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## Determination of nanoparticle surface area for improved nanotoxicology studies

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With the increasing amount of products and applications found in our everyday life based on engineered nanoparticles (Geiser & Kreyling, 2010) the concerns about possible adverse health effects of nanoparticles are being discussed intensively. Not only should the final product be safe to use for consumers, but also the exposure of the product to the environment (e.g. when consumers wash off sunscreens or cosmetics containing nanoparticles) should preferably be harmless. Moreover, the handling of particles during fabrication of the products should be carried out in a safe way. A potential risk during product fabrication and handling is inhalation of the nanoparticles and hence a number of both *in vivo* and *in vitro* studies to investigate nanoparticle toxicity for this exposure route have been performed.

For nanotoxicology investigations of air-borne particles to provide relevant results and to enable categorical studies of nanotoxicity it is ever so important that the particle exposure of, for example cells, closely resembles the “real” exposure situation, that the dosimetry is well defined, and that the characteristics of the deposited nanoparticles are known in detail (Grass *et al.*, 2010). By synthesizing the particles in the gas-phase and directly depositing them onto lung cells, the particle deposition conditions in the lung is closely mimicked. In the present work we present a setup for generation of gas-borne nanoparticles of a variety of different materials (Messing *et al.*, 2009) with highly controlled and tunable particle characteristics, and demonstrate the method by both *in vitro* and *in vivo* depositions of gold particles.

The particle characteristics identified to play the most important roll for the toxicological response include shape, mass, number concentration, solubility, surface chemistry and surface area (Maynard & Kuempel, 2005). In our setup the number and mass concentrations are measured on-line, the shape is determined by electron microscopy and the surface area is determined using three different models. The mass and surface area dose is, furthermore, calculated and from these calculations accurate deposition times to cause an inflammatory response during exposure, according to literature, are determined (Messing *et al.*, 2013; Svensson *et al.*, 2013). The generated particles are then deposited onto primary lung cells using an air-liquid-interface (ALI) chamber as well as directly inhaled by living

mice. The first major advantage of the setup used is that it allows for a comparison of the dependence of the toxicological response on surface area, when using particles of the same mass and number concentration. The second major advantage is the simplicity by which particles of different materials are produced that provides the possibility to directly compare the effect of nanoparticle material on toxicology, when all other size related parameters are chosen to be the same.

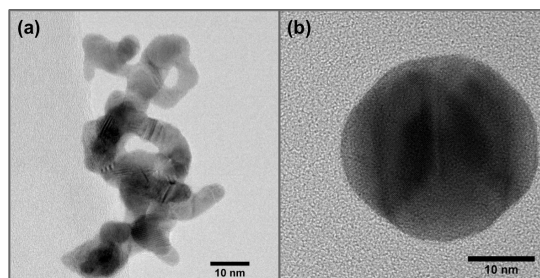


Figure 1. TEM images of (a) an agglomerate particle with a mobility diameter of 60 nm and (b) a particle with an original mobility diameter of 60 nm (compare to a) that has been sintered at 700C which reduced the mobility diameter to 31 nm and the surface area by about a factor of 6.

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