamsuzzaman M, Naqvi M, Sehgal A, et al. Polyphenols inhibiting MAPK signalling pathway mediated oxidative stress and inflammation in depression. Biomedicine and Pharmacotherapy. 2022;146:112545.
19. Gao Z-K, Shen X-Y, Han Y, Guo Y-S, Li K, Bi X. Pre-ischemic exercise prevents inflammation and apoptosis by inhibiting MAPK pathway in ischemic stroke. Transl Neurosci. 2022;13(1):495-505.
20. Park Y-J, Cheon S-Y, Lee D-S, Cominguez DC, Zhang Z, Lee S, et al. Anti-Inflammatory and Antioxidant Effects of Carpesium cernuum L. Methanolic Extract in LPS-Stimulated RAW 264.7 Macrophages. Mediators Inflamm. 2020;2020:1-14.
21. Tang Kim S, See Wesley Zhi C, Naidu R. Neuroprotective properties of zinc oxide nanoparticles: therapeutic implications for Parkinson's disease. Biosci Rep. 2024;44(11).
22. Goma AA, Salama AR, Tohamy HG, Rashed RR, Shukry M, El-Kazaz SE. Examining the Influence of Zinc Oxide Nanoparticles and Bulk Zinc Oxide on Rat Brain Functions: a Comprehensive Neurobehavioral, Antioxidant, Gene Expression, and Histopathological Investigation. Biol Trace Elem Res. 2024;202(10):4654-4673.

CC BY