Impact of Nanomaterials on Human Health: Lessons from in vitro and animal models

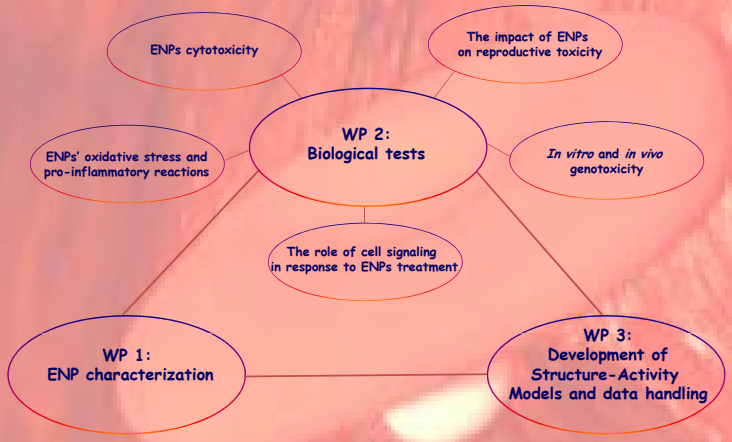
Zuzana Magdolenova[1], Maria Dusinska[1], Lise Fjellsbø[1], Per Schwarze[2], Gunnar Brunborg[2], Maria Wojewódzka[3], Małgorzata Dobrzyńska[4], Joanna Gromadzka[5], Anna Lankoff[3], Christine Instanes[2], Magne Refsnes[2], Marit Låg[2], Elise Rundén- Pran[1], Marcin Kruszewski[3]

[1]Norwegian Institute for Air Research, Kjeller, Norway
[2]Norwegian Institute of Public Health, Oslo, Norway
[3]Institute of Nuclear Chemistry and Technology, Warsaw, Poland
[4]National Institute of Public Health – National Institute of Hygiene, Warsaw, Poland
[5]Warsaw University of Life Science, Warsaw, Poland

Aim

- To elucidate the understanding of the nature of biological responses induced by engineered nanoparticles (ENPs), at different levels of biological organization (i.e. from molecules to whole organism), from which the potential risks to the human health can be determined through observation, modeling and data interpolation ultimately developing a hazard and risk assessment framework.
- Study the molecular and cellular mechanisms, pathways of action of ENPs with specific focus on oxidative stress, inflammation, genotoxicity and reproductive toxicity.

Scheme of work



Methodology

- A panel of biological methods used include determination of ENPs **cytotoxicity** by standard colorimetric assays: MTT or NRR. The LC_{50} will be estimated for each ENP. ENPs toxicity the mechanisms of cell death (apoptosis vs. necrosis) will be assessed by flow cytometry, fluorescence microscopy and Western or ELISA analysis.
- The **oxidative damage** to proteins and lipids will be assessed by immunological (ELISA) and colorimetrical (TCA, TBARs and LPO) methods. The **pro-inflammatory reactions** and oxidative stress responses will be determined using ELISA.
- Determination of ENP-induced changes in gene expression, especially **signaling** pathways will be tested using The Signal Transduction PathwayFinder™ PCR Array (SuperArray, USA), Western blot analysis.
- The ***in vitro and in vivo genotoxic potential*** of ENPs will be measured using the Comet assay with lesion specific enzymes, DNA repair, histone γH2AX foci formation, the micronucleus assay, and a cytokinesis block assay. Apoptosis/necrosis and *in vitro* proliferation rates will also be scored.
- Impact of ENP on **reproductive toxicity** (chronic and acute) will be determined *in vitro* and *in vivo*.

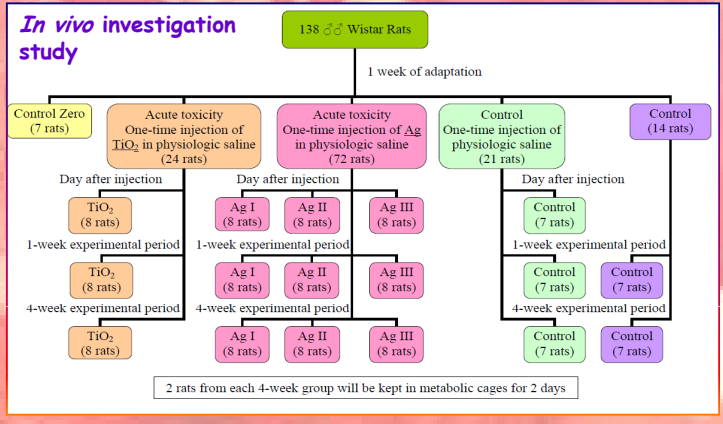
ENP characterization

- Physico-chemical properties of the selected ENPs (TiO_2, Ag), interaction with culture media components, size distributions in stock solution/dispersion and after their addition to culture media, will be determined prior to toxicological testing.

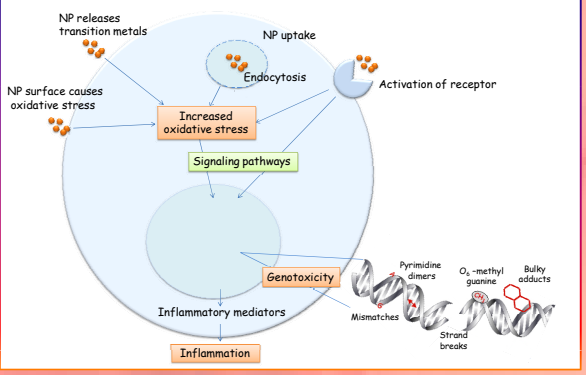
Cellular models *in vitro*

- The blood system: peripheral blood mononuclear cells
- Lung system: human A549, BEAS-2B, HIVE-26, a co-culture of epithelial and endothelial lung cells (for the study of intercellular signaling), and in selected experiments primary rat epithelial lung cells
- The central nervous system: HCEC, SH-SY5Y
- Cardiovascular system: HUVEC, HL1
- The liver: human HepG2
- Isolated testicle cells (*in vitro* experiments), sperm (*in vivo* experiments)

In vivo investigation study



Mechanism of nanoparticles action



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