Title: Toxicity profiling of wood combustion and ship diesel engine exhausts by air-liquid interface exposure

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Exposure to ambient aerosols, especially those formed during combustion processes, is linked to increased morbidity and mortality rates [1]. However, the aerosol components responsible for the adverse effects as well as the underlying mechanisms are still insufficiently characterized.

The "Helmholtz Virtual Institute of Complex Molecular Systems in Environmental Health" (HICE) is addressing this knowledge gap (http://www.hice-vi.eu). During joint measurement campaigns, controlled aerosol generation by combustion of different fuels is combined with comprehensive on-line characterization of the aerosols and in vitro investigations of their effects on human cells.

Here, we show results from in-vitro cell exposure experiments using an ALI exposure system [2] during the two HICE measurement campaigns in 2012. Exhaust emissions from a household wood-stove fueled with beech logs, as well as from a ship diesel engine fed with diesel fuel (EN 590) and heavy fuel oil and their effects on A549 lung epithelial cells were investigated.

The toxic equivalency (TEQ) values [3] of analyzed polycyclic aromatic hydrocarbons (PAH) per m^3, which were determined for the different diluted aerosols, increased in the order wood smoke < diesel fuel < heavy fuel oil. At the dilutions chosen for cell exposure, only the aerosols from the ship diesel engine caused acute cytotoxicity. Interestingly, no difference in toxicity could be seen between the complete and filtered aerosol. CYP1A1 mRNA levels, a biomarker for monitoring PAH exposure, increased after exposure to aerosols derived from heavy fuel oil but only moderately in response to wood smoke and diesel exhausts.

Our results suggest that gaseous components are the main contributors for the observed acute toxicity of diesel exhausts, demonstrating the importance of using air-liquid interface exposure techniques when investigating aerosol toxicity. As expected, the aerosol with the highest amount of PAHs also led to the strongest induction of CYP1A1. CYP1A1 induction was lower when the particulate phase was removed by filtering, thus implying particle bound compounds mediating this effect. The data also indicates that the TEQ value of PAH alone is not sufficient to predict toxicity of combustion-derived aerosols. Therefore, additional constituents, such as e.g. metals or noxious gases, have to be considered to assess the toxicity of wood smoke or exhaust emissions of ship diesel engines.

[1] Paur, H.-R., Cassee, F. R., Teeguarden, J., Fissan, H., Diabate, S., Aufderheide, M., Kreyling, W. G., et al. (2011). In-vitro cell exposure studies for the assessment of nanoparticle toxicity in the lung—A dialog between aerosol science and biology. Journal of Aerosol Science, 42(10), 668–692. doi:10.1016/j.jaerosci.2011.06.005

[2] Mülhopt, S., Diabaté, S., Krebs, T., Weiss, C., & Paur, H.-R. (2009). Lung toxicity determination by in vitro exposure at the air liquid interface with an integrated online dose measurement. Journal of Physics: Conference Series, 170, 012008. doi:10.1088/1742-6596/170/1/012008

[3] The MAK-Collection for Occupational Health and Safety: Part I: MAK Value Documentations. (2002). Weinheim, Germany: Wiley-VCH Verlag GmbH & Co. KGaA. doi:10.1002/3527600418