Developmental abnormalities in sea urchin larvae obtained from sperms exposed to engineered nanoparticles

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Engineered nanoparticles are increasingly produced to be employed in many commercial products of common use, but their possible toxic effects are not known enough. Preliminary studies report that they can play a role in damaging numerous important biological processes, including skeletogenesis on living organisms. That is very critical in vertebrates, especially during the developmental stages, when the skeleton develops in a genetically programmed way.

The aim of this study is to propose a model of nanotoxicity for aquatic organisms at the developmental stage. The research investigated whether the exposure to different concentrations of cobalt (Co), titanium dioxide (TiO_2) and silver (Ag) nanoparticles can induce skeletal damages in sea urchins (*Paracentrotus lividus)* at the pluteus stage.

P. lividus at pluteus stages were obtained from fertilized eggs exposing sperms at different nanoparticles' concentrations (from 0.0001 up to 1 μg mL^{-1}). Non exposed sea urchins' sperms served as controls. The pluteus were made permeable by treating them with glycerol solution and then they were treated *in toto* with the lectin wheat germ agglutinin (WGA), specific component for binding to the n-acetyl-glucosamine residues. WGA was fluorochrome conjugated, therefore, it was introduced into tissues and visualized through confocal (TCS SP2 Leica, Switzerland) and epifluorescence (Olympus BX60,Japan) microscopic investigations. Fluorescent WGA lectin-binding sites were observed in the mesenchyme cells and in the skeletal rods of nanoparticle-treated pluteus stages. On the contrary, no fluorescence was detected in the controls at pluteus stages. Morphological investigations of the pluteus by means of a Field Emission Gun Environmental Scanning Electron Microscope (FEG-ESEM Quanta 250–FEI, the Netherlands) coupled with an x-ray microprobe of an Energy Dispersion System verified morphological changes and the physical presence of the different nanoparticles in the bodies. No dose-dependent nanoparticle's effects were identified. Morphological anomalies such as the asymmetrical rods of the skeleton and the irregular shape of the pluteus stage were found only in the exposed pluteus.

In conclusion, the present study suggests that nanoparticles interact with WGA lectin-binding sites in developing mesenchyme and skeleton, causing skeletal alterations and damages. That phenomenon induces malformations in the offspring.

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