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Visualisation of the attachment, possible uptake and distribution of ferric based technical nanoparticles on small test organisms with electron microscopy methods

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Ferric based technical nanoparticles are beginning to be used in a wide variety of products. They have great potential in cleaning of halogen-organic contaminated ground and waste water (project Fe-NANOSIT; funded by the German Federal Ministry of Education and Research [BMBF]). But there is little known yet about the possible impacts of released nanoparticles on the environment and their organisms. On water born organisms like zebra fish, potential toxic effects should be investigated. This research also should help to understand how the particles interact with organism, organs, tissues, cells and cell organelles and whether they have negative effects on general vital functions. In order to identify these particles a simple morphological analysis with conventional electron microscopic methods is not always sufficient [1] due to the possible risks of misinterpretation. Therefore, an additional reliable identification method is necessary. One technique which allows a secure identification of nanoparticles is elemental analysis with energy dispersive X-ray spectroscopy (EDX) performed in the scanning electron microscope (SEM) [2]. In this work we demonstrate a proper identification of ferric nanoparticles both attached at zebra fish egg shells (not shown here) and larvae (Figure 1, A-F) after *in vitro* exposure of test organisms with ferric nanoparticle containing products. Further SEM, TEM and STEM investigations should also contribute to identify possible uptake (i. e. oral uptake) and distribution (i. e. gastrointestinal tract) of ferric nanoparticles within the organisms.

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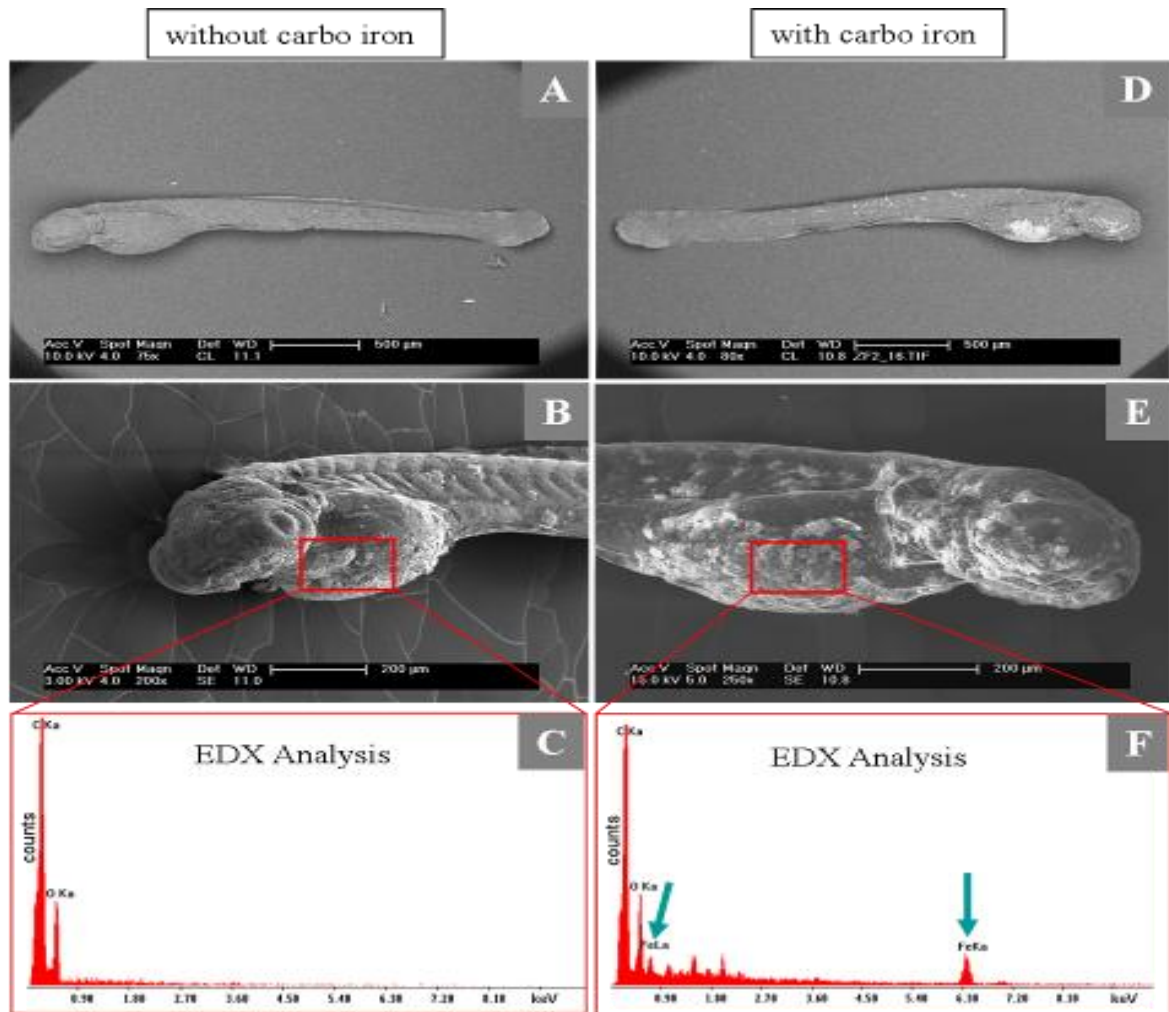


Figure 1: Zebra fish embryos (96 hours old) incubated without (Figure A – C) and with (Figure D – F) carbo iron.

a.) Back scatter electron (BSE) image of zebra fish embryo incubated without carbo iron.

d.) BSE image of zebra fish embryo incubated with carbo iron; Different BSE signal intensity on discontinuous distributed surface areas (different “material contrast”).

b.) Secondary electron (SE) image of zebra fish embryo incubated without carbo iron; plaque like depositions on the surface area (breast and head region) visible.

e.) SE image of zebra fish embryo incubated with carbo iron; also plaque like depositions on the surface area (breast and head region) visible.

c.) EDX analysis (red rectangle in B): shows only peaks for C and O.

f.) EDX analysis (red rectangle in E): besides the peaks for C and O there are also peaks for Fe (arrows) with is the active component of the carbo iron.