

# TOXICOLOGICAL EFFECTS OF INDOOR PM<sub>10</sub> IN PRIMARY SCHOOLS EXPOSED TO DIFFERENT STREET TRAFFIC INTENSITIES ACROSS THE CITY OF BARCELONA, SPAIN



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<sup>4</sup>AP23075 Barcelona, Spain. BREATHE-BRAIN dEvelopment and Air pollution ultrafine particles in sChool childrEn (Advanced Grant ERC, Seventh Framework Programme).

## INTRODUCTION



The BREATHE project funded by the European Union is currently measuring aerosols in primary schools located in the city of Barcelona in order to assess the risk that air pollution poses to the neuro-development and behavioural patterns of children.

The schools are separated into high and low-traffic areas with the primary aims being to recognize if differences in ambient traffic emissions affect the neurological system of children and if there is a relationship between their behaviour at school and the levels of air pollution in their environment. Within the BREATHE project the Complementary Action CECAT is considering the toxicological aspect of the problem by investigating the toxicity of PM by means of its ability to induce a systemic oxidative stress which damages cells and DNA molecules, and thus create subsequent inflammation to produce a disease.

A total of 20 schools have been selected and are being currently sampled. This sampling is being carried out inside the classrooms for 4 consecutive days at two different times a year (winter and summer of 2012) to take into account the effect of changes of air pollutants in different climatic conditions.



Sampling is simultaneous in Indoor and outdoor school environments



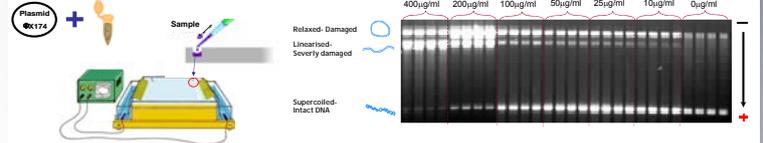
## TOXICOLOGICAL ASSAYS

One of the mechanisms that has been commonly proposed to explain the association of PM exposure and occurrence of respiratory infections, lung cancer, and chronic cardiopulmonary diseases is oxidative DNA damage through the generation of Reactive Oxygen Species (ROS). The biochemical pathways leading to cell damage involve both non-cellular characteristics of particles (including shape, size, solubility, surface reactivity, carrier function, and surface chemistry) and cellular properties (including the ability of generating ROS, alteration of signalling pathways, and initiation of inflammation).

For the determination of particle oxidative capacity, PM<sub>10</sub> in the classrooms is being collected using an Airborne Sample Analysis Platform system (ASAP; Model 2800 Thermo, USA) on polyurethane foam substrates (PUF) with a high sample flow-rate of 200 l/min. The genotoxicity, inflammatory potential and cytotoxicity of the PM<sub>10</sub> samples will be elucidated using three different but complementary biological assays:

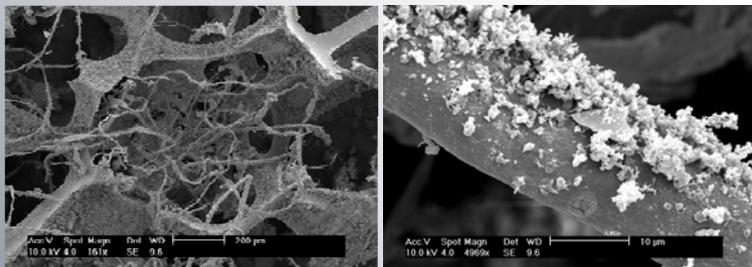
- Plasmid Scission Assay (PSA) - genotoxicity
- DCFH ROS Assay - potential pro-inflammatory
- F-actin polymerisation Assay - cytotoxicity

### Plasmid Scission Assay

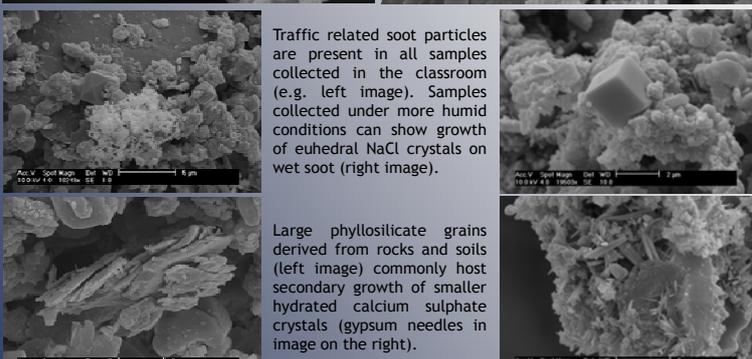
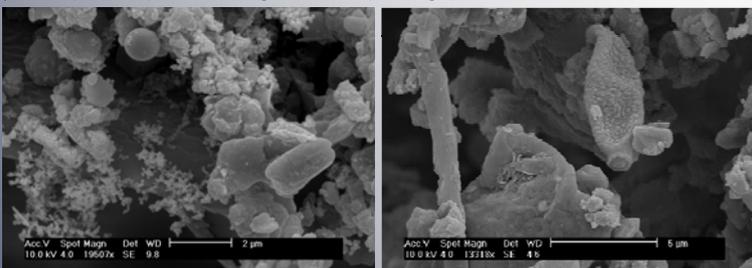


The Plasmid Assay is an in vitro method of assessing and comparing the toxicity of fine particles through their ability to produce free radicals. By incubating supercoiled plasmid DNA ( $\Phi$ X174 RF) with particles in solution, free radical activity will cause sequential nicking of the DNA. The plasmid DNA is incubated in varying concentrations of PM and separated by agarose gel electrophoresis. This results in the gradual uncoiling of the DNA until it unwinds completely to a relaxed coil form. Further free radical damage will cause the relaxed coiled DNA to linearise, and then to fragment. Separation of the different forms by agarose gel electrophoresis allows quantification of each form using densitometry.

## SCANNING ELECTRON MICROSCOPY: INDOOR SCHOOL PM

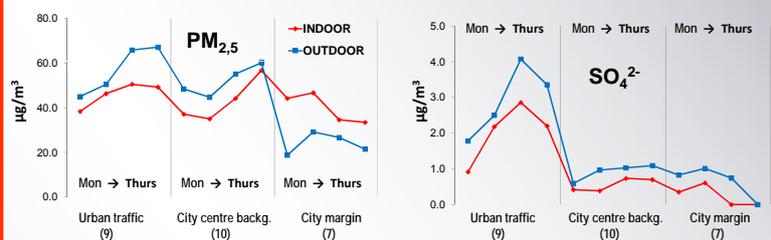


Cotton fibres (top left) presumably derived from clothing are coated with smaller particles that include abundant traffic-related soot and mineral PM (top right). Less commonly present are spherical fly ash particles (lower left) and various organic aerosols (lower right).

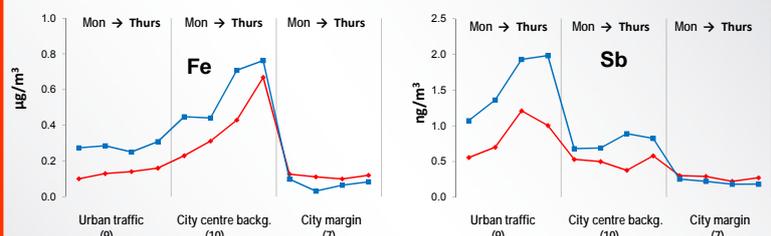


## GEOCHEMISTRY

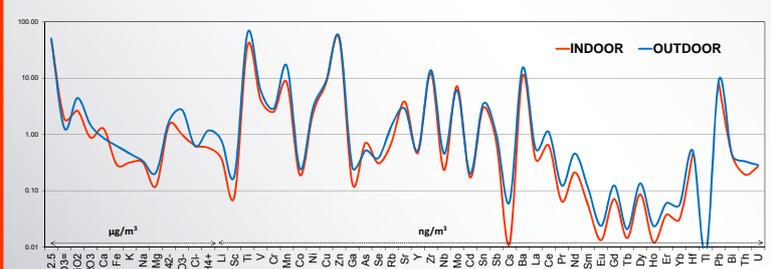
Database for all school monitoring sites includes full inorganic chemical characterisation of both indoor and outdoor air. Some initial results are shown below:



- Wed-Thurs > Mon-Tues = PM weekday build-up
- Indoor PM similar at all sites
- Outdoor > indoor in city centre & traffic spot but not in city margin



- High FePM<sub>2.5</sub> in city centre
- High outdoor Sb at traffic hot spot (brake emission) with progressively increasing weekday concentration
- Outdoor > indoor in city centre & traffic spot but not in city margin



The results that are currently being obtained and presented in this congress are helping us i) to reveal the effect of traffic emissions on air quality in school indoor environments, ii) to identify the inorganic components that may have an adverse effect on health, iii) to quantify the biological responses that subsequently cause health effects through measurements of both reactive species of oxygen and inorganic components (as well as their synergistic effects) in order to reveal exposure dependent alterations.