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Seasonal and dimensional differences in the proinflammatory potency of atmospheric particulate matter. Comparison of in vitro results with clinical effects in children exposed to the same particles from the same urban area. The Milan Prolife Project.

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One issue of major concern in the evolution of adverse health effects from environment pollution is to compare data from in vitro studies and experimental models in animals to clinically evident effects in humans. A wide range of research studies- both basic and clinical studies- have been performed, trying to relate pollution events to hospital admissions and/or major respiratory or cardiovascular adverse effects.

In particular, hospital admissions to the main pediatric service of Milan were recorded, analysed and compared with daily and seasonal variation in PM_{10} and $PM_{2,5}$ concentration, during 4 consecutive periods: winter 2007-summer 2007-winter 2008-summer 2008. There were 390 acute pediatric admissions in 2008. Respiratory disease were classified as follows: asthma or asthma like disorders; upper respiratory diseases (pharyngitis, pharingotonsillitis, otitis); lower respiratory diseases (bronchitis, bronchiolitis and pneumonia).

During 2007, there were in total 391 pediatric admissions for respiratory disease; 192 (99 males and 93 females) during the 1st winter semester, and 199 during the 2nd semester.

The mean age (SD) was 31 months +/- 38.1, range 23 mo-15y. The mean hospital stay was 5 days +/- 2.6, range 1-17 days. There was 12.7% asthma or asthma related admission; mo 55.8% due to lower respiratory illness, and 31.5 % due to upper respiratory disease. The daily coverage of PM_{10} concentration during the 1st semester 2007 was 48.3 +/- 17.9 mcg/m^3 median 47. There were 107 (59.1%) days with at least hospital admission. The mean daily concentration of PM was higher in days with (107) than without (n=74) hospital admissions ($p=0.032$ or < 0.05).

In addition, the human bronchial epithelial cell-line BEAS-2B and the human alveolar epithelial cell A549 were seeded at a concentration of 80.000 cell/well and treated after 48 hrs with both summer and winter PM 10 and PM 2.5 sampled in the main Milan urban area.

Cytotoxicity was assessed by HOECHST 33342/91 staining. Viability was calculated as the sum of viable mitotic cells. Release of the proinflammatory citockine IL was measured by sandwich ELISA assay. Oxidative stress was evaluated by chemiluminescence and genotoxicity was assessed by comet assay. It was found that whereas A549 cell viability was not significantly reduced after summer and winter PM exposure, summer PM had no significant effects on BEAS-2B viability, whereas winter PM treatment induced a decrease in cell viability, both at the dose of 25 and 50 mcg/cm^2. In addition, whereas both winter and summer $PM_{2.5}$ produced only a slight increase in IL 8 release, winter PM_{10} induced a 5 fold increase in IL 8 release in treated cells, and summer PM_{10} induced a 20 fold increase ($p< 0,05$) in IL 8 expression, which was reduced to 11- fold- increase after polimixin treatment.

In conclusion, in vitro studies using PM_{10} and $PM_{2,5}$ sampling from different seasonal samples seem to correlate with clinical data in children exposed to the same type and concentration of PM during winter and summer season. In particular, BEAS-2B resulted more responsive to PM treatment than A549. Winter PMs were more cytotoxic than summer PMs; Summer PM_{10} had a higher proinflammatory potential, which could be partly due to biological components (LPS).

Accordingly, children acute admissions were significantly affected by pollution data, with increased admission during winter time, namely for upper air tract infections, whereas lower tract inflammation and acute admission for asthma were more frequent during the spring: summer season.

Even if great caution is required when trying to relate in vitro studies to clinical effects in humans, the present report is the first study in a large urban area trying to compare “in vitro” and clinical effects of the same urban particles in different seasons of the year.

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