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Cytotoxicity of Zinc Oxide Nanoparticles: Recent Evidences and Biological Implications

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Abstract

Zinc oxide nanoparticles (ZnO-NPs) are being widely studied for biological applications due to their adjustable physicochemical characteristics and possible antibacterial and anticancer effects. However, variable toxicity results and unsolved biosafety issues preclude their clinical use. The evidence suggests that particle size, surface properties, synthesis technique, exposure dose, and biological model all have an impact on ZnO-NP cytotoxicity. This mini-review covers current in vitro and in vivo investigations, showing dose-dependent effects and selective anticancer action. Key mechanisms include oxidative stress, Zn^+ ion release, and mitochondrial malfunction.

Introduction

ZnO-NPs have attracted substantial attention because of their distinctive thermal, optical, magnetic, and biological properties. These characteristics support their application in cancer therapy, biomedical imaging, diagnostics, nanomedicine, and antimicrobial protective coatings [1, 2]. However, their expanding use raises concerns regarding unintended exposure routes, particularly ingestion and inhalation, followed by systemic distribution within the body [3]. Experimental data suggests that ZnO-NPs can bioaccumulate in important organs, disrupting zinc homeostasis, altering enzyme function, and inducing oxidative stress [4-7]. ROS production is primarily thought to play a major role in ZnO-NP-mediated cytotoxicity, which happens in many types of mammalian cells by generating membrane damage, inflammation, DNA damage, apoptosis, and endocrine disruption [8,9]. Long-term or high-dose exposure has been linked to problems associated with oxidative stress, including respiratory, cardiovascular, and neurological diseases [10]. Despite their immense therapeutic potential, ZnO-NPs would need to be carefully optimized for safety. This mini-review covers current developments in ZnO-NP cytotoxicity investigations using cellular and animal models.

Recent Advances in ZnO-NP Cytotoxicity Studies

Cytotoxicity in normal human cells: Several researches have looked at the impact of ZnO-NPs on normal human cells. ZnO-coated dental aligners were tested for cytotoxicity against Hu-

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man Gingival Fibroblasts (HGFs) using MTT assays at 0, 7, and 14 days. After lengthy exposure (days 7 and 14), the coated aligners revealed mild cytotoxicity, but the uncoated aligners did not show any. Importantly, increasing nanoparticle size resulted in lower cytotoxic effects, suggesting that particle size serves a protective function in reducing cellular These results indicate that ZnO-coated biomedical devices should be used with caution, particularly under conditions of long-term exposure [11]. In a similar study, Peripheral Blood Mononuclear Cells (PBMCs) were exposed to ZnO-NPs generated using the Pechini technique, resulting in a significant decrease in cell viability, increased LDH release, and apoptosis at high doses. Although no micronucleus development was seen, substantial evidence of mitochondrial dysfunction and membrane damage was described, suggesting cellular absorption of ZnO-NPs or Zn^{2+} ions through endocytic pathways [12].

Cytotoxicity in selective anticancer activity: Green synthesis methods have shown promising anticancer selectivity with little toxicity to normal cells. ZnO-NPs produced from Pentatropis capensis leaf extract showed cytotoxicity against HT-29 colon cancer cells with an IC_{50} of 95.37 µg/mL, which is comparable with previous results utilizing Morus laevigata extracts [13-15]. Similarly, ZnO-NPs made using H. scoparia extract displayed dose-dependent cytotoxicity against MCF-7 breast cancer cells. Un-calcined nanoparticles reduced cell viability by more than 50% at doses below their IC_{50} [16]. Green-synthesized ZnO-NPs utilizing Catharanthus pusillus showed substantial cytotoxic effects against A549 lung cancer cells (IC_{50} = 36.63 µg/mL), triggering death through mitochondrial breakdown and oxidative stress while sparing normal cells. Morphological alterations such as membrane blebbing and apoptotic body formation supported programmed cell death [17].

Another study found that ZnO-NPs generated with Fioria vitifolia leaf extract and stabilized with poly vinyl pyrrolidone were more stable and selectively cytotoxic to A549 lung cancer cells, while being less hazardous to L929 fibroblasts. The obtained IC_{50} values (86.6-93.6 µg/mL) indicate their selective anticancer potential. This increased activity has been linked to bioactive phytochemicals such as anthraquinones and saponins, which may influence oxidative stress pathways and cause apoptotic cell death [18]. Similarly, aluminum-doped ZnO-NPs showed improved lethal selectivity against MDA-MB-231 breast cancer cells while having negligible impact on normal Mammary Epithelial Cells (MCF-10A). Higher dopant levels increased the formation of reactive oxygen species and mitochondrial-mediated cell death in cancer cells, suggesting that dopant-driven redox regulation may offer a technique for targeted cancer treatment [19].

In vivo and ecotoxicological evidence: In vivo investigations consistently demonstrate ZnO-NPs' dose-dependent toxicity. Exposing marine medaka (Oryzias melastigma) to ZnO-NPs at concentrations ≥10 mg/L caused considerable mortality and suppressed genes linked to neurological and cardiovascular function. Although heart rate did not change much, extended exposure raised concerns about environmental danger [20]. In animal models, oral treatment of ZnO-NPs (40 and 70 nm) for 50 days resulted in testicular toxicity, decreased sperm count and motility, hormonal abnormalities, inflammation, and oxidative stress. Nanoparticles caused higher toxicity than bulk ZnO, with bigger particles having more significant negative effects [21].

Mechanistic Insights into ZnO-NP Cytotoxicity: ZnO-NP-induced cytotoxicity is primarily caused by reactive oxygen species production, Zn^{2+} ion release, mitochondrial malfunction, membrane damage, and apoptosis [12,17,19,21]. The toxicity output is largely controlled by aspects such as size, surface, synthesis method, and dopants used during synthesis [11,18,19]. It is noteworthy that green methods in almost all instances reduce non-target toxicity without compromising or even improving anti-cancer potency [14,17,18]. As a result, green synthesis methodologies are regarded as crucial for rationally designing and formulating ZnO-based nanoparticles for a variety of biological applications.

Conclusions and Future Perspectives

In conclusion, as evident from the recent studies data, ZnO-NPs reveal considerable dose-response, size-response, and model-response cytotoxicity, with a growing number of reports confirming their selective anticancer efficacy when rationally designed using "green synthesis," functionalization, or doping. However, the current data were obtained in short-term in vitro models, which may be inadequate to be taken into account in the complex in vivo conditions characterized by degradation characteristics, ion release, and accumulation patterns influencing the NPs' toxicity profile. Therefore, it is necessary to develop a new toxicological approach with a clear differentiation between particle- and ion-related phenomena in ZnO-NPs and to conduct in vivo biosafety and reproductive toxicity studies. It is crucial to optimize the surface chemistry of ZnO-NPs to minimize non-specific toxicity as well as to maintain efficacy to ensure the safe application of these NPs in a wide range of biological targets, including nanomedicine.

Conflict of Interest: The authors report no conflicts of interest in this work.

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References

1. S Dey, D lochan Mohanty, N Divya, V Bakshi, A Mohanty, et al. A critical review on zinc oxide nanoparticles: Synthesis, properties and biomedical applications, Intelligent Pharmacy. 2025; 3(1): 53-70.

2. A Ragu Prasath, K Selvam. A review of the zinc oxide nanoparticles synthesis and their emerging biomedical potential, Biomedical Materials & Devices. 2025: 1-25.

3. L He, Y Liu, A Mustapha, M Lin. Antifungal activity of zinc oxide nanoparticles against Botrytis cinerea and Penicillium expansum, Microbiological research. 2011; 166(3): 207-215.

4. A R Pinho, F Martins, M E V Costa, A M R Senos, O A B da Cruz e Silva, et al. In vitro cytotoxicity effects of zinc oxide nanoparticles on spermatogonia cells, Cells. 2020; 9(5): 1081.

5. M Ghosh, S Sinha, M Jothiramajayam, A Jana, A Nag, et al. Cytogenotoxicity and oxidative stress induced by zinc oxide nanoparticle in human lymphocyte cells in vitro and Swiss albino male mice in vivo, Food and Chemical Toxicology. 2016; 97: 286-296.

6. J Musarrat, Q Saquib, A Azam, S A H Naqvi, Zinc oxide nanoparticles-induced DNA damage in human lymphocytes, International Journal of Nanoparticles. 2009; 2(1-6): 402-415.

7. C Mihai, W B Chrisler, Y Xie, D Hu, C J Szymanski, et al. Intracellular accumulation dynamics and fate of zinc ions in alveolar epithelial cells exposed to airborne ZnO nanoparticles at the air-liquid interface, Nanotoxicology. 2015; 9(1): 9-22.

8. S R Saptarshi, A Duschl, A L Lopata. Biological reactivity of zinc oxide nanoparticles with mammalian test systems: An overview, Nanomedicine. 2015; 10(13): 2075-2092.

9. J Zhao, L Xu, T Zhang, G Ren, Z Yang. Influences of nanoparticle zinc oxide on acutely isolated rat hippocampal CA3 pyramidal neurons, Neurotoxicology. 2009; 30(2): 220-230.

10. A Beegam, P Prasad, J Jose, M Oliveira, F G Costa. Environmental Fate of Zinc Oxide Nanoparticles: Risks, Toxicology: New aspects to this scientific conundrum. 2016; 81.

11. I Ravi, V Kailasam. Assessment of cytotoxicity of clear aligners coated with zinc oxide nanoparticles, Journal of Oral Biology and Craniofacial Research. 2025; 15(2): 262-265.

12. J V R Angulo, J F Valenzuela, S I Freire-Bernal, V E Nino-Castano, J E R Paez, et al. Cytotoxicity and genotoxicity of zinc oxide nanoparticles in human peripheral blood mononuclear cells, Mutation Research-Genetic Toxicology and Environmental Mutagenesis. 2025; 901: 503838.

13. S Paranthaman, C S Shivakumar, S Kalaipriya, H N Venkatesh, J Gireesha, et al. One-pot green synthesis of zinc oxide nanoparticles using Morus laevigata aqueous extract and evaluation of its anticancer potential against HT-29 cell line, Main Group Metal Chemistry. 2024; 47(1): 20240016.

14. L Ganesan, M Arumugam, V Maluventhen. Green Synthesis of Zinc Oxide Nanoparticles Using Pentatropis capensis (Lf) Bullock Leaf Extract: Characterization, Antibacterial, Antioxidant, Anti-Inflammatory and Cytotoxicity Studies, Biomedical Materials & Devices. 2026; 4(1): 896-908.

15. D Elumalai, T Y Suman, M Hemavathi, C Swetha, R Kavitha, et al. Biofabrication of gold nanoparticles using Ganoderma lucidum and their cytotoxicity against human colon cancer cell line (HT-29), Bulletin of Materials Science. 2021; 44(2): 132.

16. C Benine, D A Boutlelis, L A Touhami, E Lanez, D G Amara, et al. Green synthesis, characterization, antioxidant, interaction with DNA/BSA, and investigation of cytotoxicity against MCF-7 cancer cells of zinc oxide nanoparticles using Hammada scoparia (Pomel) Iljin extract, International Journal of Biological Macromolecules. 2025; 295: 139709.

17. Y S Wu, E S Sam, A R Prasath, K Selvam, V Sangameshwaran, et al. Green synthesis of zinc oxide nanoparticles using Catharanthus pusillus: Characterization, antibacterial, cytotoxicity of A549 cells, and photocatalytic degradation of Eosin (E) dye, Journal of Water Process Engineering. 2025; 77: 108606.

18. A Nandhini, P Anilkumar, J Jasmin, S Balamurali. Green synthesis, characterization, structural, morphological, antibacterial, and cytotoxicity evaluation of zinc oxide nanoparticles using Fioria vitifolia extract, Biophysical Chemistry. 2025; 323: 107440.

19. A Khorsand Zak, A M Hashim, H Khorsand Zak. Investigating the cytotoxicity of Aluminum-Doped zinc oxide nanoparticles in normal versus cancerous breast cells, Scientific Reports. 2025; 15(1): 25227.

20. S Shohag, Y Horie. Neurotoxicity and cardiovascular toxicity of zinc oxide nanoparticles to Oryzias melastigma, Journal of Applied Toxicology. 2025; 45(3): 452-459.

21. D H Ahmed, N M El-Beih, E A El-Hussieny, W M El-Sayed. Zinc oxide nanoparticles induced testicular toxicity through inflammation and reducing testosterone and cell viability in adult male rats, Biological Trace Element Research. 2025; 203(4): 1934-1948.