Reference particles for toxicology studies of biomass combustion generated ash particles

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The health risks of biomass combustion have been widely recognized (Mills *et al.*, 2009). However there is contradictory understanding of the factors that cause the adverse effects on human health. There is also a lack of knowledge regarding the relevant dose of particular pollutants causing the adverse health effects (Pope & Dockery, 2006).

In order to understand the toxicological responses, the properties of the exposing particles must be clearly defined. Pure substances or chemicals have been used in toxicological analyses as reference. However the size, crystallinity and solubility, for example, are major determinant factors for the toxicological response (Becker *et al.*, 2003). The properties of reference particles that are typically used in these studies differ, in general, from the ones of the pollutants. Therefore, it can be difficult to correlate toxicity of the references and the pollutants.

The ash of efficient combustion is mainly composed of alkali metal sulphates and chlorides, containing also various amounts of transition metal oxides such as ZnO (Sippula *et al.* 2009). The particle diameter is typically below 100 nm and thus fulfils the definition of nanoparticles (NPs, EC 2011). However, these efficient conditions are not typical for biomass combustion. In incomplete combustion condensed organic compounds are likely to participate in the particle formation. There are indications that organic compounds are capable of inducing radical alterations to the rheological behaviour, such as solubility, of the particles. Therefore the behaviour of the particles in contact with cells in a biological medium becomes more unpredictable.

In this study we have synthesized NPs containing ZnO, alkali salts and carbon in order to respond to the need for biomass combustion reference particles for toxicological studies. The NPs were designed to be similar in size, shape and chemical composition to the ash particles released from biomass combustion. Moreover, the physicochemical and rheological properties of the particles were characterised in order to identify the factors affecting their bioavailability. The effects on the cell viability of mice macrophage cells caused by the synthesized NPs were also evaluated.

The NPs were manufactured with chemical vapour synthesis (CVS, Moisala *et al.* 2006), using liquid Zn acetate dehydrate and alkali salts in water as a precursor solution. Purified air was used as carrier gas. The precursor was dispersed to gas with an ultrasonic atomizer and lead through a high-temperature (1000-1400°C) vertical flow reactor, and diluted rapidly with ambient temperature air to quench the chemical reactions. Particles were then analysed with on-line and off-line methods.

The resulting particles (Fig. 1) were characterised for size distribution, chemical composition, crystallinity, single particle morphology, thermal behaviour, surface properties, IR absorbance and rheological properties e.g. zeta potential and solubility. Cytotoxicity of the synthetic NPs was determined by a dose-response study which was carried out using mice RAW264.7 macrophage cells. The detected toxicological responses were compared to the real biomass combustion generated ash particle responses as described by Jalava *et al.* (2012).

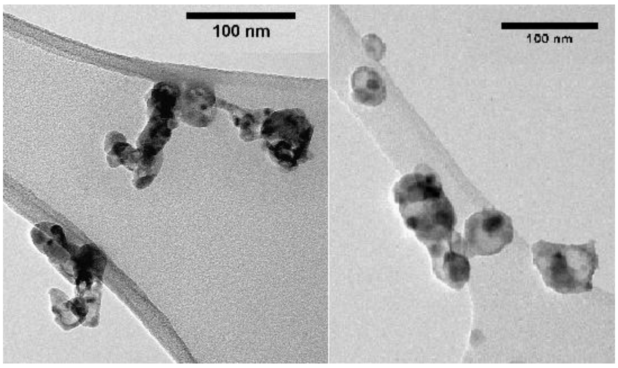

Figure 1. TEM images of ash particles released in biomass combustion (left figure), and ash–like NPs, synthesized with CVS process (right figure).

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