Online Dose Determination For In Vitro Experiments With Nano Particles In The Karlsruhe Exposure System

Background
For the quantitative assessment of the toxicity of airborne nanoparticles the dose – response relationship is tested in in vitro test systems using bioassays of cell cultures as sensor. For the air-liquid interface exposure of cell cultures towards aerosols the Karlsruhe Exposure System (Mülhopt et al) (Fig. 1) was developed. After exposure the responses of the cells are analyzed to measure the biological responses such as viability, inflammatory or oxidative stress (Diabaté et al). For the determination of the dose response relationship accurate knowledge of the dose is an essential question (Teeguarden et al).

Aim
Online measurement of deposited particle dose in in vitro exposure experiments for assessment of particle toxicity.

Methods for Dose-Determination
Two alternatives were developed and compared:

1. Fluorescein sodium dosimetry (FSD): a highly sensitive method for a precise determination of the deposited mass exclusively on the membrane.

Fluorescein sodium aerosol is used as calibration particles (Fig. 3). After an exposure experiment the cell culture membrane is cut out of the Transwell® insert, washed in 10 ml ultra pure water and treated by sonication. The fluorescein sodium mass in the solution is analyzed by fluorescence spectroscopy.

2. Quartz crystal microbalance (QCM): an online measurement of the deposited mass per cell surface unit and time providing a control on the exposure quality.

In the Karlsruhe Exposure System one of the 6 exposure chamber the membrane is replaced by the sensor crystal of a QCM. It monitors the deposited particle mass per area unit and time.

Results
The FSD method and the QCM online monitoring are compared. Further experiments are required to improve the reproducibility of the measurement.

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<tr>
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<th>Deposited mass by FSD [µg/cm²]</th>
<th>Deposited mass by microbalance [µg/cm²]</th>
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<tbody>
<tr>
<td>Average of 5 values</td>
<td>1,50±18%</td>
<td>1,67±13%</td>
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<tr>
<td>determined over 2 hours</td>
<td>1,74</td>
<td>2,37</td>
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<td>Calibration factor (m_{QCM}/m_{FSD})</td>
<td>1,16</td>
<td>1,42</td>
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References


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