Green-Synthesized Titanium Dioxide Nanoparticles: Biocompatibility in *Drosophila* and Antibacterial Efficacy

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Keywords: (up to 4, Arial font, size 9, italic style, separated by comma)

ABSTRACT

Metal oxide nanoparticles (NPs), such as titanium dioxide (TiO_2), are increasingly studied for their potential in medical therapies, including photodynamic therapy. As an inorganic compound, TiO_2 is photoactive and can generate reactive oxygen species (ROS) when exposed to UV light in an aqueous solution, inducing cell death. This property makes TiO_2 a promising candidate for treating diseases and combating antibiotic-resistant bacteria. However, concerns regarding its biocompatibility remain. To mitigate environmental impacts and enhance safety, green synthesis of TiO_2 using plant sources has emerged as a sustainable and effective approach for NP production. The aim was to evaluate the biocompatibility of biosynthesized TiO_2-NPs derived from the bark of *Stryphnodendron adstringens* (TiO_2-SA) and *Origanum vulgare* (TiO_2-OV) in the model organism *Drosophila melanogaster* (fruit fly). The TiO_2-NPs were synthesized and mixed with a standard culture medium at final concentrations of 0.125, 0.250, and 0.500 mg/ml. Six vials were used per test concentration, and a negative control containing only the culture medium was also included. In each vial, thirty (n=30) first-instar larvae were introduced, and daily monitoring of larval development, pupation rates, and lethality was conducted. In addition to biosynthesized TiO_2-SA, TiO_2 nanoparticles derived from TiO_2-OV were also tested, with a TiO_2-anatase sample used as a positive control. Moreover, strains of *Staphylococcus aureus* (ATCC 25923) and *Escherichia coli* (ATCC 25922) were tested to evaluate the antimicrobial activity of the NPs using the disk diffusion technique. Both TiO_2-SA and TiO_2-OV demonstrated high biocompatibility compared to TiO_2-anatase, with developmental parameters such as larval lethality and total pupation rate showing results similar to the negative control group. This suggests that biosynthesized TiO_2-NPs are less cytotoxic than the TiO_2-anatase control, supporting their potential suitability for biological applications. Surprisingly, daily pupation analysis of TiO_2-SA revealed a higher pupation rate in treated groups compared to controls over time, suggesting that these NPs may induce larvae to accelerate pupation as an adaptive response. This accelerated pupation may minimize exposure to adverse conditions, highlighting the complex interaction between biosynthesized TiO_2-SA and *Drosophila* development. This response underscores the need to assess not only lethality but also developmental timing and behavioral changes in biocompatibility studies. Despite their observed biocompatibility, TiO_2-OV and TiO_2-SA did not exhibit bactericidal activity against the tested strains of *S. aureus* and *E. coli* in disk diffusion tests, indicating limited antimicrobial efficacy under these conditions. In summary, biosynthesized TiO_2-SA and TiO_2-OV nanoparticles demonstrated strong biocompatibility in *D. melanogaster*, with no significant effects on development or lethality. However, their lack of antibacterial activity limits their application in antimicrobial therapies. Future research should focus on optimizing synthesis methods, incorporating dopants to improve antibacterial efficacy, and examining a wider range of concentrations and conditions. Additionally, studies on long-term and multi-generational impacts on development and behavior are needed to thoroughly assess the safety profile of these nanoparticles for biomedical and environmental applications.

Keywords: nanotoxicity, fruit fly, post-embryonic development, green synthesis