PM Oxidative Potential:

response of acellular assays to predict PM-induced oxidative stress activity

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Ascorbic acid a-cellular assay for measure of PM oxidative potential: effect of the composition of the artificial respiratory tract lining fluid

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This work describes the ascorbic acid assay, as one of the most common cell-free methods used to quantify the oxidative potential (OP) of particle matter (PM). It uses the ascorbate (Asc), as the most abundant antioxidant found in lung fluids, which has a vital role in oxidant production from redox-active species.

To make the OP^{AA} response a surrogate measure of the in vivo capacity of PM to induce ROS, there is the need of designing an assay set up that most closely simulates the interactions of the inhaled particles with interstitial macrophages in the lung. With this purpose, this work investigates different experimental conditions representing an artificial respiratory tract lining fluid (RTLF). In comparison to the simplest surrogate containing only ascorbate, composite solutions were used by adding three endogenous lung components at physiological levels, namely reduced glutathione (GSH), urate (UA) and citrate (CIT).

The study was performed on 20 real $PM_{2.5}$ samples collected at an urban and rural site in the Po Valley. The obtained results clearly show that the OP^{AA} responses change with the composition of the synthetic RTLF, as they significant decreased ($p < 0.05$) by adding other antioxidants to ascorbate (Fig 1).

Although each antioxidant shows a different effect, a general order is followed for all the analyzed samples:

Asc > Asc + CIT > Asc + UA > Asc + GSH > Asc + CIT+GSH ~ Asc + CIT + GSH + UA.

All the investigated surrogates generate linearly correlated responses ($R^2 \geq 0.8$), indicating that the effect of the various RTLF surrogates on OP^{AA} responses is similar for all the samples. This suggests that all the tested assay set ups are useful for measuring OP^{AA}.

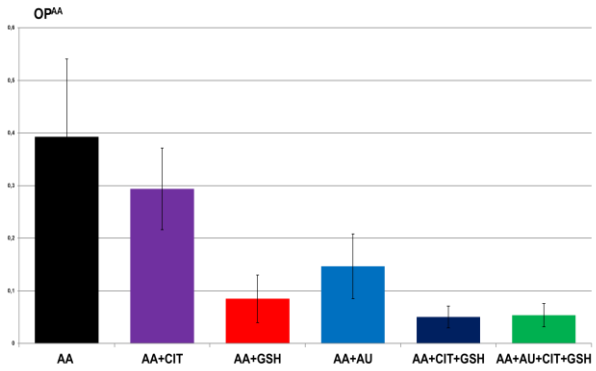

Figure 1. OP^{AA} responses (nmol min^{-1} m^{-3}) of $PM_{2.5}$ real samples measured in six different RTLF surrogates.

For comparison, the study was expended to standard solutions of redox-active species that are known to give positive response to the AA assay, namely: Cu^{2+}, Fe^{2+}, 1,2-naphthoquinone, 1,4-naphthoquinone and 9,10-phenantrenequinone.

By comparing the dependence of OP^{AA} on RTLF composition, we could infer that Cu^{2+} and quinones were the redox active species most responsible of the OP^{AA} response of the analysed $PM_{2.5}$ samples.

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