**Chronic exposure to engineered nanomaterials: sedimentation kinetics by Centrifugal Separation Analysis (CSA)**

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The extensive uses and applications of nano-based products lead to potential releases of engineered nanomaterials to the environment [1]. For this reason and due to the high complexity of ENMs fate processes, environmental risk assessment and, in particular, environmental exposure assessment to ENMs, have received a particular attention over the last years [2]. Although a lot of studies on fate and behavior of ENMs were already published, no standardized protocols has been provided to assess the characterization of ENMs stability in dispersion for medium and long-term exposure assessment.

In the present work, the characterization of the stability of the inorganic Aeroxide® P25 TiO2 and the organic NanocylTM NC7000 Multi-Walled Carbon Nanotubes (MWCNTs), the latter from EU-FP7 SUN project, was performed by Centrifugal Separation Analysis (CSA) over a maximum of 30 days, employing the Multi-wavelength Dispersion Analyzer LUMiSizer® 651 (L.U.M. GmbH, Berlin). Intensity of the transmitted light as function of time and position, at 470 nm, over the entire sample length, was collected. Transmittance values at the midpoint of the length of the cuvette, for different relative centrifugal force (RCF) and ENM concentrations tested, were converted to absorbance and then to concentration. The settling rate constant, k, was calculated at different RCF and concentrations by the equation C=(C0-plateau)•e-kt+plateau, in order to obtain k values at force gravity for each ENM concentration. According to ecotoxicological studies, each ENM was dispersed in the artificial freshwater OECD 203 at the following concentrations: 5, 10, 15, 20, 50, 100 mg/l for P25 and 6, 10, 14, 20, 40 and 100 mg/l for NC 7000. Humic acid (0.2% w/w) was added to NC7000 dispersions in order to avoid the very fast sedimentation that would not allow settling measurements.

The overall results showed that ENMs sedimentation was affected by the initial concentration: k at force gravity increased with the concentration for both ENMs but with an order of magnitude less for P25 (10-4) compared to NC7000 (10-3).

To our knowledge, this is one of the first studies that calculates the sedimentation rates of ENMs in an ecotoxicological medium by CSA up to 30 days. The adopted method can be proposed to be implemented in a Standard Operating Procedure (SOP) for supporting chronic exposure assessment for similar ENMs, with regards to physico-chemical properties.

References

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