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The Effect of TiO_2 Nanoparticles on Zebrafish Embryos

Authors : Elena Maria Scalisi

Abstract : Currently, photodegradation by nanoparticles (NPs) is a common solution for wastewater treatment. Nanoparticles are efficient for removing organic and inorganic pollutants, heavy metals from wastewater and killing microorganisms through environmentally friendly. In this context, the major representative of photocatalytic technology for industrial wastewater treatment are TiO_2 nanoparticles (TiO_2-NPs). TiO_2-NPs have a strong catalytic activity that depends to their physicochemical properties. Thanks to their small size (between 1-100 nm), nanoparticles occupy less volume, then their surface area increases. The increase in the surface-to-volume ratio results in the increase of the particle surface energy, which improve their reactivity potential. However, these unique properties represent risks to the ecosystems and organisms when unintentionally TiO_2-NPs are release into the environment and absorbed by living organisms. Several studies confirm that there is a high level of interest concerning the safety of TiO_2-NPs in the aquatic environment, furthermore, ecotoxicological tools are useful to correctly evaluate their toxicity. In the current study, we aimed to characterize potential toxic effects of TiO_2-NP suspension to zebrafish during embryo-larval stages to evaluate parameters such as survival rates, malformation, hatching, the overall length of the larvae heartbeat, and biochemical biomarkers that reflect the acute toxicity and sublethal effects of TiO_2-NPs. Zebrafish embryos were exposed to titanium dioxide nanoparticles (TiO_2-NPs at 1mg/L, 2mg/L, and 4mg/L) from fertilization to the free swimming stage (144hpf). Every day, we recorded the toxicological endpoints, moreover, immunohistochemical analysis has been performed at the end of the exposure. In particular, we have evaluate the expression of the following biomarkers: Heat Shock Protein 70 (HSP70), Poly ADP-Ribose Polymerase-1 (PARP-1), Metallothioneins (MTs). Our results have shown that hatch ability, survival, and malformation rate were not affected by TiO_2 NPs at these exposure levels. However, TiO_2-NPs caused an increase of heartbeat and reduction of body length; at the same time, TiO_2-NPs have inducted the production of ROS and the expression of oxidative stress biomarkers HSP70 and PARP-1. Hight positivity for PARP-1 at all concentration tested was observed. As regards MT, positivity was found in the expression of this biomarker in the whole body of the embryo, with the exception of the end of the tail. Metallothioneins (MT) are biomarkers widely used in environmental monitoring programs for aquatic creatures. At the light of our results i.e. no death until the end of the experiment (144hpf), no malformation and expression of the biomarkers mentioned, it is evident that zebrafish larvae with their natural detoxification pathways are able to resist the presence of toxic substances and then they can tolerate the presence of metal concentrations. However, an excessive oxidative state can compromise cell function, therefore the uncontrolled release of nanoparticles into the environment is severe and must be constantly monitored.

Keywords : nanoparticles, embryo zebrafish, HSP70, PARP-1
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