Influence of Vehicular Transport on Air Pollution in Oporto, Portugal: Particulate-bound Polycyclic Aromatic Hydrocarbons

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Abstract — Considering particulate matter among the health-damaging air pollutants, the objective of this work was to evaluate the influence of road transport on levels of fine ($PM_{2.5}$) and coarse ($PM_{2.5-10}$) particles in urban area and to assess its polycyclic aromatic hydrocarbon content and the associated health risks. Samples were collected for period of 40 days during 2007 at an urban site situated in Oporto metropolitan area in Portugal; 17 PAHs recommend by U.S. EPA as priority pollutants were quantified using microwave assisted extraction combined with liquid chromatography. The results showed that $PM_{2.5}$ and $PM_{2.5-10}$ daily means were 28.7 ± 9.8 μg m^{-3} and 13.9 ± 7.9 μg m^{-3}, respectively. The mean concentration of 17 particulate-bound PAHs (Σ_{PAHs}) was 13.3 ± 10.0 ng m^{-3} in $PM_{2.5}$ and 1.0 ± 0.3 ng m^{-3} in $PM_{2.5-10}$; PAHs with 5-6 aromatic rings were the most abundant compounds in $PM_{2.5}$ accounting for 70% of Σ_{PAHs}. The estimated values of lifetime lung cancer risks considerably exceeded (220 and 44 times) the health-based guideline level. Finally, the results showed that evaluation of benzo[a]pyrene (regarded as a marker of the genotoxic and carcinogenic PAH) alone would underestimate the carcinogenic potential of the studied PAH mixtures.

Keywords — Air pollution, Health risks, PAHs, PM

1 Introduction

Climate change is one of the greatest environmental, social and economic threats of nowadays. It is likely that most of the warming can be attributed to the greenhouse gas emissions caused by human activities, transport being one of the most important ones. To mitigate climate changes, reduction of emissions from transport must clearly be part of a global commitment. The European Union is adamant about addressing emissions from all forms of transport including international maritime and aviation sectors that are currently not covered by the Kyoto Protocol. Concurrently, it is expected that climate change mitigation efforts will also improve air quality. In that regard, particulate matter (PM) is of interest as it is considered among the most health-damaging air pollutants.

Particulate matter is a complex mixture of solid and/or liquid particles suspended in the air [1]. The particles vary considerably in their origin, shape, size, and chemical properties; PM consists of many organic and inorganic substances, covering a wide range of particle diameters, from <0.1 μm up to some 100 μm. It is common to classify the particles by their aerodynamic diameter and in that regard PM tends to be divided into two principal groups [2]: coarse particles ($PM_{2.5-10}$) that are larger than 2.5 μm and lesser than 10 μm in aerodynamic diameter, and fine particles ($PM_{2.5}$) that are smaller than 2.5 μm in aerodynamic diameter.

The reason why PM gained so much attention was is due to its health implications. Various epidemiological studies showed a strong association between exposure to increased levels of PM and increases in mortality and morbidity rates caused by pulmonary and cardiovascular diseases [3-6]. Furthermore, the more recent studies of air pollution impacts estimated that in Europe hundreds of thousands people annually die prematurely due to the outdoor air pollution caused by fine particulate matter ($PM_{2.5}$) alone [7]. The need for further reduction of this pollution is essential as even the lowest concentrations of particulates in air can cause adverse health effects. Therefore, the European Union made several attempts. Although the implementation of some policy measures led to reduction of PM emissions for approximately 20% during the last decade, ambient PM levels remained largely the same [7], possibly also due to the increased temperature caused by climate change that affects air quality. Continuing its efforts to improve air quality, in 2008 the European Union revised the current legislation and issued a new Directive 2008/50/EC [8]. Despite the fact that the values of daily and annual of PM_{10} (i.e. particles smaller than 10 μm in aerodynamic diameter) current limits themselves have not changed, all the modifications

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made in this legislation led to reduction of the public health protection compared to the levels of protection given in the previous directive. However, for Europe the directive still represents a significant step forward as for the first time $PM_{2.5}$ standards were promulgated. As the values of proposed $PM_{2.5}$ standards will be revised in 2013, the European Union consequently urges research in the respective area.

Composition of particulate matter is as nearly as important as levels of particles themselves as it has been shown that it also contributes to PM adverse health effects [9]. Health-hazardous PM components, such as carcinogenic metals (arsenic, cadmium, and nickel), or polycyclic aromatic hydrocarbons (PAHs) have gained the highest interest. The health concerns of PAHs have been traditionally focused on their potential cytotoxicity, mutagenicity and carcinogenicity in humans [10]. PAHs are genotoxic compounds and their carcinogenicity is probably mediated by their ability to damage the DNA [11]. Especially PAHs associated with particulates matter are potentially harmful to humans, because they are the ones with highest molecular weights, and they are the most carcinogenic [12]. As the carcinogenic potency of individual PAH widely varies, U.S. Environmental Protection Agency (U.S. EPA) listed 16 priority PAH compounds [13], with benzo[a]pyrene being classified by International Agency for Research on Cancer (IARC) as known carcinogen to humans [14], whereas other PAHs are considered as probable and possible human carcinogens [14], [15]. Out of 16 compounds, benzo[a]pyrene is probably the most known PAH carcinogen. In many studies that estimate human cancer risk benzo[a]pyrene is used as a surrogate for other carcinogenic PAHs. Even current European legislation for ambient air [16] uses benzo[a]pyrene as indicator for carcinogenic PAHs. Nevertheless, the suitability of this approach started to be questioned [17] by new findings on the presence of more potent PAHs, such as dibenzo[a,l]pyrene or dibenz[a]anthracene that are estimated to have a carcinogenic potency approximately 100 and 5 times higher, respectively, than benzo[a]pyrene [18].

As traffic emissions are one of the biggest sources of fine particles, more information on PM need to be provided, namely concerning the levels and carcinogenic PAH composition. Nevertheless, there is a great inconsistency in evaluation carcinogenic PAHs, as previous studies mostly focused on benzo[a]pyrene, omitting other carcinogenic compounds. Thus, aiming to further understand the impact of traffic particulate matter on public heath, this study evaluated the influence of vehicular emissions on $PM_{2.5}$ and $PM_{2.5-10}$, considering: (1) concentrations; (2) carcinogenic PAH composition; and (3) associated health risks. The developed work includes the detailed characterization of $PM_{2.5}$ and $PM_{2.5-10}$ sampled at one urban site directly influenced by traffic emissions. The 17 PAHs (16 PAHs considered by U.S. EPA as priority pollutants, and dibenzo[a,l]pyrene) including 9 carcinogenic ones were quantified by microwave-assisted extraction combined with liquid chromatography.

2 MATERIALS AND METHODS

2.1 Sample Collection

PM_{10} and $PM_{2.5}$ bound PAHs were collected in north of Portugal in Oporto city for a period of 40 days during winter 2007, at an urban site (latitude 41° 10' 40" N, longitude 8° 35' 54" W and altitude 121 m). The site was close to an access point of one of the most important high-ways connecting Lisbon and Oporto, being also the main road connection to the north of Spain. Other two major thoroughfares that provide the traffic connection within the city as well as with surrounding towns also pass through this area; traffic emissions are the main source of atmospheric pollutants in the area [19-21].

The particulate samples were collected daily for a period of 24 hours by TCR TECORA Bravo H2 constant flow samplers, combined with PM sampling heads in compliance with the norm EN12341 (PM_{10}) and EN14907 ($PM_{2.5}$) using air flow rate of 2.3 m^3 h^{-1}. The different fractions of particles, i.e. PM_{10} and $PM_{2.5}$, were collected on polymethylpentene support ring (2 μm porosity, ∅47 mm, SKC Ltd.).

PM masses were determined gravimetrically by subtracting the initial average mass of the blank filter from the final average mass of the sampled filter. The filters were stored in Petri dishes and the same analytical balance (Mettler Toledo AG245) was always used. The steps of gravimetric mass determinations were the following: 24 h to equilibrate filters before weighing at room temperature, followed by weighing during the following 24-48 h. The filters were repeatedly weighed until three reproducible values were obtained. After sampling filters were immediately weighed, stored in Petri dishes covered in parafilm, and kept in freezer (-18 °C) until they were further analysed.

Dibenzo[a,l]pyrene and more 16 PAHs identified as priority pollutants by U.S. EPA were determined in the collected particulate samples: naphthalene (NA), acenaphthylene (ACY), acenaphthene (ACE), fluorene (FLUOR), phenanthrene (PHEN), anthracene (AN), fluoranthene (FLUR), pyrene (PY), benz[a]anthracene (B[a]A), chrysene (CHRY), benzo[b]fluoranthene (B[b]F), benzo[k]fluoranthene (B[k]F),, benzo[a]pyrene (B[a]P, dibenz[a,h]anthracene [D[a,h]A), benzo[ghi]perylene (B[ghi]P), and indeno[1,2,3-cd]pyrene (IN).

2.2 PAH Extraction and Quantification

The extraction of PAHs from particles was performed by Microwave Assisted Extraction (MAE; MARS-X 1500 W Microwave Accelerated Reaction System for Extraction and Digestion, CEM, Mathews, NC, USA) that was previously optimised [22]. Filters were transferred to the glass extraction vessels with 30 mL of acetonitrile (Sigma-Aldrich) and the MAE of both PM_{10} and $PM_{2.5}$ was performed for 20 min at 110 °C. After the extraction vessels were allowed to cool at room temperature, extracts were carefully filtered through a PTFE membrane filter (0.45 μm) and reduced to a small volume using a rotary evaporator (Buchi Rotavapor, R-200) at 20 °C. A gentle stream of nitrogen was used to dry the extracts under low temperature; the residue was then re-dissolved in 1000 μL of acetonitrile immediately before analysis.

To quantify PAHs, extracts were analysed using a Shimadzu LC system (Shimadzu Corporation, Kyoto, Japan) equipped with a LC-20AD pump, DGU-20AS degasser and photodiode array SPD-M20A (PAD) and fluorescence RF-10AXL (FLD) detectors on line. Separation of the compounds was performed in a C18 column (YMC, MP-PAH C18, 50×4.0 mm; 3 μm particle size) and the injected volume was 15.0 μL. A mixture of water (ultra-pure grade, prepared by Milli-Q simplicity 185 Millipore system, Millipore, Molsheim, France) and acetonitrile was used as the mobile phase. The initial composition of the mobile phase was 45% of acetonitrile and 55% ultra-pure water, and a linear gradient to 100% of acetonitrile was programmed in 15 min, with a final hold of 7 min. Initial conditions were reached in 3 min and maintained for 10 min before next run. The total run time was 35 min with a flow rate of 0.8 mL min^{-1}. Fluorescence wavelength programming was used to perform better sensitivity and minimal interference. Each compound was detected at its optimum excitation/emission wavelength pair: 260/315 nm (naphthalene, acenaphthene and fluorene), 260/366 nm (phenanthrene), 260/430 nm (anthracene, fluoranthene, pyrene, benz[a]anthracene, chrysene, benzo[b]fluoranthene, benzo[k]fluoranthene, benzo[a]pyrene, dibenz[a,h]anthracene, benzo[ghi]perylene and dibenzo[a,l]pyrene), and 290/505 nm (indeno[1,2,3-cd]pyrene). Acenaphthylene, which does not show fluorescence, was analysed at 254 nm in PAD. Each analysis was performed at least in triplicate.

All materials used for MAE-LC analysis were regularly cleaned in acetone (Fluka, commercial grade), and hexane (supplied by Valente & Ribeiro, commercial grade).

The overall analytical procedure was previously validated by systematic recovery experiments using the standard reference material 1650b "Diesel particulate matter". Under the microwave-assisted extraction conditions, PAHs were extracted from particulate matter with recoveries ranging from 81.4 ± 8.8% to 112.0 ± 1.1%, for all the compounds except for naphthalene (62.3 ± 18.0%) and anthracene (67.3 ± 5.7%). Regarding the reproducibility of the optimised methodology, expressed as relative standard deviation, values were lower than 9% for all the targeted contaminants except for naphthalene (18%). Limits of detection (LODs) and limits of quantification (LOQs) were calculated and expressed as PAH concentration in solution (μg L^{-1}) and in air samples [22]. LODs between 0.0016 ng m^{-3} (0.090 μg L^{-1}) for benz[a]anthracene and 0.027 ng m^{-3} (1.5 μg L^{-1}) for naphthalene were obtained, with corresponding LOQs in the range 0.0054-0.089 ng m^{-3} (0.30-4.90 μg L^{-1}).

2.3 Health Risks Analysis

To assess the risks associated with inhalation exposure to PAHs, the toxicity equivalency factors (TEF) [10], [23] were used; consequently the corresponding lifetime lung cancer risks were estimated [24].

2.4 Statistical Analysis

For the data treatment, the Student's t-test was applied to determine the statistical significance ($P<0.05$, two tailed) of the differences between the means determined for both sites.

3 Results and Discussion

3.1 Particles and Particulate-bound PAH Levels

The mean concentration of PM_{10} ranged from 15.4 to 91.0 μg m^{-3} being fractioned into $PM_{2.5}$ with a daily mean of 28.7 ± 9.8 μg m^{-3} and $PM_{2.5-10}$ (i.e. particles with aerodynamic diameter between 2.5 and 10 μm) with mean of 13.9 ± 7.9 μg m^{-3}. On average, $PM_{2.5}$ fraction accounted for approximately 67% of PM_{10}. In general the obtained results were in agreement with other aerosol studies performed in Oporto Metropolitan Area [20], [21], [25] and in other European and non-European urban environments [24], [26].

The total concentration of 17 PAHs in $PM_{2.5}$ and $PM_{2.5-10}$ was 13.3 ± 10.0 ng m^{-3} and 1.0 ± 0.3 ng m^{-3}, respectively. The levels of PAH groups with 2-6 rings associated with both PM fractions are shown in Fig. 1.

From this figure it is clear, that lower molecular weight PAHs (i.e. with 2 rings), such as naphthalene, corresponded to less than 1% of Σ_{PAHs} in $PM_{2.5}$ probably due to their predominant presence in gas phase [27], [28].

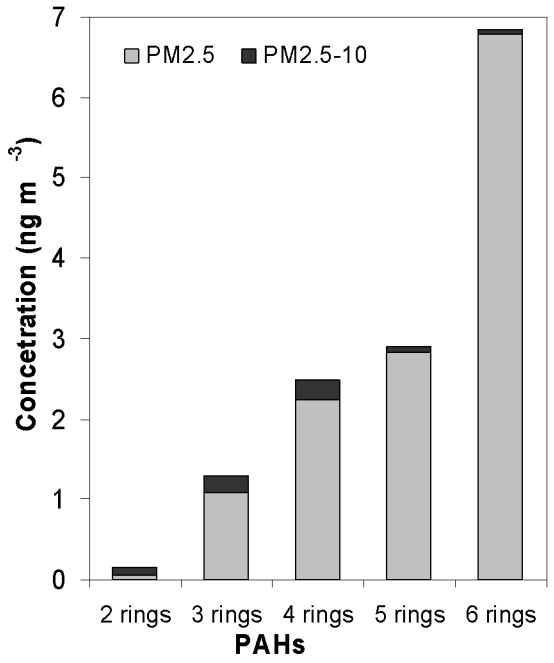

Fig. 1. Concentrations of PAHs associated with $PM_{2.5}$ and $PM_{2.5-10}$. Concentrations are presented as sums of individual compounds according to the number of aromatic rings, i.e. groups with 2, 3, 4, 5 and 6 rings, respectively.

On the contrary, compounds with 5-6 rings that have high molecular weight (i.e. benzo[b]fluoranthene, benzo[k]fluoranthene, benzo[a]pyrene, dibenz[a,h]anthracene, benzo[ghi]perylene, indeno[1,2,3-cd]pyrene, and dibenzo[a,l]pyrene) contributed about 70% of Σ_{PAHs} in $PM_{2.5}$, being primarily associated with particulate phase [29]. Dibenz[a,h]anthracene was the most abundant compound $PM_{2.5}$. This compound reached mean concentration of 4.27 ± 2.13 ng m^{-3}, respectively, accounting for approximately 30% of Σ_{PAHs}. Its percentage was considerably higher than those for other abundant $PM_{2.5}$-bound PAHs, that were, by descending order: benzo[b]fluoranthene and indeno(1,2,3-cd)pyrene (approximately 10%), benzo[ghi]perylene (approximately 8%), and benzo[a]pyrene and chrysene (approximately 7%). According to several studies [29-31], those PAHs (dibenz[(a,h]anthracene, benzo[b]fluoranthene, indeno(1,2,3-cd)pyrene, benzo[ghi]perylene, benzo[a]pyrene and chrysene) are considered as indicators of traffic emissions, suggesting that these emissions were the major source of PM pollution at the selected urban site.

The results in Fig. 1 also show that particulate PAHs were predominantly associated with particles of smaller sizes, i.e. $PM_{2.5}$. On average PAHs associated with $PM_{2.5}$ accounted for 93% of Σ_{PAHs} thus showing the importance of fine particles. Consequently, PAHs on particles bigger than 2.5 μm accounted for only 7.0%. These results are in agreement with previous studies [12], [28], [32]. These findings are especially health-relevant because fine particles cause many adverse health effects including cardiopulmonary diseases and lung cancer. Although, it is not yet clear if it is the physical or the chemical PM characteristics, or both, that are responsible for the health effects, PAHs present in these fine particles may significantly contribute to (or eventually even enhance) those adverse health effects [33], [34]. It is, however, necessary to point out that PAHs in $PM_{2.5-10}$ had different contribution profiles (Fig. 1.). Compounds with 3 and 4 rings were the most abundant PAHs in $PM_{2.5-10}$ accounting for 31 and 34% of Σ_{PAHs}, respectively. The highest levels were observed for anthracene (3 rings) and pyrene (4 rings) that in $PM_{2.5-10}$ reached mean concentrations of 0.160 ± 0.80 and 0.102 ± 0.63 ng m^{-3}, respectively. These results thus indicate that coarse particles collected at the urban site might originate from other emission sources [20], [21], [35], [36].

In order to confirm if particulate PAH emissions originated from traffic, diagnostics ratios for PAHs emitted by traffic were calculated (Table 1). Typically the value of phenanthrene/(phenanthrene+anthracene) ratio higher than 0.70 and of benzo[a]pyrene/benzo[ghi]perylene ratio lower than 1.0 indicate that PAHs are emitted by traffic [30], [37]; specifically, a ratio of indeno[1,2,3-cd]pyrene/(indeno[1,2,3-cd]pyrene+benzo[ghi]perylene) higher than 0.30 indicates significant emissions from diesel vehicles [30] and these emissions are also indicated by benzo[a]pyrene/benzo[ghi]perylene ratio with values between 0.46-0.81 [37].

Table 1. Diagnostic ratios of PAHs emitted by traffic

Ratios	Range	$PM_{2.5}$	$PM_{2.5-10}$
PHEN/(PHEN+AN)	> 0.70	0.74	0.39
B[a]P/B[ghi]P	< 1.0 0.46-0.81	0.80	1.04
IN/(IN+B[ghi]P)	> 0.30	0.55	0.28

As it can be clearly seen in Table 1, for $PM_{2.5}$ all ratio values were within the required range. These results thus corroborate the previous findings confirming that traffic emissions were the major source of PAHs. On the contrary, PAHs in $PM_{2.5-10}$ did not fit the required interval, suggesting another emission sources for this fraction; as previously shown emissions from a power plant situated nearby the Oporto Metropolitan Area also contribute to the respective PM levels [20], [21], [35].

3.2 Health Risks

To evaluate the health risks associated with exposure to particulate PAHs, TEF approach was applied. TEF

values reported by Nisbet and LaGoy [38] and Okona-Mensah et al. [18] were used to calculate the TEF-adjusted concentrations (based on benzo[a]pyrene) of the determined 17 PAHs. The evaluation was performed for PAHs in $PM_{2.5}$ only, as this fraction contained majority of the PAH content. Furthermore, acenaphthylene and acenaphthene were detected in less than 10% of samples; therefore their detailed study was not done. The results are shown in Table 2.

Table 2. TEF adjusted concentrations for 17 PAHs associated with $PM_{2.5}$ (pg m^{-3})

Compound	TEF	$PM_{2.5}$ x 10^1
Naphthalene	0.001	0.519
Fluorene	0.001	0.314
Phenanthrene	0.001	2.75
Anthracene	0.01	76.9
Fluoranthene	0.001	2.23
Pyrene	0.001	4.85
Benz[a]anthracene	0.1	647
Chrysene	0.01	88.2
Benzo[b]fluoranthene	0.1	1370
Benzo[k]fluoranthene	0.1	512
Benzo[a]pyrene	1	9330
Dibenzo[a,l]pyrene	100	21 800
Dibenz[a,h]anthracene	5	213 800
Benzo[ghi]perylene	0.01	113
Indeno[1,2,3-cd]pyrene	0.1	1340
∑PAHs	-	250 000

Dibenz[a,h]anthracene was previously the most abundant PAH. The levels of this compound ranged from 0.270 to 10.5 ng m^{-3} and on average it represented 32% of Σ_{PAHs}. Due to its TEF of 5, dibenz[a,h]anthracene remained the largest contributor to $\Sigma_{TEF\text{-}PAHs}$, with 85%. On the contrary, dibenzo[a,l]pyrene appeared previously in $PM_{2.5}$ in the lowest levels (mean of 21.8 ± 16.7 pg m^{-3}, i.e. less than 0.2% of Σ_{PAHs}), nevertheless due to its high TEF (100) it was the second largest contributor to $\Sigma_{TEF\text{-}PAHs}$ contributing 8.7%. These results confirm and emphasize the importance of the analysis and evaluation of these two potent carcinogens that are being currently discussed as possible surrogate compounds for PAH mixtures from various environments [18]. Furthermore, the associated health risks of PAHs in PM are expected to be higher than those predicted by an additive model of these two pollutants.

The values of $\Sigma_{TEF\text{-}PAHs}$ were used to estimate the corresponding lifetime lung cancer risks for exposed groups. Regarding the lung cancer risk via the inhalation route, the World Health Organization suggested the unit risk of 8.7×10^{-5} (ng m^{-3})$^{-1}$ for lifetime (70 years) PAH exposure [24]. Thus, the corresponding lifetime lung cancer risk was 2.2×10^{-3}. Taking into the consideration that people spent outdoors only 20 to 25% of their time, the value of lifetime lung cancer risk lowered to 4.4×10^{-4}. It is however important to point out that both values exceeded the health-based guideline level of 10^{-5} (approximately 220 and 44 times, respectively) [39]. These estimations show that despite the limited time spent outdoors, PAHs from outdoor sources and especially from traffic emissions represent a serious risk to public.

4 Conclusions

Pollutants from traffic emissions may promote serious problems, thus in order to protect public health it is fundamental to reduce traffic emissions, mainly in relation to particulate pollution.

The mean concentration of 17 PAHs (Σ_{PAHs}) associated with $PM_{2.5}$ was 13.3 ± 10.0 ng m^{-3}. This fraction contained 93% of the total PAH content. PAHs with 5-6 aromatic rings were the most abundant compounds in $PM_{2.5}$ accounting for 70% of Σ_{PAHs}. The diagnostic ratios confirmed that traffic emissions were the major source of $PM_{2.5}$ and its PAH content.

The total concentration of 17 compounds associated with $PM_{2.5\text{-}10}$ was 1.0 ± 0.3 ng m^{-3}, with 3-4 rings PAHs being the most abundant ones (61% of Σ_{PAHs}); these particles originate from other emission sources.

The estimated values of lifetime lung cancer risks considerably exceeded the health-based guideline level. Furthermore, the results showed that evaluation of benzo[a]pyrene (regarded as a marker of the genotoxic and carcinogenic PAH) alone would probably underestimate the carcinogenic potential of the studied PAH mixtures.

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