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Aero-dispersed mutagenicity attributed to particulate and semi volatile phase in an urban environment

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HIGHLIGHTS

- Both chemicals and meteo-chemical parameters can influence the mutagenicity of air pollution.
- The gas phase and particulate phase mutagenicity can be different and affected by season.
- The gas phase accounted for only 1% of the observed mutagenicity.
- The particulate mutagenicity is approximately 5-fold higher during winter.
- The contribution of the nitro-derived compounds seems to be crucial.

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ABSTRACT

Commonly the atmospheric pollution research is focussed on particulate indicators especially when mutagenicity was studied. On the other hand the volatile and semi-volatile compounds no adsorbed on to the particles can be genotoxic and mutagenic. Moreover some mutagenic compounds, such as polycyclic aromatic hydrocarbons, are present both in the particulate and in the gas-phase in according to chemical conditions. This work is focussed on the assessing of the total mutagenicity shifting the gas-phase and particulate phase, during two seasons, in Turin. Two sampling sessions are conducted for total particulate matter and gas-phase pollutants. Moreover meteorological and usual air pollution monitoring data were collected at the same sampling station. The Salmonella assay using the strains TA98 and YG1021 was conducted on each organic extract. The mean level of total suspended particles, PM10 and PM2.5 were 73.63 ± 26.94, 42.85 ± 26.75 and 31.55 ± 26.35 μg m^{-3}. The observed mutagenicity was PM induced YG1021 > PM induced TA98 > PM induced TA98+S9 ≫ non-particle induced YG1021 > non-particle induced TA98 > non-particle induced TA98+S9. The multivariate regression is significant when we consider air pollution and meteorological indicators and chemical conditions as predictors.

1. Introduction

Air pollution is one of the most important worldwide health concerns (WHO-Europe, 2013). Particularly in the last 10 years, in both the US and Europe, new directives and regulations supporting more restrictive pollution limits were published (Krzyzanowski, 2008). However, the early effects of air pollution cannot be avoided, especially for the urban population (EEA, 2012). A recent Eurobarometer survey showed that European citizens are deeply concerned about the impact of air pollution and that more than 70% of the European population is worried that air pollution and air quality is worsening over time (EU, 2013). The decision to designate 2013 as the Year of Air reflects both the economic seriousness of the problem but also the impacts on humans. Approximately 3% of cardiopulmonary and 5% of lung cancer deaths are attributable to particulate matter (PM) pollution worldwide (HEI, 2013), while the disease burden related specifically to PM2.5 pollution accounts for approximately 3.1% of the global disability-adjusted life years (Lim et al., 2012).

The total suspended particulate (TSP) air pollution is widespread and consists of a mixture of solid and liquid particles suspended in

Abbreviations: PAHs, polycyclic aromatic hydrocarbons; PCR, polymerase chain reaction; TSP, total suspended particles; PM, particulate matter; PM10, particulate matter with an aerodynamic diameter <10 μm; PM2.5, particulate matter with an aerodynamic diameter <2.5 μm.

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the air. The physical and chemical characteristics of TSP vary by site. Common chemical constituents of PM include sulphates, nitrates, ammonium and other inorganic ions, but also include organic carbon, crustal material, particle-bound water, metals, aromatic hydrocarbons such as polycyclic hydrocarbons and their nitrated, oxidised, sulphated forms (Claxton et al., 2004; Breysse et al., 2013). Especially in urban polluted locations, the secondary particulates formed from precursor gases are the prevalent toxic agents. Particle accumulation and coagulation reactions in the atmosphere produce a fine fraction of particulate matter (PM2.5) that often constitutes more than fifty percent of the TSP (Dimitriou and Kassomenos, 2013). The emitted chemicals, the dispersion conditions, and physical parameters such as humidity and temperature (Zhang et al., 2012) can all influence particle formation.

A large number of studies provide evidence of a correlation between both for short term and long-term exposure to PM pollution and health effects such as morbidity and mortality from cardiovascular and respiratory diseases, as well as from lung cancer (Krzyzanowski, 2008). At the end of 2013, outdoor air pollution and its major component, outdoor particulate matter were classified as carcinogenic for humans (1 Group) (Loomis et al., 2013).

Many mutagenic and genotoxic compounds are present in air pollution, and the effects are widely known and reviewed (de Kok et al., 2006; Claxton and Woodall, 2007a; Valavanidis et al., 2008; DeMarini, 2013).

The finest air pollution fractions, PM10 (particles with a diameter of less than 10 μm) and PM2.5 (particles with a diameter of less than 2.5 μm) show greater genotoxicity (Claxton et al., 2004), while the ultrafine particles (particles having a diameter of less than 0.1 μm) are the subject of in-depth analyses (Hoek et al., 2010; Kovats et al., 2013). The studies conducted using *in vivo* and *in vitro* models show the induction of mutations and genotoxic effects. However, non-genotoxic effects also occur and various studies focused on the epigenetic effects of the ambient particles (Ji and Hershey, 2012).

Among the typical air pollution chemicals, Polycyclic Aromatic Hydrocarbons PAHs have a relevant role in air pollution toxicity. These compounds are reactive in the atmosphere and primarily form oxidised products, the most notable being oxy-derivatives (mostly quinones) and nitrated compounds (Kim et al., 2013). Some of these compounds, such as benzo(a)pyrene and 6-ditrochrysene and the 7,12-dimethylbenz(a)anthracene, are also present in primary emissions. Benzo(a)pyrene is the reference compound for the carcinogenic relative potency factor, while others previously cite PAHs as having a carcinogenic factor of 10 and 64, respectively. Also among the secondary PAHs are compounds with high carcinogenic relative potency factors such as benz(j)aceanthrylene (60) and 1,6-dinitropyrene (10) (ATSDR, 1995). The historic list of 16 USEPA priority PAHs is an important source of information, but was developed when knowledge of the relative toxicity of PAH congeners was more limited than at present. As such, it is useful as reference for monitoring but limited for the assessment of human health risks attributable to air PAH mixture exposition (Yang et al., 2007).

Vapour–particle partitioning of mutagens can be quantified using the gas–particle-partitioning coefficient for each compound. This coefficient is influenced by both the adsorption and absorption processes and is strongly temperature dependent (Albinet et al., 2008). Moreover, volatile and semi-volatile organic compounds associated with particulate matter can be influenced by heterogeneous photochemical reactions in the atmosphere (Fraser et al., 2000; Xie et al., 2013). Our typical samplings were conducted using standard methods that are affected by relevant limits (Liu et al., 2007; Forbes et al., 2012).

The aim of this work is to assess the mutagenicity of particulate and not-particulate air pollution and to determine the effects of seasonality and the contribution of nitro-compounds to the mutagenic effects in an urban environment.

2. Materials and methods

2.1. Sampling

Sampling was performed from 20 November to 22 December 2009 for the winter period and from 4 May to 4 June 2010 for the spring period at a meteorological–chemical station of the Environmental Protection Regional Agency (Piedmont A.R.P.A.) located at Torino, in the northwest of the Padana Plain, Italy. The sampling site, called Lingotto, was located outdoors in a small green area within an enclosed zone classified as urban background (ARPA Piemonte, 2010). Turin has 872,367 inhabitants and a population density of approximately 7000 inhabitants per km^2; thus, the pressure on the territory that is associated with human activity is very high (ISTAT, 2012). Moreover, the climate and topographical characteristics of the area contribute to critical air pollution (Poncino et al., 2009; Eeftens et al., 2012). The Total Suspended Particles (TSP) were collected on glass micro-fibre filters (Type Fiberfilm T60A20, 150 mm, SKC, 863 Valley View Road Eighty Four, PA 15330, USA) and micro-pollutants were collected in Polyurethane Foam (PUF) Sorbent Tubes (SKC, 226-131 Valley View Road Eighty Four, PA 15330, USA) using an AirFlowPuf Sampler and conforming with the US EPA methods TO-4A, TO-9A, TO13A, ASTM D-6209 and ISO-12884, ISO-16362 (Analitica Strumenti Samplers, via degli Abeti 144 61100 Pesaro, Italy).

The TSP were collected on glass fibre filters, and the polyurethane foam (PUF) cartridge was placed in series after the glass fibre filter. The volatile compounds, which were not trapped on the filter, were retained in the PUF cartridge.

The sampling flow was electronically controlled to be 250 L min^{-1}. Each sampling duration was controlled by a timer that was accurate to ±15 min over a 48-h sampling period. The exact flow rate was calculated daily and corrected for variations in atmospheric pressure and actual differential pressure across the filter. The filters were conditioned for 48 h and were weighed using an analytical balance (±10 μg) before and after sampling to calculate the mass of the TSP trapped on the filter. The procedures were conducted according to the European Committee for Standardization. Additionally, the PUF had been pre-cleaned by 24 h Soxhlet extractions using acetone.

2.2. Extraction and mutagenicity assays

Each sample was extracted with acetone in a Soxhlet apparatus for a minimum of 80 cycles. The samples were dried in a Rotavapor instrument, and suspended in dimethyl sulfoxide (DMSO) to obtain an equivalent concentration of 0.1 m^3 of sampled air per μL of solution. The mutagenicity assay was conducted as previously described (Maron and Ames, 1983; Traversi et al., 2009). Defined amounts of organic extract were tested to generate a dose–response curve (2, 5, 10, 20, 30 air equivalent m^3 for the TSP extracts and 10, 20, 30, 50, 100 air equivalent m^3 for the PUF extracts). The slope of the dose–response curve (revertants m^{-3}) was calculated by the least squares linear regression beginning at the first linear portion of the dose–response curve (Traversi et al., 2011). All experiments were performed in triplicate using at least three doses. The results are expressed as net revertants per cubic metre (rev m^{-3}) (the total revertants minus the spontaneous revertants) and were calculated using the dose–response curve (Cassoni et al., 2004; Claxton et al., 2004). The mutagenic activity of the airborne particulate extracts was determined using the Salmonella typhimurium strain TA98, with and without S9 mix, as well as the YG1021 strain. YG1021 is