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Individual exposure level following indoor and outdoor air pollution exposure in Dakar (Senegal)

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Abstract

The consequences of indoor and outdoor air pollution on human health are of great concern nowadays. In this study, we firstly evaluated indoor and outdoor air pollution levels (CO, CO₂, NO, NO₂, PM₁₀) at an urban site in Dakar city center and at a rural site. Then, the individual exposure levels to selected pollutants and the variations in the levels of biomarkers of exposure were investigated in different groups of persons (bus drivers, traders working along the main roads and housemaids). Benzene exposure levels were higher for housemaids than for bus drivers and traders. High indoor exposure to benzene is probably due to cooking habits (cooking with charcoal), local practices (burning of incense), the use of cleaning products or solvent products which are important emitters of this compound. These results are confirmed by the values of S-PMA, which were higher in housemaids group compared to the others. Urinary 1-HOP levels were significantly higher for urban site housemaids compared to semirural district ones.

Moreover, urinary levels of DNA oxidative stress damage (8-OHdG) and inflammatory (interleukin-6 and -8) biomarkers were higher in urban subjects in comparison to rural ones.

The air quality measurement campaign showed that the bus interior was more polluted with PM₁₀, CO, CO₂ and NO than the market and urban or rural households. However, the interior of households showed higher concentration of VOCs than outdoor sites confirming previous observations of higher indoor individual exposure level to specific classes of pollutants.

Keywords: Air pollution; Volatile organic compounds exposure; Biomarkers of exposure; Indoor and outdoor air pollution exposure; Personal exposure evaluation.

Capsule: Outdoor and indoor air pollution analysis and human biomarker exposure evaluation

Introduction

Indoor and outdoor air pollution is among the main causes of morbidity and mortality in the world (Cohen et al., 2017; IARC, 2014; Merbitz et al., 2012; Raaschou-Nielsen et al., 2013). The assessment of environmental exposure to air pollution is, therefore, a major challenge for understanding and evaluating the potential impact of this pollutant in exposed population. Road traffic is an important emission source of various organic compounds (such as volatile compounds - VOCs, like benzene, toluene, xylenes, and polycyclic aromatic hydrocarbons - PAHs) and others compounds deriving mainly from vehicle exhausts. Outdoor VOCs and PAHs extensively affect outdoor air quality and are mainly generated during the incomplete combustion of fuels (both gasoline and diesel), while indoor VOCs are related to specific sources which determines the type of molecules observed in confined environments (Sahlberg et al., 2013; Salthammer, 2016). Many of these organic compounds are known to be carcinogens (IARC, 2014; Filippini et al., 2015) and to cause pulmonary and cardiovascular diseases and immune system impairments (Alshaarawy et al., 2016; Haussmann, 2012). Since the majority of the studies on air pollution and related human exposure has been conducted in developed countries (Maitre et al., 2002; Schneider et al., 2001; Smith et al., 2007; Sorensen et al., 2003), there is a urgent need of similar information from studies performed in developing countries and especially from those areas (such as the sub-Saharan Africa) where, in the last years, an uncontrolled urbanization and old fleets of circulating vehicles generate a significant emission of air pollutants (Assamoi & Liousse, 2010; Baumbach et al., 1995). Dakar, the capital of Senegal, can be considered representative of these conditions. In Dakar the circulating vehicles are mainly characterized by old and inefficient combustion technologies. As a consequence, higher concentration of air pollutants are registered alongside roadways, and the individuals working along the major streets of the city may incur in substantial exposures to traffic-related air pollution (Knibbs & Morawska, 2012). Beside outdoor air pollution, in the last years specific attention has been paid also to indoor air pollution and the consequent human exposure (Huang et al., 2019). Although the relation between outdoor and indoor air pollution, which vary depending on factors that can influence the indoor levels, e.g. climate, emission sources, human activity and building ventilation (Monn, 2001), it is well recognize that indoor emissions may greatly influence building air quality (Huang et al., 2019; Tang et al., 2016). Epidemiological investigation may provide important information on the actual exposure of a population thanks to the selection of biomarkers of exposure/effect. Indeed biomarkers may reflect the total exposure of an individual to a selected (class of) pollutant. Several studies

used the urinary concentration of selected metabolites of organic compounds and/or other biomolecules to show a significant increase of these biomarkers in population close to the specific source of emission, such as traffic exhausts (Amodio-Cocchieri et al., 2001; Arayasiri et al., 2010; Fustinoni et al., 2005).

Among the different biomarkers of exposure S-phenylmercapturic acid (S-PMA), muconic acid (t,t-MA) and catechol are representative of the metabolic transformation of benzene, (Snyder et al., 1993; Waidyanatha et al., 2001). The urinary excretion of S-PMA and t,t-MA has been used in epidemiological investigation in occupational and environmental studies when benzene was considered the primary pollutant of interest (Ayi Fanou et al., 2006; Borgie et al., 2014; Fustinoni et al., 2005). Unfortunately, under environmental conditions, people are exposed to several airborne compounds among which also PAHs (Fan et al., 2012). One of the biomarkers normally used to evaluate the exposure to PAHs mixtures is the urinary concentration of 1-hydroxypyrene (1-OHP) (Ovrebo et al., 1995). Although biomarkers of exposure give significant information on the interaction of the selected population with the pollutants of interest, it is also important to evaluate also the levels of markers representative of the possible effects induced by air pollutants. Oxidative stress is considered a significant process of damage activated by air pollutants, such as PAHs and VOCs. Uncontrolled oxidative stress may in turn determine the oxidative damage of biological macromolecules, such as DNA, RNA and phospholipids. A largely used biomarker of oxidative damage is the level of 8-hydroxy-2'-deoxyguanosine (8-OHdG) in urine. This molecule results from oxidation of the DNA base guanine and has been already proposed as biomarker to evaluate the damages determined by ambient air pollution (Svecova et al., 2009) and indoor air pollution (Commodore et al., 2013). Finally, among the processes triggered by air pollution, lung epithelia inflammatory responses are considered of primary importance (Podechard et al., 2008; Umannova et al., 2011; Vogel et al., 2005) for understanding the health impacts of airborne pollutants. The inflammatory mediators, such as tumor necrosis factor alpha (TNF- α), interleukin-6 (IL-6), interleukin-8 (IL-8), may be released also in the systemic circulation and finally excreted in urine (Sawyer, 2010; Van Eeden et al., 2001) and may therefore serve as additional biomarkers of the negative effects of air pollutants on exposed population.

The aim of this study was therefore to assess, for the first time in a sub-Saharan region, the individual exposure level to air pollution, mainly volatile organic compounds (namely benzene, toluene, ethylbenzene and xylene – BTEX) and PM, and its potential health impact, by the quantification of selected biomarkers of exposure/effects. The biomarkers were quantified in groups of person representative of professionals (bus drivers and traders)

exposed mainly to outdoor air pollution and of housemaids mainly affected by indoor air pollution, selected in Dakar city center. Moreover the effects of indoor air pollution on the levels of biomarker of exposure/effects were also evaluated in a group of housemaids working in a semi-rural area located 45 km from the city center to evaluate possible differences between urban and non-urban areas on indoor air quality and related potential effects.

1. Materials and methods

1.1. Study population

This cross-sectional study took place in the city of Dakar, at the urban district of HLM where the road traffic is particularly intense and at a semirural district, Toubab Dialaw (TD), that is located 45 Km west from the urban site (**Fig. S1**).

A non-probability sampling technique was used to select subjects. 57 professionals and 59 housemaids were selected. Professionals were representative of bus drivers (27 participants included) and traders (30 participants located along the street where traffic is more intense and in the main market of HLM). These two categories were considered exposed mainly during their working hours and mainly to outdoor air pollution. Housemaids (29 recruited at the urban site and 30 at rural site) were selected as working exclusively inside the houses during their working hours and therefore exposed mainly to indoor air pollution (for more details see supplementary material).

1.2. Sample collection

Individual exposure to BTEX (benzene, toluene, ethylbenzene and xylene) was monitored from March 13 to 31, 2017 using badges GABIE (Gas Adsorbent Badge for Individual Exposure). These passive samplers were worn by the subjects, as close as possible to the inhalation area (mouth/nose), daily during 8 h of the working time and then kept at -80 °C until chemical analysis. Urine spot samples were collected by each subject at the end of the 8 h of monitoring period each day for the determination of S-phenylmercapturic acid (S-PMA), trans, trans-muconic acid (t,t-MA), 1-hydroxypyrene (1-OHP), 8-hydroxy-2'-deoxyguanosine (8-OHdG) and inflammatory cytokines (TNF- α , IL-1 β , IL-6 and IL-8). Each urine sample was divided into aliquots and quickly frozen at -80 °C until analyses.

1.3. Analysis of BTEX

BTEX were desorbed from the activated charcoal by using 5 mL of carbon disulfide under ultrasound for 15 min. The extract was transferred into a 2 mL vial then analyzed by gas chromatography (CP-3800, Varian USA) coupled to a flame ionization detection as reported

in Liu et al. (2009). Xylenes were resolved as ortho (o-) and meta (m-) + para (p-) xylene. As a result of the selected column used for the chromatography procedure, m- xylene and p- xylene were not discriminated.

1.4. Urinary metabolites

1.4.1. S-phenylmercapturic acid (S-PMA)

Urinary S-PMA was analyzed using a Waters Acquity ultra-performance liquid chromatography (UPLC) coupled with a tandem mass spectrometer detector (Waters XEVO TQS Waters, Manchester, UK). The analysis was performed at 40 °C using a gradient elution of ammonium formate and acetonitrile (**Table S1**). Results were collected by TargetLynx 4.1 software (Waters) (for more details see supplementary material).

1.4.2. Trans,trans-muconic acid (t,t-MA)

The t,t-MA was extracted from urine samples by a semi-liquid phase extraction using disposable cartridges (Varian bond elut SAX 500 mg 3 mL) and quantification of the benzene metabolite was carried out by a high pressure liquid chromatography (HPLC) coupled with a UV spectrometer (ALLIANCE 2695, Waters, Watford, UK) coupled with a PDA UV detector (2998 WATERS) at 259 nm wavelength (for more details see supplementary material).

1.4.3. 1-Hydroxypyrene (1-HOP)

Determination of 1-HOP was performed according to by Li et al. (2005) using a Waters Acquity ultra performance liquid chromatography (UPLC) coupled with a detector Waters XEVO TQS tandem mass spectrometer (Waters, Manchester, UK). Results were acquired by TargetLynx software 4.1 (Waters) (for more details see supplementary material).

1.4.4. 8-Hydroxy-2- α -désoxyguanosine (8-OHdG)

8-OHdG concentration in urine was studied using a commercially available enzyme immunoassay (Highly Sensitive 8-OHdG Check, Gentaur France SARL), as published elsewhere (Leclercq et al., 2017).

1.4.5. Urinary creatinine

Values of urinary metabolites were related to those of creatinine, as evaluated by the Jaffé method using a Roche Diagnostics kit (Roche Diagnostics, France). Samples with urinary creatinine < 350 mg/L were excluded from the study.

1.5. Cytokines concentration measurement

TNF- α , IL-1 β , IL-6 and IL-8 concentrations in urine were determined using commercially available enzymatic immunoassays (Milliplex[®]Map, Human Cytokine/Chemokine Magnetic Bead Panel Luminex[®], EDM Millipore, USA), scrupulously following the manufacturer's instructions. The results were expressed as pg.mL⁻¹.

1.6. Air quality analysis by EVM-7 and AQ Pro

The campaign of the air quality measurement in Dakar was held from November 11, 2017 to March 30, 2018. Two different devices were adopted: the EVM-7 environmental monitor and the AQ Pro indoor air quality monitor. The EVM-7 (version 1.05 Quest Technologies) is a compact environmental monitor for the measurement of the airborne concentrations of PM₁₀ and gases (CO₂ and CO). It also measures meteorological parameters such as humidity and temperature. A simultaneous, multi-sensor gas detector, AQ Pro indoor air quality monitor, was configured to detect the concentration of NO and NO₂ in ppm.

As a first step, we measured the level of air quality at the four main roads of the HLM market, at the rate of 3 measurements per axis and in the 27 buses involved in the campaign. In a second step, we simultaneously assessed the indoor and outdoor air quality levels of the 29 households at urban site (HLM) and the 30 households at rural site (TD). The devices were placed about 1 m above the ground, in the living room (inside) and on the sidewalk at the house's door (outside). The measurements were carried out for 8 hours.

1.7. Statistical analysis

Data processing was performed with the statistical analysis software SPSS for windows, v.22.0.0, Oct. 2016: p-values less than 0.05 were considered significant. One-factor ANOVA followed by Tukey tests were used to compare means values obtained between the different groups. Moreover, Spearman correlation analyses were used to evaluate the relationship between BTEX and urinary biomarkers. Data from multi-pollutants measurements between interior and exterior of dwellings are reported as mean and standard deviation and U non-parametric test Mann-Whitney was used to determine the statistical differences.

2. Results and discussion

2.1. BTEX concentrations

BTEX analysis by gas chromatography coupled to flame ionization detection revealed that toluene was the most encountered pollutant with concentrations ranging from 800 to 1950 $\mu\text{g}/\text{m}^3$. Benzene and xylenes were ranging from 150 to 650 $\mu\text{g}/\text{m}^3$ while ethylbenzene concentration was about 50 $\mu\text{g}/\text{m}^3$ (**Fig 1** and **Table S2** in supplementary information).

These results are in agreement with previous works where toluene was reported to have the highest concentration of BTEX in urban areas: Miri et al. (2016) ($16 \mu\text{g}/\text{m}^3$) in Tehran (Iran), Pekey and Yilmaz (2011) ($35 \mu\text{g}/\text{m}^3$) in Turkey, Hazrati et al. (2016) ($45 \mu\text{g}/\text{m}^3$) in Ardabil (Iran) and Borgie et al. (2014) ($11520 \mu\text{g}/\text{m}^3$) in Lebanon. **Fig. 1B** highlights the differences of toluene concentrations between outdoor professional groups (1922 and $1023 \mu\text{g}/\text{m}^3$ for traders and bus drivers respectively) and TD housemaids ($867 \mu\text{g}/\text{m}^3$). The outdoor professional activities might determine a prolonged proximity to main streets and/or congested vehicular traffic and therefore a substantial increased occupational exposures to traffic-related air pollution (Knibbs & Morawska, 2012) during the working hours of the selected participants, as also suggested by the toluene concentration exposure here reported. However, the average concentration of BTEX found in HLM housemaids group ($1454 \mu\text{g}/\text{m}^3$) was significantly higher than those of drivers. This result may suggest a major exposure for those groups of participants spending their working hours at a specific urban site (regardless of whether the activity is indoor or outdoor) rather than the one moving around the city.

Benzene concentrations (**Fig. 1A**) were significantly higher for housemaids (649 and $568 \mu\text{g}/\text{m}^3$ respectively for urban and rural sites) than for professionals (192 and $425 \mu\text{g}/\text{m}^3$ respectively for bus drivers and traders). This difference is probably due to local culinary habits (coal cooking), specific indoor practices (important incense burning) and use of products that are important emitters of benzene (Olsson & Petersson, 2003), but is also likely the indoor accumulation and concentration of outdoor emissions (Weisel, 2010). Benzene concentration in our study was higher than the level reported in Cotonou, Benin ($60 \mu\text{g}/\text{m}^3$) (Ayi Fanou et al., 2006), in Lagos, Nigeria ($250 \mu\text{g}/\text{m}^3$) (Baumbach et al., 1995), in Torino, Italy ($10.3 \mu\text{g}/\text{m}^3$) (Bono et al., 2003), in Grenoble, France ($23.5 \mu\text{g}/\text{m}^3$) (Maitre et al., 2002) and in Copenhagen, Denmark ($2.5 \mu\text{g}/\text{m}^3$) (Sorensen et al., 2003).

Ethylbenzene was the least representative VOC with concentration of about $50 \mu\text{g}/\text{m}^3$ whatever the population considered (53, 59, 63 and $45 \mu\text{g}/\text{m}^3$ for rural and urban housemaids, traders and bus drivers, respectively) (**Fig. 1C**).

Finally, among the xylene isomers (**Table S2** and **Fig. 1 D-E**), o-xylene was the most abundant in all the analyzed samples and higher for the urban groups (traders and housemaids) than for the other groups. In Borgie et al. (2014) traffic policemen were exposed mainly to m- and o-xylene, similarly Kim et al. (2012) reported higher urban air concentration of m+p xylene than o-xylene. This difference may be related to the different sources of emissions or to different secondary reaction in the atmosphere of the xylene isomers (Zhang et al., 2019).

Although below the recommended values by the Occupational exposure limit in France (3250, 76800, 88000 and 221000 $\mu\text{g}/\text{m}^3$ for benzene, toluene, ethylbenzene and xylene, respectively), BTEX concentrations here reported go beyond ATSDR's chronic MRL (Minimal Risk Level according to the Agency for Toxic Substances and Disease Registry): 9.8 $\mu\text{g}/\text{m}^3$ for benzene, 300 $\mu\text{g}/\text{m}^3$ for toluene and 200 $\mu\text{g}/\text{m}^3$ for xylene (INRS, 2016).

Just a minor correlation (between o-toluene and xylene) was found among the parameters analysed for the drivers and housemaids in rural site, whereas excellent spearman correlation ($p < 0.001$) between the different BTEX for the traders and housemaids of the urban site is reported (**Table S3**). These results are in agreement with previous studies that reported strong correlations between BTEX compounds (Dehdari et al., 2014; Hoque et al., 2008; Miller et al., 2012) at urban sites of sampling.

This means that the BTEX of the first two groups (traders and urban housemaids) are likely to come from common urban emission sources, and therefore it is likely that indoor levels are influenced also by outdoor emission sources, while those of the rural housemaids probably have different sources of emission. Drivers' case differs from the others since this group is representative of persons that are always moving around the city and their exposure to BTEX can hugely be influenced by other local/transient sources.

In a general, the total BTEX concentration found in this study was considerably higher than that obtained by other studies conducted in developed countries, such as Germany (46.5 $\mu\text{g}/\text{m}^3$) (Schneider et al., 2001) and USA (6.7 $\mu\text{g}/\text{m}^3$) (Smith et al., 2007). The differences observed can be explained by specific characteristics of traffic such as the quality of fuels and/or the number and the mean technology of circulating vehicles while domestic activities significantly impact the levels of indoor pollution.

2.2. Biomarkers of exposure

Benzene can be transformed into primary and secondary metabolites that are usually excreted in urine and can serve as biomarkers of professional or environmental exposure. In this study, urinary S-PMA and t,t-MA levels were considered as biomarkers of environmental exposure to benzene for the enrolled subjects at the urban site (drivers, traders and HLM housemaids) and the ones enrolled at the semirural district (TD housemaids).

Due to low sensitivity of the technique, the urinary concentration of t,t-MA was not detected (values below the LOD) in all the collected samples. S-PMA concentration was found, for the sites we selected, to be a better biomarker of benzene exposure than t,t-MA. Results showed that the median S-PMA values were higher for housemaids (1.3 and 1.2 $\mu\text{g}/\text{g}$ creatinine for

urban and rural group, respectively) than for professionals (1.1 and 0.8 $\mu\text{g/g}$ creatinine for traders and drivers, respectively). However, post hoc analysis indicated that only city housemaids had a significant higher urinary excretion of S-PMA compared to the traders (**Fig. 2A**). S-PMA contents were lower than those found in a previous study conducted in Benin, where a mean S-PMA concentration of 9.3 $\mu\text{g/g}$ creatinine was reported for motorcycle taxi drivers compared to 4.2 $\mu\text{g/g}$ for residents living in a village without traffic emissions (Ayi Fanou et al., 2006). Similarly, in a previous study in Lebanon (Borgie et al., 2014), the pre and post-shift S-PMA levels for traffic policemen (6.2 and 5.8 $\mu\text{g/g}$ creatinine, respectively) and for office policemen (2.6 and 2.7 $\mu\text{g/g}$ creatinine) were higher than those reported in this study despite here we reported higher individual levels of benzene exposure. This difference could be related to a co-exposure of benzene with other chemicals and additives emitted from gasoline vehicles (Barbieri et al., 2008) such as toluene (a competitive inhibitor of benzene for CYP metabolism) which could affect the pharmacokinetics and metabolism of benzene, including conjugation with glutathione and subsequent mercapturic acid excretion (Johnson et al., 2007).

The median level of 1-HOP, used to assess pyrene exposure was slightly higher in drivers (0.4 $\mu\text{g/g}$ creatinine) compared to others groups (0.2, 0.3 and 0.2 $\mu\text{g/g}$ creatinine respectively for traders, urban housemaids and semirural housemaids), but without significant differences. Statistical analysis showed that urinary 1-HOP levels were significantly higher for urban site housemaids compared to semirural district ones (**Fig. 2B**). These values were higher than those observed in diesel exhaust-exposed workers in Denmark (0.11 $\mu\text{g/g}$; Nielsen et al., 1996), in Brazil (0.11 $\mu\text{g/g}$; Brucker et al., 2013) and in Copenhagen (0.25 $\mu\text{g/g}$; Autrup et al., 1999) but lower compared to the level reported for workers of coke oven (5.5 $\mu\text{g/g}$; Ovrebo et al., 1995).

The excretion of 1-OHP in urine can be significantly influenced also by lifestyle (Castano-Vinyals et al., 2004). Food has previously been shown to be an important source of pyrene (Knize et al., 1999) and specific cooking activities may determine a high exposure to organic compounds known to pose significant risks to health (Weinstein et al., 2017).

The high concentrations of BTEX here reported may be related to common local practices, such as the use of wood and charcoal as cooking fuels, incense burning and lack of ventilation of indoor environments. Also the use of cleaning products or solvents may increase the personal exposure to volatile compounds. Lifestyle is therefore an important contributor to PAHs and VOCs exposure and might explain the differences between the pyrene biomarker of exposure between the indoor urban and rural population (**Table S3**). The correlation of 1-

HOP with other VOCs (toluene and xylene concentrations $p < 0.05$) for housemaids at the urban site may indicate that this population is exposed at the same time to indoor and outdoor sources. Interestingly, S-PMA had significant correlation with benzene among housemaids in rural site ($p < 0.05$) and traders ($p < 0.01$) therefore confirming the importance of this metabolite for assessing personal exposure to benzene (**Table S3**).

These results clearly showed that indoor air pollution was a major driver for the personal exposure to BTEX in the selected populations and as consequence this poses a urgent need to regulate or, at least, to activate campaigns of information to share the potential impact of indoor activities in generating volatile organic pollutants.

2.3. Oxidative stress and inflammation biomarkers

Urinary levels of 8-OHdG were higher for urban site housemaids (2.7 $\mu\text{g/g}$ creatinine) when compared to rural site ones (1 $\mu\text{g/g}$ creatinine) (**Fig. 3**). These results support the importance of urban air pollution emissions in determining biological effects and the higher impact of urban pollution compared to rural one. Road traffic is an important source of air pollution in African cities. United Nations estimates that roads account for up to 90% of urban air pollution in developing countries (UNEP, 2011). We have recently reported (Ndong et al, 2019) that the traffic emissions are major contributors to air pollution at the urban site compared to the rural one. These emissions contribute to the risk of chronic respiratory diseases such as asthma and Chronic Obstructive Pulmonary Disease (COPD) among professionals exposed to air pollution related to road traffic. Indeed, recent studies found a high prevalence of allergic rhinitis among vendors in Dakar city (Sylla et al., 2017) and showed a higher prevalence of chronic respiratory diseases and a decreased lung function in bus drivers in the HLM neighborhood in Dakar (Sylla et al., 2018). The results here reported are in agreement with these studies showing that urinary level of 8-OHdG is significantly higher in HLM housemaids in comparison to the ones of the rural site Toubab Dialaw. The lack of high positive correlation (**Table S3**) between 8-OHdG and the VOCs we analysed suggests also that the increase in 8-OHdG may be related to other organic and/or inorganic compounds not analysed in this study and suggest the possibility that other air pollution parameters (such as fine particulate matter) should be taken into account for understanding the observed levels of this biomarker.

The 8-OHdG values found in this study were lower compared to the level in women exposed to wood smoke in a cook stove emissions reported in Commodore et al. (2013) which analysed this biomarker in San Marcos, Peru (132.6 $\mu\text{g/g}$ creatinine; 2013), and lower than the

values reported for pregnant rural Guatemalan women cooking and heating with solid fuels (91.3 $\mu\text{g/g}$ creatinine; Weinstein et al., 2017) and in a cross-sectional sample of 48 green space workers in Brussels, Belgium (between 7.05 and 20.92 $\mu\text{g/g}$ creatinine; Guilbert et al., 2019). The variability in results may be due to the variety of confounding factors associated with urinary oxidant concentrations, such as age, physical activity and vitamin status (Pilger and Rudiger, 2006; Romanazzi et al., 2013) and above all to the exposure to oxidative damaging compounds inhaled or intake by diet or other exposure routes.

As shown in **Fig. 4**, a release of pro-inflammatory biomarkers, such as IL-6 and IL-8 was higher for housemaids at urban and rural sites than for professionals but no significant differences were found between the different groups. IL-1 β and TNF- α were low and did not vary between subjects. Mean IL-6 values here reported (6.7, 6.1, 8.3 and 6.4 pg/mL for drivers, traders, city and semirural housemaids, respectively) were similar to those observed in healthy control subjects population (8 pg/mL) and clearly lower than subjects with congestive heart failure (24 pg/mL) assessed in Spain while TNF- α concentrations were slightly higher (2 and 4 pg/mL respectively for control and sick subjects) (Sirera et al., 2003). This might suggest that specific indoor contaminants could determine the increase of the inflammatory response.

2.4. Air quality measurements

The results of air quality measurements showed that CO and CO₂ concentrations were significantly higher for the drivers population (1.7 ppm for CO and 633.2 ppm for CO₂) than in others environments (**Fig. 5A** and **5B**). Similar CO concentration rates were found in Hong Kong buses (1.9 ppm) (Chan et al., 2002), therefore suggesting a specific occupational exposure. However, the values reported by Limasset et al. (1993) were 11.3 ppm and 12.5 ppm in Paris and Bordeaux buses, respectively. In a comparative study between various travelling habits (walking, motorcycles, light vehicles and buses), Saksena et al. (2008) reported a mean concentration of 11.5 ppm for CO in buses. the American Conference of Governmental Industrial Hygienists (ACGIH) has set a 25-ppm TWA (Time-Weighted Average) threshold in the workplace, and the National Institute for Occupational Safety and Health (NIOSH) has proposed a maximum average concentration (TWA-8 hours) of 35 ppm with a maximum acceptable limit of 200 ppm for 8 hours of work. Considering these limits, the CO concentrations here reported comply with international standards on indoor air quality and occupational exposures and should not pose any risk for the exposed population.

NO, released into the atmosphere in presence of ozone from the air and under the influence of solar radiation, becomes NO₂. **Figure 5C** revealed that NO levels were significantly higher in

the buses (0.1 ppm) than those found at the urban site houses (0.0 ppm). No significant difference was found between NO₂ concentrations at the different environments (**Fig. 5D**). NO concentrations in our study are lower than the accepted average exposure value of 25 ppm (INRS, 2006). Our results agree with those reported in Limasset et al. (1993) which found NO concentrations ranging from 0.5 to 0.6 ppm over a 90 to 130 minute paths in Paris and concentrations from 0.07 to 0.1 ppm for NO₂ for the same travel times.

PM₁₀ (Particulate Matter) are particulates with aerodynamic diameter less than 10 µm. Air pollution guidelines, proposed by the WHO and the European Union, define a threshold of 50 µg/m³ of PM₁₀ (calculated as daily average) to protect the health of exposed population (WHO, 2005). On the other hand the Senegalese air quality law has set a higher limit value (260 µg/m³ for 24 hours). The results of this study showed levels of PM₁₀ concentration lower at the rural site houses (30 µg/m³) than those found at urban sites, houses (200 µg/m³), market (100 µg/m³) and buses (300 µg/m³) (**Fig. 6A**). Concentration levels of PM₁₀ in the buses exceeded the national recommended limit values and are significantly higher than the international standards. Similar results were obtained by Saksena et al. (2008) who showed PM₁₀ concentrations as high as 262 µg/m³ in buses in Vietnam. Similarly Praml et Schierl (2000) found an average concentration of 155 µg/m³ considering a multi buses trip analysis in Munich.

Median temperature values were slightly lower for the urban housemaid population (25 °C) than all the other populations studies (29.2, 28 and 28 °C for drivers, commerciants and rural housemaids, respectively) (**Fig. 6B**) possibly in relation to air conditioning or other type of temperature controlling systems.

This study clearly shows that the interior of the buses circulating in the highly busy urban area of Dakar determines a higher exposure to gases and particles compared to the other environments here investigated. These differences could be explained by the congestion of the roads due to the density of traffic, traffic jams and high emission from old combustion technologies of the circulating vehicles fleet, as well as local topographical properties (such as street canyons) that prevent aeration and dispersion of pollutants and promoting their accumulation (Saksena et al., 2008; Morin et al., 2009; Zagury et al., 2000).

The comparison of indoor and outdoor air quality showed that average PM₁₀ concentrations were significantly higher indoors (0.5 mg/m³) than outdoors (0.2 mg/m³) at the urban site. Similarly, CO levels were significantly higher indoors (3.9 ppm) than outdoors (0.5 ppm). For CO₂, the concentrations found were significantly higher within households in the rural site (**Fig. S2**). No significant differences were observed for the meteorological parameters

(temperature and humidity) between indoor and outdoor sampling areas, both for the urban and the rural site.

These results were similar with other studies where many pollutants were found at higher concentrations in indoor sampling sites in comparison to outdoor ones (Bardana et al., 2001; Kirchner et al., 2011). The accumulation of pollutants in indoor environments may be related to several conditions such as the ventilation of the houses; the season of sampling; the level of outdoor air pollution; the proximity to high traffic roads and the presence of specific indoor emission sources such as gas or wood equipment for cooking (Gerber et al., 1997).

Conclusion

For the first time here we report data on the personal exposure levels of different selected populations to indoor and outdoor air pollution in the sub-Saharan region of Dakar. The data provide an important milestone for the knowledge of the exposure levels to VOCs, PAHs and other air pollutants in developing countries.

Housemaids at urban and rural sites were more exposed than professionals to volatile organic air pollution. Benzene indoor concentrations are probably due to local indoor practices (charcoal cooking, incense burning etc...) and this exposure correlates with the median values of S-PMA (**Table S3**), which were higher among housemaids compared to the other groups.

In addition, urinary levels of oxidative stress (8-OHdG) and inflammatory (interleukin-6 and -8) biomarkers were higher in urban subjects comparing to rural ones therefore suggesting a general major impact of urban air pollution compared to the rural one.

The air quality measurement campaign showed that the bus interior was more polluted with PM₁₀, CO, CO₂ and NO than the market and urban and rural indoor sites. The comparison of air quality between indoor and outdoor sites revealed that, especially at the urban site, indoor air quality may be worse than the corresponding outdoor one, confirming previous observations of potential higher individual exposure level to pollutants in indoor environment.

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465

466 **Declaration of interest**

467 The authors declare no conflict of interest.

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Table S1

Gradient elution of ammonium formate (A1) and acetonitrile (B1).

Table S2

Mean concentration (and number of measurements), minimum, maximum, median first (Q1) and third (Q3) quartile of exposed population (Housemaids-HLM, Housemaids-TD, Traders and Bus drivers).

Table S3

Spearman correlation coefficients between different variables in bus drivers, traders, in housemaids in urban and rural site (*= $p < 0.05$; **= $p < 0.01$; ***= $p < 0.001$).

Fig. 1. Distribution of individual Benzene (A), Toluene (B), Ethylbenzene (C), m+p Xylenes (D) and o-Xylene (E) exposure in between bus drivers, traders and housemaids in urban and rural sites. Values are expressed as median, upper and lower quartiles, range and the outliers (^o) (ANOVA–Tukey test: **a** = $p < 0.05$; **b** = $p < 0.01$, **c** = $p < 0.001$).

Fig. 2. Exposure biomarkers (S-PMA(A) and 1-HOP (B)) in urine between bus drivers, traders and housemaids in urban and rural sites illustrated by the median, the upper and lower quartiles, the range and the outliers (*, ^o) (ANOVA–Tukey test: **a** = $p < 0.05$; **b** = $p < 0.01$).

Fig. 3. 8-Hydroxy-2- α -deoxyguanosine (8-OHdG) in urine between bus drivers, traders and housemaids in urban and rural sites. These values are depicted as median, upper and lower quartiles, range and the outliers (*, ^o) (ANOVA–Tukey test: **a** = $p < 0.05$).

Fig. 4. Protein secretion of Tumor Necrosis Factor-alpha (TNF- α) (A), Interleukin 1-bêta (IL-1 β) (B), Interleukin 6 (IL-6) (C) and Interleukin 8 (D) in urine between bus drivers, traders and housemaids in urban and rural sites. These values are depicted as median, upper and lower quartiles, range and the outliers (*, ^o) (ANOVA–Tukey test).

Fig. 5. Distribution of CO₂ (A), CO (B), NO (C) and NO₂ (D) in between bus market, houses in urban (HLM) and rural (TD) sites. Values are expressed as median, upper and lower quartiles, range and outliers (*, ^o) (ANOVA–Tukey test: **a** = $p < 0.05$; **b** = $p < 0.01$, **c** = $p < 0.001$).

Fig. 6. Distribution of PM₁₀ (A), Temperature (B), and Humidity (C) in between bus market, houses in urban (HLM) and rural (TD) sites. Values are expressed as median, upper and lower quartiles, range and outliers (*, ^o) (ANOVA–Tukey test: **a** = $p < 0.05$; **b** = $p < 0.01$, **c** = $p < 0.001$).

Fig. S1. Location map of the study

Fig. S2. PM₁₀ (A), CO₂ (B), CO (C), Temperature (D), and Humidity (E) levels between outdoor and indoor air in urban (HLM) and rural (TD) sites. Values are reported as mean and standard deviations. * = $p < 0.05$; ** = $p < 0.01$; *** = $p < 0.001$ (Mann–Whitney U; outdoor versus indoor). p indicates differences between HLM outdoor and TD outdoor and a = $p < 0.05$; c = $p < 0.001$. t stays for differences between HLM indoor and TD indoor and b = $p < 0.01$; c = $p < 0.001$.

Figure 1

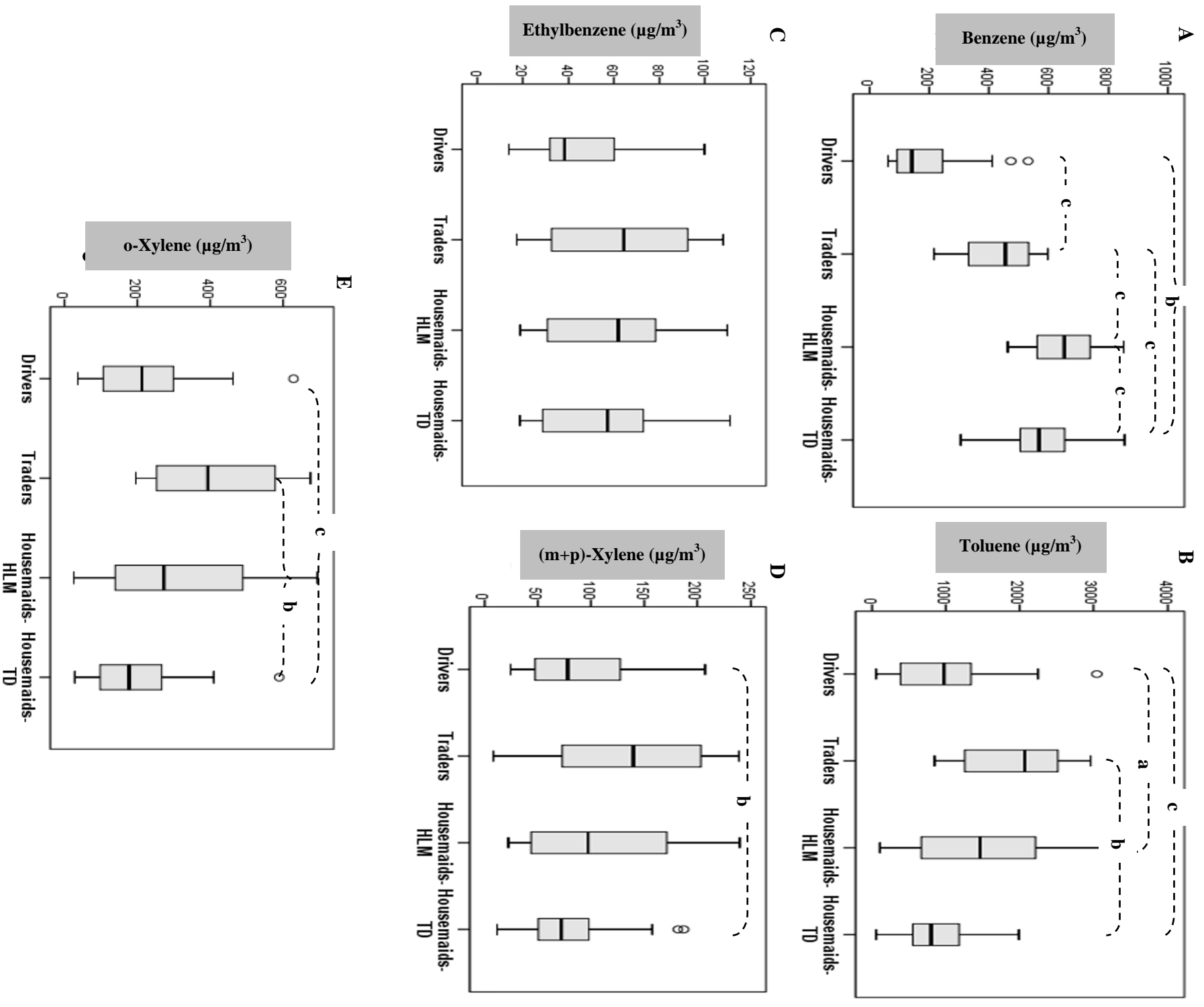


Figure 2

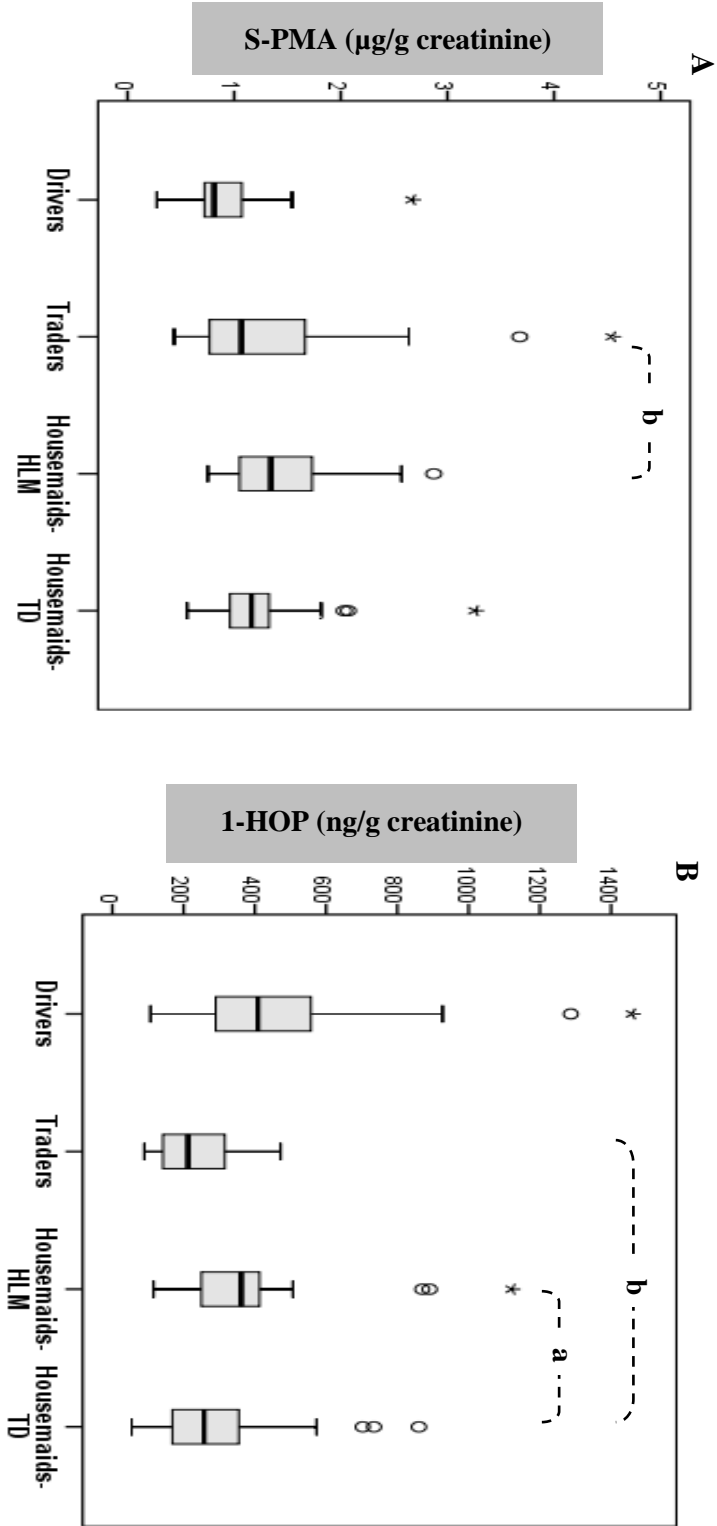


Figure 3

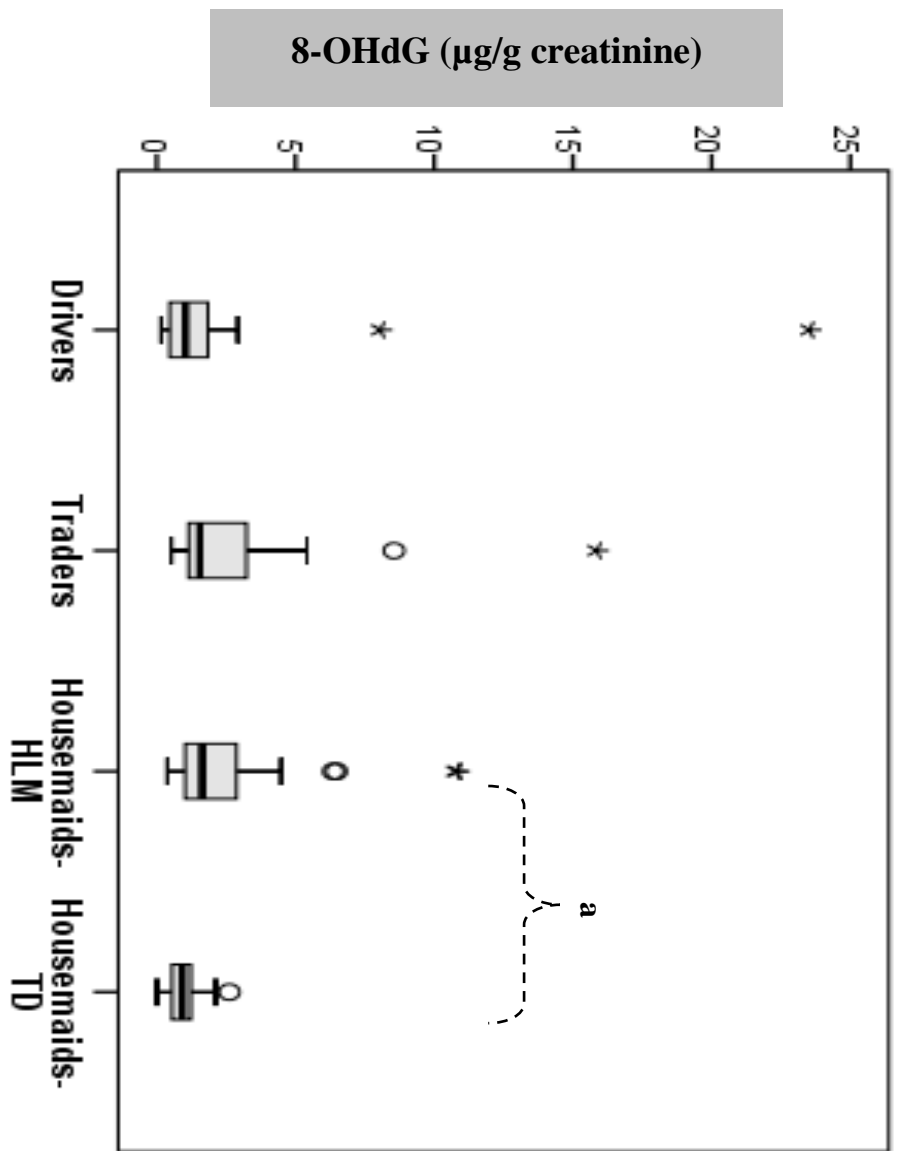


Figure 4

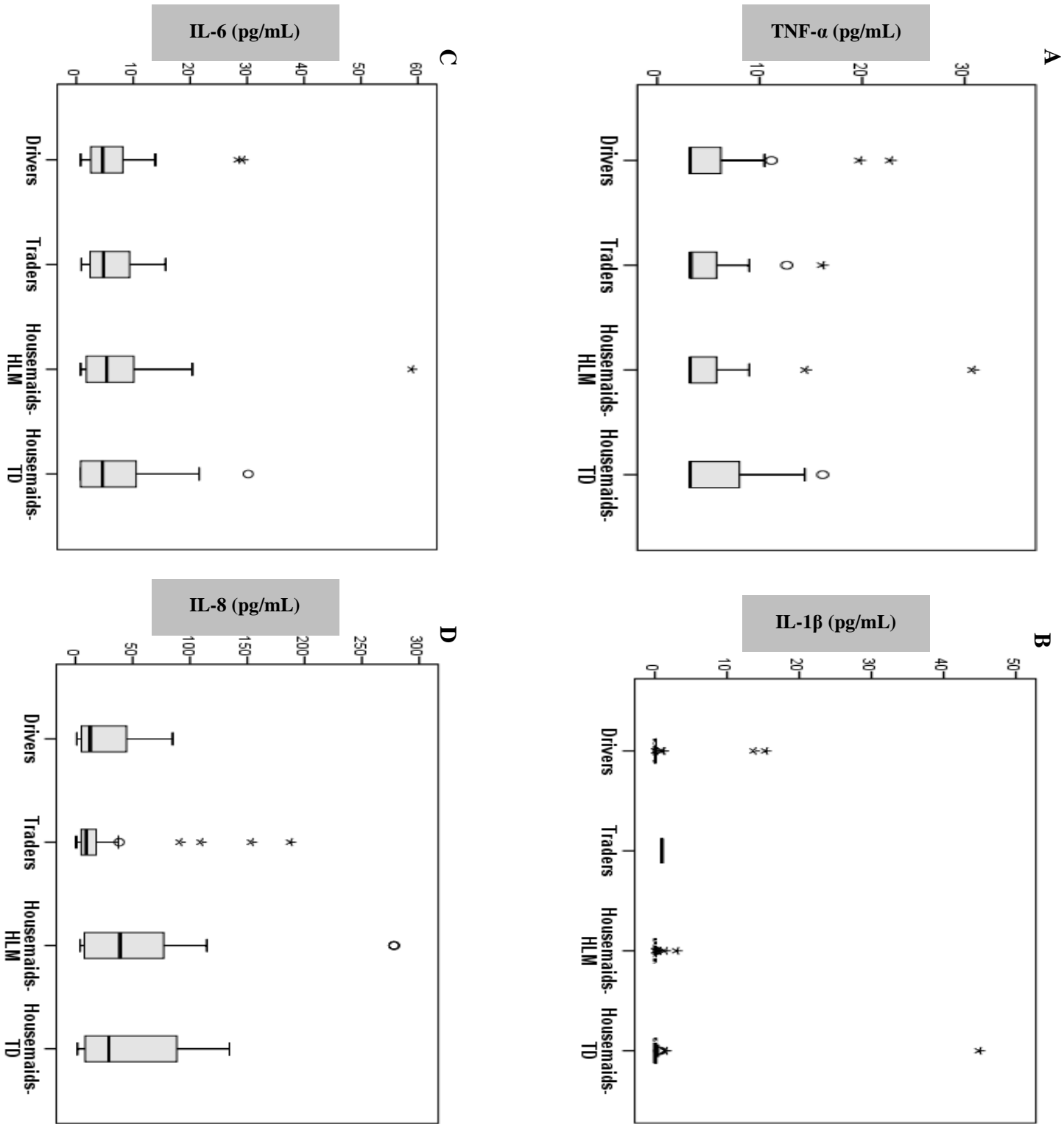


Figure 5

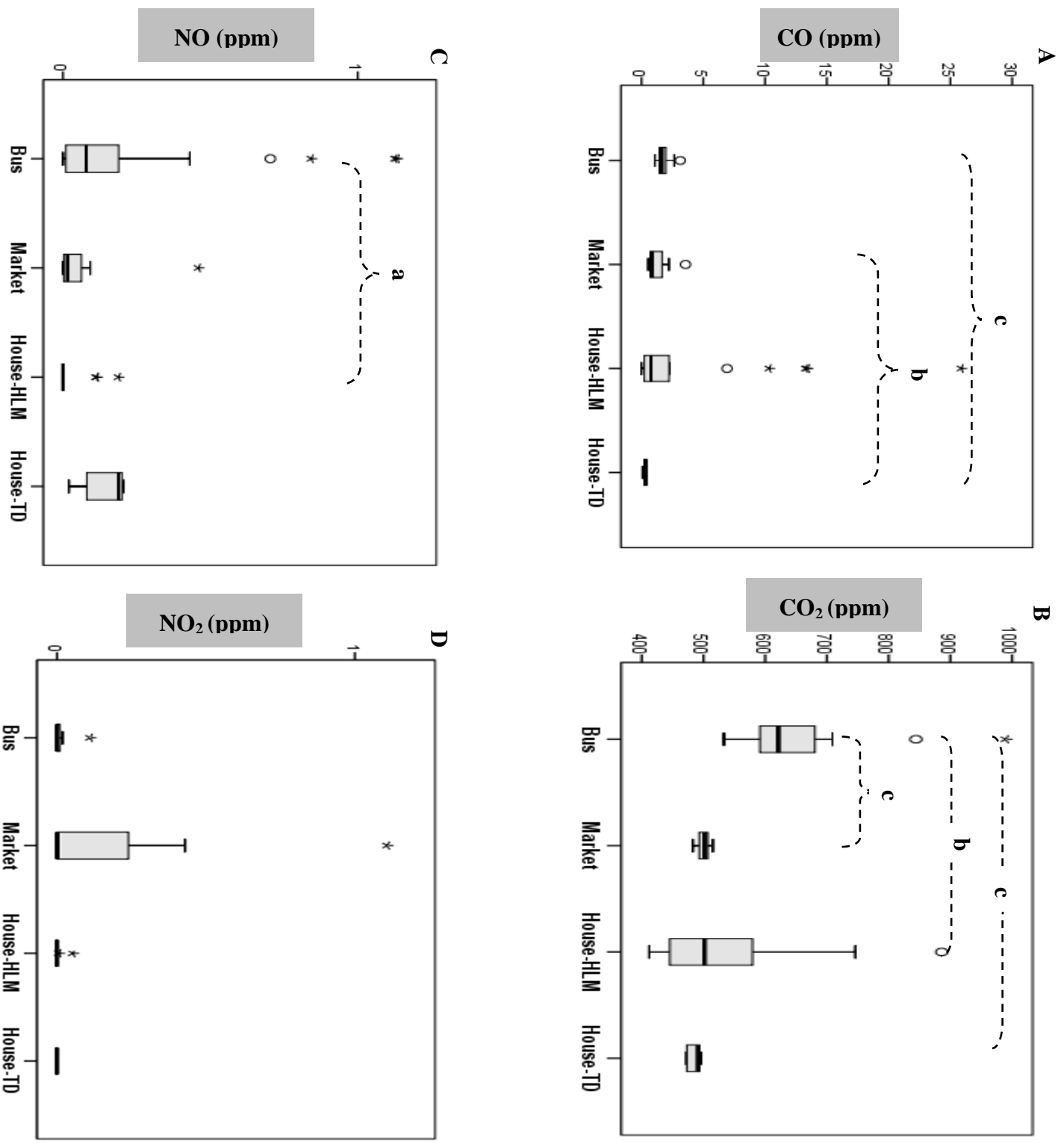
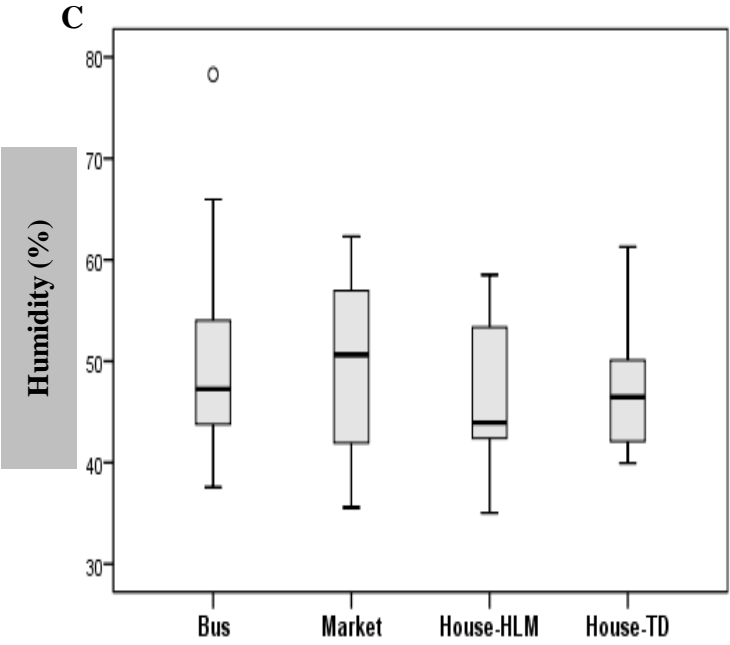
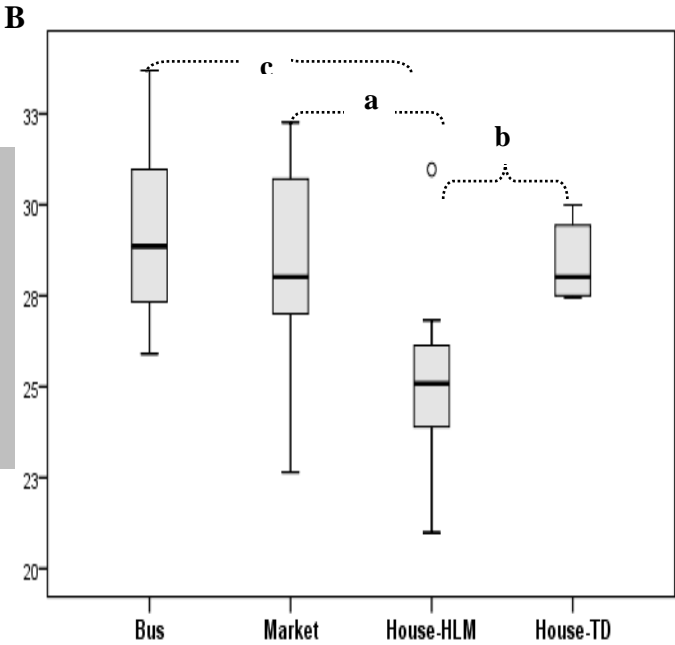
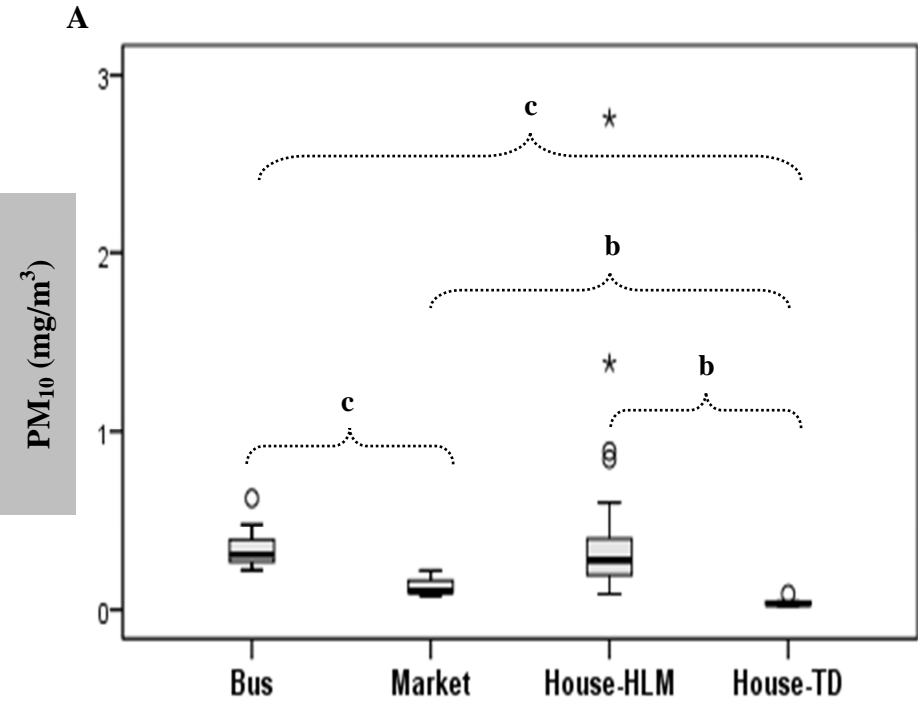


Figure 6



- Indoor organic volatile pollution higher than outdoor one
- Highest urinary levels of oxidative stress and inflammatory biomarkers in urban subjects
- Importance of indoor air pollution on the benzene metabolite S-PMA
- Relevance of indoor habit to determine the exposure levels to selected VOCs
- Correlation of biomarker of exposure with selected VOCs



Urban site



Rural site

Air quality analysis



EVM-7

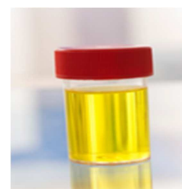


AQ Pro

Human exposure



GABIE badges: BTEX analysis



Urine sampling: biomarkers determination