

## **Portable air-liquid interface exposure chamber for field emissions toxicity assays**

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Exposure of living cell cultures at air-liquid interface, mimicking, for example, the surface of human lungs, is believed to be one of the most realistic means to model toxicity of complex mixtures of pollutants on human health. Such assays, along with measurements under realistic conditions, such as portable emissions monitoring systems (PEMS) for measurement of real driving emissions, are important components of monitoring the anticipated effects of new technologies and emissions reduction efforts, typically based on surrogate measurements such as total particle number or mass. In this work, the concepts of air-liquid interface exposure and real-world emissions monitoring are combined into a portable exposure chamber, along with a mobile toxicological laboratory base, for field studies.

Living cell cultures are contained in standard 24-well holders, placed in airtight 17x13x9 cm exposure boxes, with a symmetrical head distributing the sample into 8 wells at 25 cm<sup>3</sup>/min per well with standardized 6 mm inserts. Sample and control air are conditioned to 5% CO<sub>2</sub> by volume, heated to 37°C, and relative humidity is increased to above 85% by membrane humidifier with deionized water. Up to four exposure boxes, two for sample and two for control, are placed in a commercial small scale (40x35x45 cm inner dimensions) incubator, housing sample conditioning, pump and various accessories. The mobile base laboratory, deployable in a van or indoor, includes a small laminar flow box for manipulation with cell cultures, an additional incubator for housing exposure boxes that are not actively undergoing exposition, a freezer and pressure bottles with CO<sub>2</sub> (conditioning) and synthetic air (control sample).

The system has undergone an extensive field validation sampling diluted vehicle exhaust as well as ambient air, including 4 h of exposure and 2 h transport in a vehicle each day for 5 days, 5-day operation in vans and sheds at -10 to +30°C outside temperatures, and transport of up to 800 km per week. Sampling system losses characterised with 4-200 nm carbonaceous and metallic particles and particle deposition rates characterized with transmission electron microscope on silver nanoparticles will be reported on and discussed. Toxicological tests performed in four localities on ambient air included measurements of cytotoxicity, oxidative stress, DNA damage and gene expression changes. The analyses are ongoing, however, so far, the most pronounced effects were observed near major road in Prague.

This setup overcomes the distance between the source and the toxicological laboratory, which is, in the opinion of the authors, the main hurdle of toxicity studies. While the toxicological side of the experiment is neither simple nor low-cost, it is the opinion of the authors that the mobile setup allows for field evaluation of toxicity of emissions from combustion engines, friction brakes, local heating appliances, nanomaterial production and handling facilities, and other sources, and of highly polluted ambient air.

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