TOXICITY OF SIZE SEGREGATED AEROSOL IN THE AMBIENT FROM HEAVILY POLLUTED CITY OF OSTRAVA CZECH REPUBLIC

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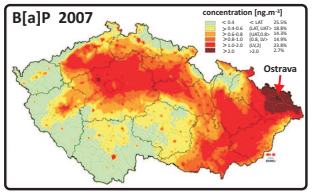


BACKGROUND

Exposure to air pollutants significantly contributes to morbidity and mortality of inhabitants living in affected regions. Ostrava, one of the most industrialized cities in the Czech Republic, suffers from serious air pollution problems especially during winter seasons. Toxic compounds bound to particulate matter of different size, including ultrafine particles, may play crucial role in the adverse health effects of the air pollution.

AIM

The aim of our study was to analyze toxicity of extractable organic matter (EOM) from particulate matter (PM) of four different PM size fractions including nanoparticles (< 0.17 µm – nanoparticles ; 0.17 – 0.5 µm – lower accumulation mode; 0.5 – 1 µm – upper accumulation mode; 1 – 10 µm – coarse fraction). The contribution of the size fractions to various toxicity endpoints will be assessed.

METHODS

Sample Collection

PM samples were collected during winter 2012 (January 26 – February 20, 2012) using a high volume cascade impactor (BGI 900, USA) on polyurethane foam (PUF) with an integrating time of 23 h. PUFs were extracted with dichloromethane and chemical analysis of polycyclic aromatic hydrocarbons (PAHs) was performed using HPLC with electrochemical detection. EOMs from PMs of individual size fractions were then pooled into three groups according to the inversion episode and concentration of PAHs: group 1a contained highest PAH concentrations, followed by group 1b and group 2 (see Fig. 1).

Toxicity studies

Cell cultures and treatment:

Toxicity studies were conducted using A549 cells, a model human lung epithelial cell line. Cells were treated with a subtoxic dose of EOMs corresponding to 3 m^3 of the sampled air for 24 hours.

Cytotoxicity:

Cytotoxicity was measured by lactate dehydrogenase (LDH) test.

Genotoxicity:

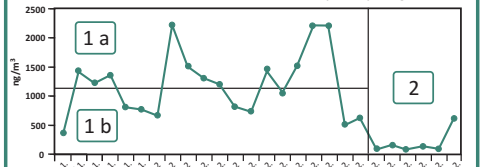
Bulky DNA adduct levels were assessed using ^{32}P-postlabeling with nuclease P1 enrichment.

Oxidative damage of DNA, lipids and proteins:

8-Oxodeoxyguanosine (8-oxodG), a marker of DNA oxidation, was analyzed by HPLC-MS/MS; ELISA was used to analyze 15-F_{2t}-isoprostane (15-F_{2t}-IsoP), a marker of lipid peroxidation, and protein carbonyls, a marker of protein oxidation.

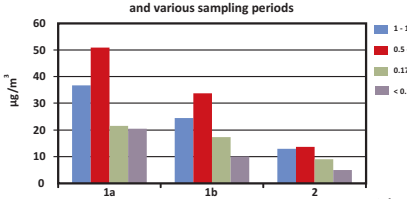
RESULTS

Fig. 1. Time course of the total PAH levels bound to PM [ng/m^3] within sampling period in Ostrava-Bartovice and the distribution of samples for pooling.



The whole sampling period might be divided in 2 major parts: 1. Cold weather with frequent inversion episodes, 11 days (1a) and periods between inversion episodes, 9 days (1b); 2. Windy and warm weather (temperature was 15-20°C higher than in the rest of sampling period, 6 days (2).

Fig. 2. Aerosol mass in individual size fractions and various sampling periods



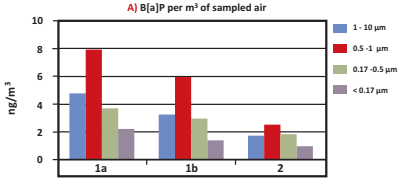
Very high PM concentrations were found during inversions (PM ~10 130 µg /m^3) PM size fraction of 0.5 - 1 µm is the most abundant for all sampling periods.

CONCLUSIONS

- Upper accumulation mode (0.5-1 µm) is the most abundant PM fraction for all sampling periods representing 33-40% of total PM mass. PM levels were ~3-fold higher during inversion period 1a compared to warm and windy period 2.
- B[a]P and c-PAH levels per m^3 are also highest in upper accumulation mode. The B[a]P and c-PAH levels are ~3-fold higher during inversions and are generally extremely high. As per mg PM, the distribution of PAH among PM size fraction is uniform.
- Highest bulky DNA adduct levels are induced by EOMs from upper accumulation mode in all three pooled groups. Group 1a representing inversion period exhibits approx. 5-fold higher genotoxicity compared to group 2. As per mg of PM, ultrafine fraction (<0.17 µm) became more significant for the overall genotoxicity of EOMs.
- The results of oxidative damage to biomolecules did not indicate clear effects of EOMs; however, there was a trend of decrease of levels of oxidative stress markers, particularly peroxidized lipids.
- In contrast to genotoxicity (DNA adducts), ultrafine PM fraction is the most efficient oxidant of biomolecules.

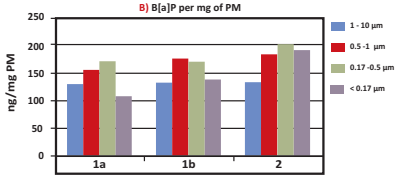
Fig. 3. Concentrations of benzo[a]pyrene (B[a]P) bound to size segregated aerosol

A) B[a]P per m^3 of sampled air



Concentrations of B[a]P are extraordinary high. Total B[a]P levels in PM10 varied between 7-18 ng/m^3 while limit recommended by WHO is 1 ng/m^3. The fraction 0.5-1 µm is the main carrier of B[a]P.

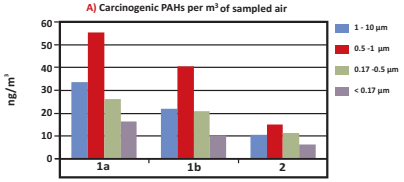
B) B[a]P per mg of PM



As per mg PM all the fractions have similar content of B[a]P.

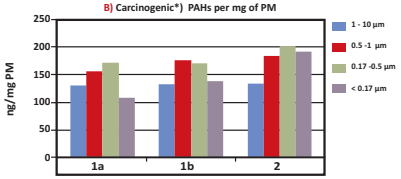
Fig. 4. Concentrations of carcinogenic PAHs*) bound to size segregated aerosol

A) Carcinogenic PAHs per m^3 of sampled air



The distribution of seven carcinogenic PAHs among various size fractions is very similar to the distribution of B[a]P. Again, the c-PAH levels are generally very high.

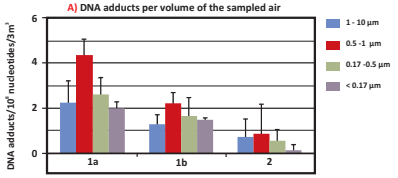
B) Carcinogenic*) PAHs per mg of PM



*) Carcinogenic PAHs include: benz[a]anthracene (B[a]A), chrysene (CHRY), benzo[b]fluoranthene (B[b]F), benzo[k]fluoranthene (B[k]F), benzo[a]pyrene (B[a]P), dibenzo[a,h]anthracene (DB[ah]A), and indeno[1,2,3-cd]pyrene (I[cd]P).

Fig. 5. Total DNA aduct levels induced by EOMs extracted from size segreated aerosol in A549

A) DNA adducts per volume of the sampled air



The results suggest highest bulky DNA adduct levels after cell treatment with EOMs from particle size fraction 0.5 – 1 µm in all three pooled groups. Highest DNA adduct levels were also found after treatment of the cells with group 1a EOMs. These samples exhibit approximately 5-fold higher genotoxicity compared to group 2.

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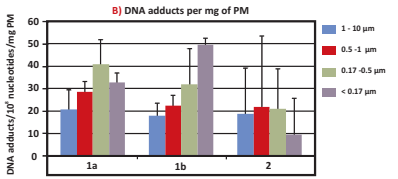
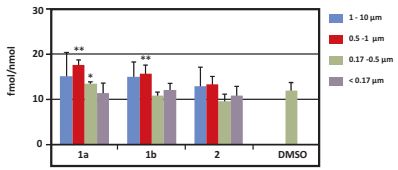


As per mg of PM, ultrafine fraction became more significant for the overall genotoxicity of EOMs.

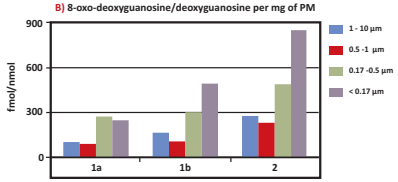
Fig. 6. Oxidative damage of DNA induced by EOMs extracted from size segregated aerosol in A549

A) 8-oxo-deoxyguanosine/deoxyguanosine per volume of the sampled air



Weak increase in oxidative DNA damage was observed.

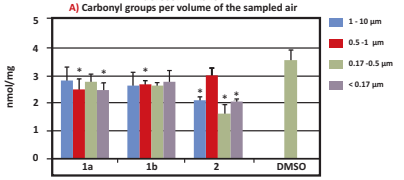
B) 8-oxo-deoxyguanosine/deoxyguanosine per mg of PM



As per mg of PM, ultrafine fraction is the most effective fraction in oxidative DNA damage for all sampling periods.

Fig. 7. Oxidative damage of proteins by EOMs extracted from size segregated aerosol in A549:

A) Carbonyl groups per volume of the sampled air



Slight, but significant decrease of the oxidative damage of proteins was observed for several PM size fractions.

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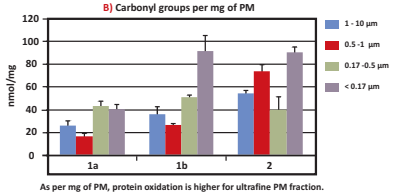
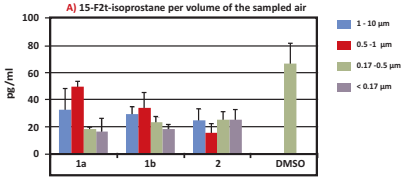


As per mg of PM, protein oxidation is higher for ultrafine PM fraction.

Fig. 8. Oxidative damage of lipids by EOMs extracted from size segregated aerosol in A549:

A) 15-F2t-isoprostane per volume of the sampled air



Interestingly, the decrease of 15-F2t-isoprostane levels resulted from incubations from all EOMs. This effect may be related to the activity of cyclooxygenase 2 (COX2) that is induced by carcinogenic PAHs and EOM exposure. COX2 decreases the levels of arachidonic acid (AA), a substrate for 15-F2t-IsoP formation, resulting in limited availability of AA for 15-F2t-IsoP synthesis.

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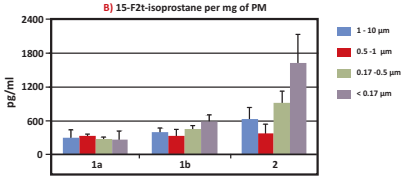

As per mg of PM, lipid peroxidation (LPO) is higher for ultrafine fraction collected out of inversion periods. This result suggests differences in chemical composition of various PM fraction as well as main air pollution sources at various sampling periods.

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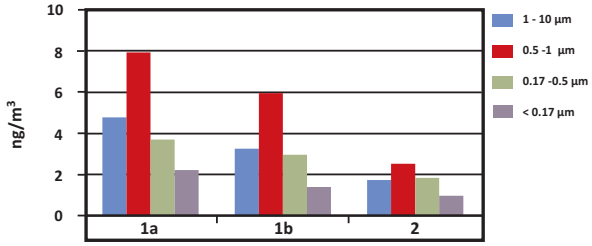

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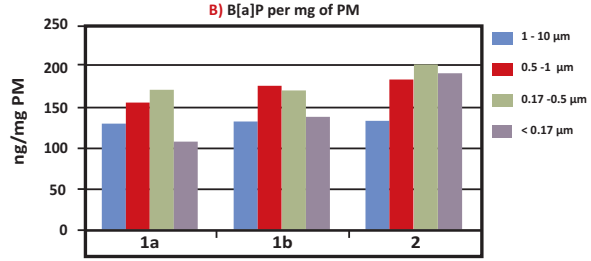

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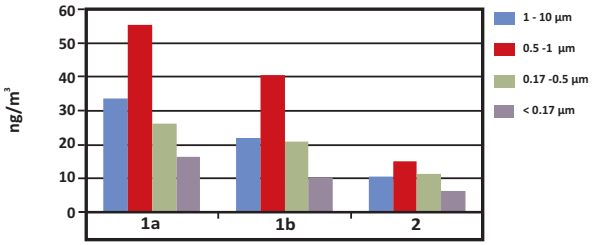

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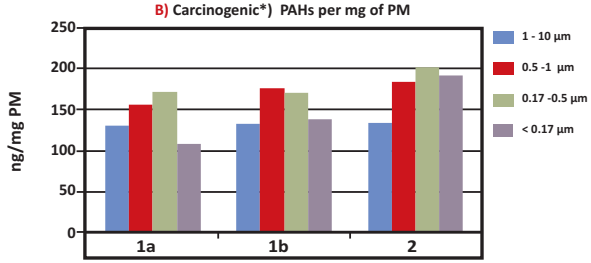

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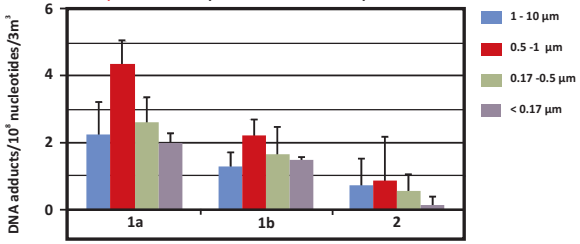

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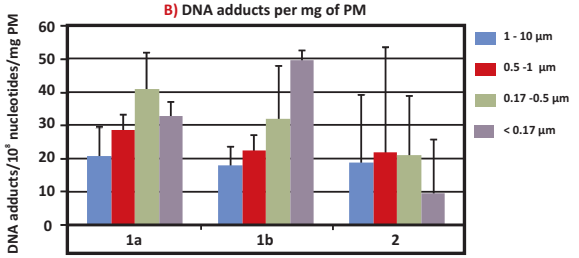

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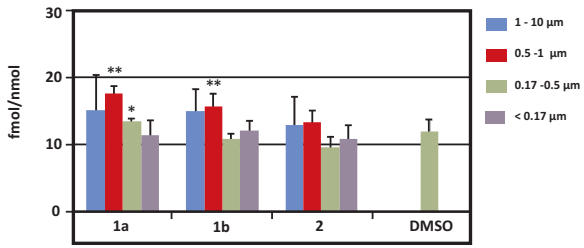

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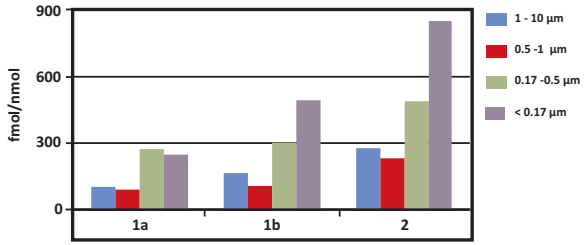

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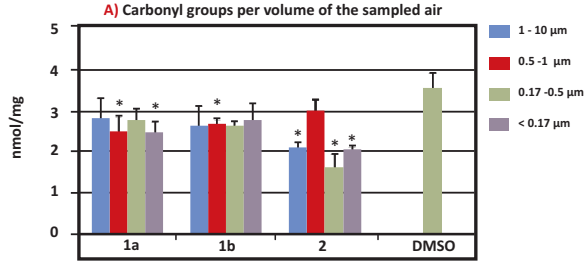

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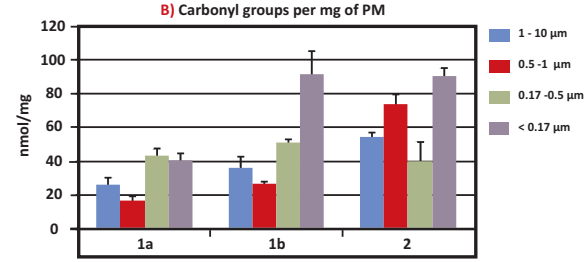

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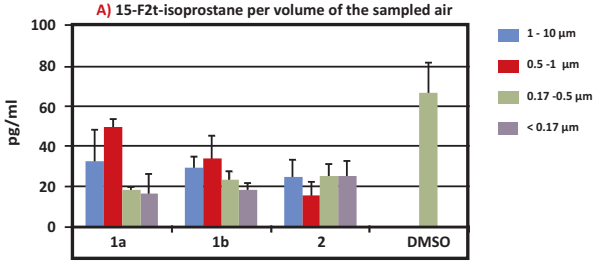

Interestingly, the decrease of 15-F2t-isoprostane levels resulted from incubations from all EOMs. This effect may be related to the activity of cyclooxygenase 2 (COX2) that is induced by carcinogenic PAHs and EOM exposure. COX2 decreases the levels of arachidonic acid (AA), a substrate for 15-F2t-IsoP formation, resulting in limited availability of AA for 15-F2t-IsoP synthesis.

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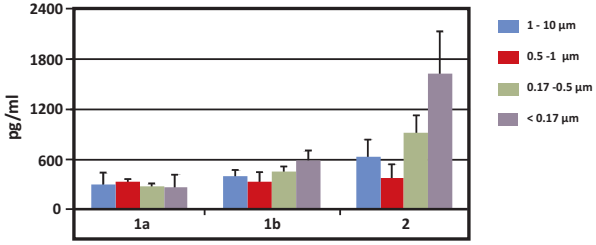

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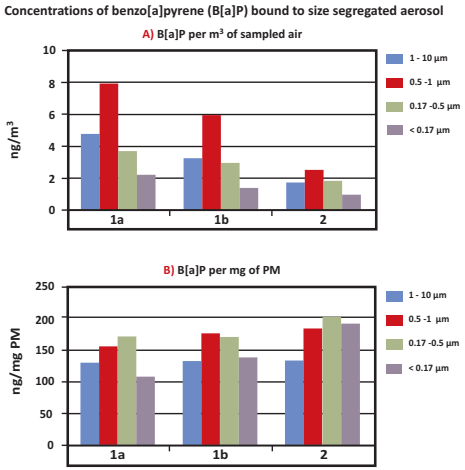
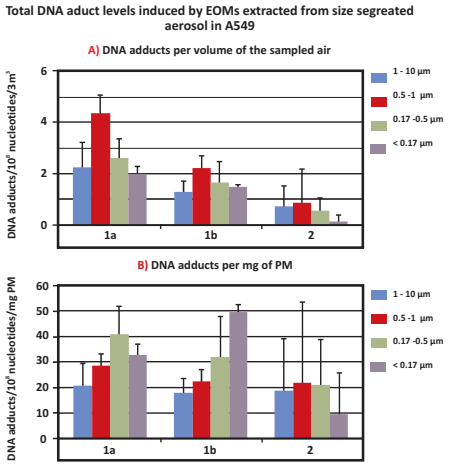
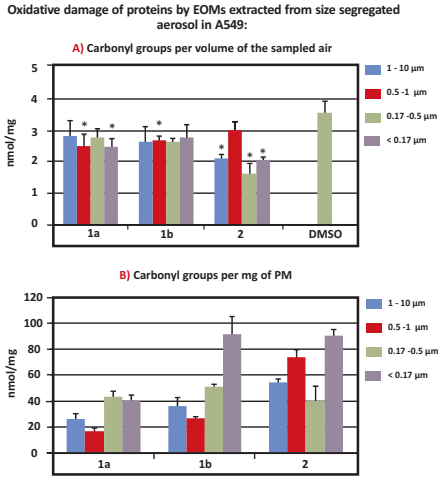
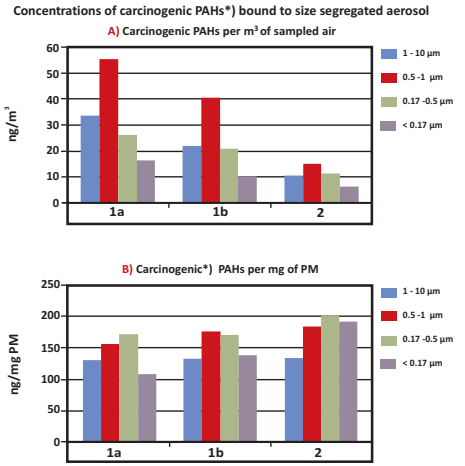
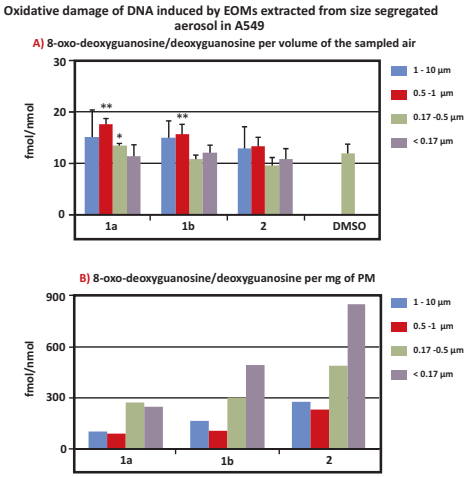
ještě jsem ani neodjel a už mám pro Vás další práciJ.
Potřeboval bych ze všech Figures na mém posledním posteru udělat slidy v PowerPointu. Jen prosím vynechte číslování a komentáře k obrázku. Legenda k Fig by však měla zůstat stejná. Z hlediska termínu to stačí do konce příštího týdne.

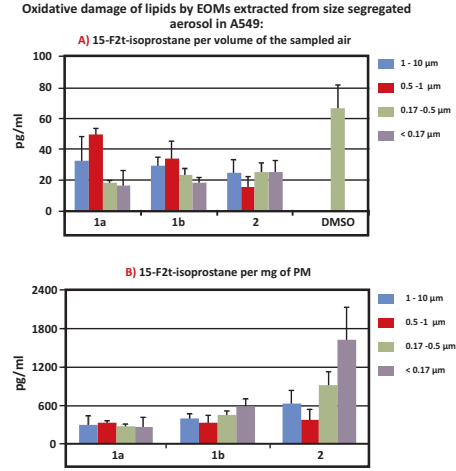












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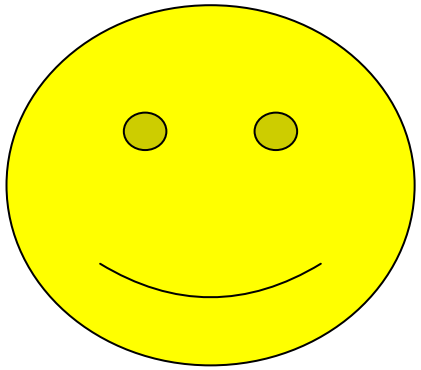

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