Toxicity of biomass combustion generated ultrafine particles: evidence from stack-sampled and airborne UFPs

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Wood burning for domestic heating is a relevant source of fine and ultrafine particles in urban areas. Nevertheless, there is still a gap of knowledge on health impacts associated to UFPs (dp<100 nm) because they are usually not monitored on a routine basis and because data on their physiochemical properties are very scarce in the literature.

The TOBICUP collaborative project (TOxicity of BIomass COmbustion generated Ultrafine Particles) intended to investigate the composition of ultrafine particles (UFPs) emitted by wood combustion and to elucidate the related toxicity. The project was developed with two parallel research lines: the first line was focused on UFP samples collected directly from residential wood combustion sources under burning cycles reflecting real-life situations; the second line was focused on airborne UFP samples collected at a sampling site where biomass burning for residential heating is widely used. Both research line shared the same methodology for sample collection and chemical and biological investigations.

UFPs were collected by means of three multistage cascade impactors (1 Small Deposit Impactor - SDI, Dekati - and 2 Micro-Orifice Uniform-Deposit Impactors - MOUDI by MSP Corporation): only UFPs collected on the two lower impaction stages and the back-up filter were analyzed in order to select particles with dae<100nm in each sampling. The impactors operated on different substrates, depending on the subsequent analysis to be performed: SDI collected UFPs on polycarbonate impaction foils for elemental analysis. One MOUDI operated with pre-fired quartz fiber filters for chemical analyses and on the other MOUDI aluminum foils were used as impaction substrates for collecting UFPs devoted to toxicological tests. UFPs chemical characterization included elements (Al, As, Ba, Cd, Co, Cu, Fe, Mn, Mo, Ni, P, Pb, Sr, Ti, V, Zn) by Inductively Coupled Plasma Atomic Emission Spectroscopy (ICP-AES), water soluble ions by Ion Chromatography (IC) levoglucosan and its isomers (mannosan and galactosan) by High Performance Anion-Exchange Chromatography (HPAEC-PAD), total carbon by Thermal Optical Transmittance (TOT), and PAHs (benzo(a)anthracene, chrysene, benzofluoranthene, benzo(a)pyrene, phenanthrene, anthracene, fluoranthene, pyrene) by GC-MS analysis; biological analyses were focused on the induction of the pro-inflammatory cytokine interleukin-8 (IL-8) by UFPs in two human cells lines (A549 and THP-1) and on UFP-induced oxidative stress and genotoxicity in A549 cells, investigated by comet assay and γ-H2AX evaluation.

Combustion tests were performed burning two common types of wood (beech and fir) in a commercial pellet (11.1 kW) and in a wood (8.2 kW) stove. UFP mass emission factors averaged to 424 mg/kgfuel for all the tested stove and wood type combinations except for beech logs burned in the wood stove (838 mg/kgfuel). Compositional differences were observed for pellet and wood

UFP samples, with higher TC levels characterizing the wood log combustion and potassium salts prevailing in pellet combustion samples. Additionally, wood samples contained more potentially carcinogenic PAHs with respect to pellets samples.

UFPs generated by pellets and wood logs combustion resulted in pro-inflammatory effects in THP-1 and A549 cells. Both cell lines responded to UFPs producing interleukin-8 (IL-8), but UFPs sampled from the flue gas of wood logs were more active compared to UFPs from pellet. With the exception of a higher effect observed with beech wood log UFPs in THP-1, the ability of soft or hard woods to induce IL-8 release was similar. In addition, on weight mass, IL-8 release was similar or lower compared to diesel exhaust particles (DEP), arguing against higher biological activity of smaller size particles. The higher activity of beech wood log UFP in THP-1 was not due to higher uptake or endotoxin contamination. Qualitatively different protein adsorption profiles were observed, with less proteins bound to beech UFPs compared to fir UFPs or DEP, which may provide higher intracellular availability of bioactive components (i.e. levoglucosan and galactosan) toward which THP-1 were more responsive compared to A549 cells.

Genotoxicity assessment through the comet assay and γ-H2AX evaluation performed on A549 human lung carcinoma cells showed significant DNA damage after 24 h treatment. The tail length data from the comet assay showed significant DNA damage (both single- and double-strand breaks) in cells treated with all UFP samples: pellet data were twice that for control cells and logwood data were twice as high as pellet data. Induction of DNA breaks investigated by γ-H2AX showed no statistical difference among samples but all showed a significant increase in DNA double-strand breaks compared to controls. In all appliance and fuel-type combinations investigated, the study of UFP chemical composition suggested a combined effect of anhydrosugars (especially levoglucosan), elemental content (especially Fe, Al), and PAHs.

Ambient UFPs collection was carried out during summertime and wintertime 2015 at the alpine town of Morbegno (Sondrio), Northern Italy. Sampling campaigns were performed in a winter period when wood burning was expected to be a relevant source (Jan-Feb) and in a summer period when conversely wood burning was considered to be very limited and almost negligible (Jun-Jul). Due to difference in average particulate matter concentrations during the two periods, sampling times were integrated over three/four days in the winter and seven days in the summer campaign.

UFPs concentrations in ambient air did not show significant seasonal differences (about 2.2 $\mu g/m^3$); opposite to mass concentration, UFP composition was season-dependent for some detected species. Total PAHs contribution was higher during wintertime compared to summertime and tracers of wood burning emission (i.e. levoglucosan and its isomers, K^+, and benzo(a)pyrene) were characterized by significant seasonal differences and very high (N8) winter to summer ratios.

Biological analyses showed that ambient UFPs can evoke a pulmonary inflammatory response by inducing a dose-related IL-8 production and DNA damage, with different responses to summer and

winter UFP samples. Airborne UFPs induced a dose-related IL-8 release in both A549 and THP-1 cells. with particles collected during summertime being more active. THP-1 cells were more sensitive than A549 cells. Compared to DEP, UFPs sampled during wintertime were less active in both cell lines, while UFPs collected in summer showed a similar or higher activity. On a weight basis data did not support a higher biological activity of ambient UFPs compared to DEP. Due to their ability to cross lung epithelial barrier, UFPs can reach systemic circulation and activate blood leukocytes, as demonstrated by the production of IL-8 in the whole blood assay. Comet assay and γ-H2AX evaluation showed a significant DNA damage especially in winter UFPs compared to control samples; this damage resulted related to PAHs concentration thus pointing at a possible mechanism of oxidative damage.

Further references for the TOBICUP project

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