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Chemical and biological characterization of air particulate matter 2.5, collected from five cities in China

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ABSTRACT

Fifteen polycyclic aromatic hydrocarbons (PAHs) in $PM_{2.5}$ samples collected in five different cities (Hong Kong (HK), Guangzhou (GZ), Xiamen (XM), Xi'an (XA) and Beijing (BJ)) in China in the winter 2012–12 were analyzed by gas chromatography-mass spectrometry. The biological effects of organic extracts were assayed using the human bronchial epithelial cells BEAS-2B. All sixteen priority PAHs can be found in the $PM_{2.5}$ samples of XA and BJ, but not in HK, GZ and XM, demonstrating the differential spatial source and distribution of PAHs. Our results showed that the total PAHs ranged from 3.35 to 80.45 ng/m^3 air, leading by BJ, followed by XA, XM, GZ and HK. In the cell culture study, transcript levels of pro-inflammatory cytokine interleukin-6 (IL-6), CYP1A1 and CYP1B1 were found to be induced in the treatment. The cells exposed to extracts from XA and BJ demonstrated significant migratory activities, indicating a sign of increase of tumorigenicity.

1. Introduction

Air pollution problem in Northern China has reached alarming levels. In the winter of 2012–13, transboundary air pollution from China was reported and became a global concern. A remarkable increase in the prevalence of lung cancers in the country in the past decades was found to be related to particulate matter (PM) with an aerodynamic diameter of less than 2.5 μm (Chen et al., 2013). The latest report from World Health Organization (WHO) stated that air pollution has become the world's single biggest environmental health risk, linked to around 7 million – or nearly one in eight deaths in 2012. The figures suggested that outdoor pollution from traffic fumes, coal and wood burning may kill more people than smoking and diabetes combined. Outdoor air pollution came as a result of stroke, cardiovascular and pulmonary disease and the vast majority was found to be in Asia and America. The situation worsens with the exponential growth in population together with the industrialization in this area.

It has been well known that ambient PM suspended in the atmosphere as aerosol are adversely affecting human health, especially to the respiratory and cardiovascular systems (Brunekreef et al., 2009; Langrish et al., 2012; Lee et al., 2007; Park et al., 2011). PM_{10} and $PM_{2.5}$ which are differentiated by the particulate diameters (μm) are commonly used in the air quality monitoring scheme all over the world (USEPA). Of which, $PM_{2.5}$, as compare to PM_{10}, can penetrate into the deeper area of the lung, directly affect the respiratory surfaces and dissolve into blood which may cause systemic toxic effects. According to the Environmental Protection Agency (USEPA), the ambient concentration of $PM_{2.5}$ was set as 35 μg/m^3 (24 h mean) and 12 μg/m^3 (annual mean) for national ambient air quality standard (USEPA, 2013). The pollutants which attached on the $PM_{2.5}$, which can pass all along the respiratory tract to the deeper alveolar sac, are the disease-causing reason of air pollution. Organic and inorganic pollutants were adsorbed on $PM_{2.5}$ found globally (Cachon et al., 2014; Chang et al., 2006).

Polycyclic aromatic compounds (PAHs) had been accepted as a class of ubiquitous environmental pollutants which are mostly products of energy production such as coal and petroleum, natural

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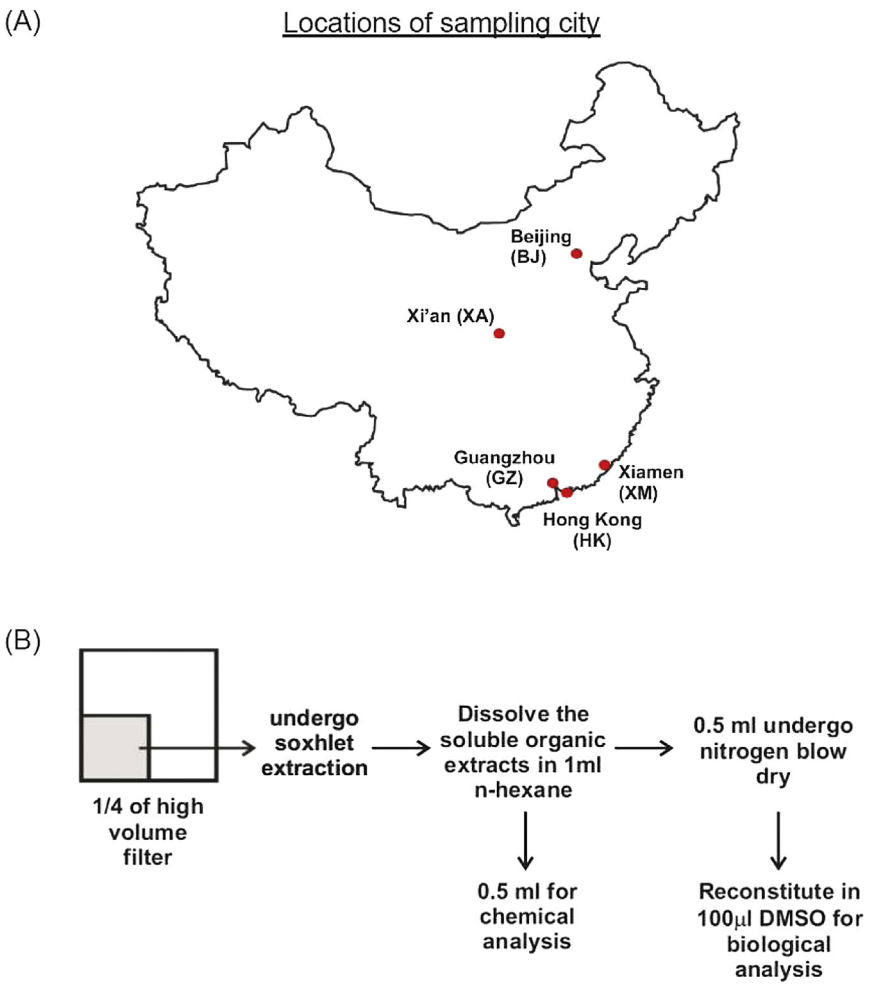

Fig. 1. Experimental set up of present study. (A) Location of five sampling cities on a China map. (B) The workflow of handling and processing the air filters. One fourth of the air filter we collected underwent soxhlet extraction and the organic extract was dissolved in n-hexane. Half of the dissolved extracts performed chemical analysis by GC–MS while the other half underwent nitrogen dry and reconstituted in 1 mL DMSO for biological analysis using cell line.

gas and fossil fuels, as well as cigarette smoking and waste incineration (Bostrom et al., 2002; Kong et al., 2010). The sources of PAHs production may varies from countries to countries; in Sweden, phenanthrene is the dominant PAHs found in 1994–2000 (Bostrom et al., 2002); while fluoranthene being the most abundant PAHs detected in Beijing, China (Wang et al., 2008). The USEPA has classified 7 PAHs as probable human carcinogens: benz(a)anthracene, benz(a)pyrene, benzo(b)fluoranthene, benzo(k)fluoranthene, chrysene, dibenz(a,h)anthracene and indeno(1,2,3-cd)pyrene.

Due to its small size, $PM_{2.5}$ is able to penetrate to the lower respiratory tract to alveolar sacs and induce cell damage. There are two major routes of inhaled PAHs to the blood circulations. The majority of the PAHs deposits on the thinner alveolar epithelium where they readily absorbed and enter the circulation; while about 10–20% of PAHs inhaled will deposit on the thicker bronchial epithelium, slowly absorbed and transport through the epithelium. Due to the lipophilic properties of most PAHs, a fraction of compounds can be retained in the bronchial tissues and accumulated to attain a high local dose even at low environmental exposure levels (Bostrom et al., 2002). Industrial development and exponential growth in population in China raises the needs of energy and subsequent different kind of pollutions to the environment. On Feb 2014, Beijing recorded dangerously high level of suspended $PM_{2.5}$ for over 15 times of the recommended levels of WHO limits (25 μg/m^3 24 h mean $PM_{2.5}$) for consecutive 6 days (Chen et al., 2013). This leads to our concern to the air quality monitoring and its potential health effects to lung, which is the first organ encountered with the suspended pollution in air. Thus, in this study, human bronchial epithelial Beas-2b cell line was used to evaluate the cytotoxicity of $PM_{2.5}$ collected from the five cities in China.

2. Materials and methods

2.1. Air sample collection

Two northern (BJ and XA) and three southern (GZ, XM and HK) Chinese cities were selected in the present study (Fig 1A). The samples were taken for six to eight days during the air pollution episode of haze from the end of January to the beginning of February in 2013. The $PM_{2.5}$ samples were collected on quartz fiber filter (8 inch × 10 inch) using a high-volume sampler at a flow rate of 1.05–1.16 m^3 min^{-1}. Seven to eight air filters were collected from each sampling

Table 1
List of primers used in this study.

Primer	Forward primer sequences	Reverse primer sequences
CYP1A1	AGCAGCTGGATGAGAACGCC	GCCGTGACCTGCCAATCACT
CYP1B1	TTGTGCCTGTCACTATTCCTC	ATCAAAGTTCTCCGGGTTAGG
TNF-α	GGGCCTGTACCTCATCTACT	TAGATGGGCTCATACCAGGG
IL-6	AGCCCACCGGGAACGAAAGA	TGTGTGGGGCGGCTACATCT
IL-8	AAGCCACCGGAGCACTCCAT	CACGGCCAGCTTGGAAGTCA
h-Actin	GACTACCTCATGAAGATCCTCACC	TCTCCTTAATGTCACGCACGATT