Exposure to Air Pollutants During Physical Activity

Radiation Science and Technology Department

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EXPOSURE TO AIR POLLUTANTS DURING PHYSICAL ACTIVITY

Exposure to Air Pollutants During Physical Activity

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To my family

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1 Introduction

1.1 Motivation

A clean environment is essential to human health and well-being, however the interactions between environment and health are complex and difficult to evaluate. Environmental factors have an increasing burden on human health. Globally, the environmental pollution, radioactivity and lack of sanitary conditions are the most notorious environmental issues to health concerns. In Europe, a main issue with greater concerns on the effect on human health is indoor and outdoor air pollution. Depending on the person and the situation, a human being can stand up to four weeks without eating and five days without drinking, but can't stand more than a few minutes without breathing, proving that air is a precious asset for life.

In contemporary societies sedentary jobs associated with poor and incorrect diets lead to health problems that can be prevented with physical activity and exercise, thereby becoming another important factor to quality of life. The regular practice of physical activity, such as walking, cycling or practice any kind of sport presents benefits to health and disease prevention. Although, when people are exercising they become more susceptible to the exposure of air pollutants primarily due to increased inhalation rates and volumes.

Due to previous evidence and knowing that research points out that people spent 80-90% of their time indoors, therefore, investigation in the scientific field of air pollution and physical activity is imperative. Growing scientific evidence in recent years indicates that polluted air inside buildings can be a more serious problem than outdoor air, a reality that emphasizes the relevance of indoor air quality (IAQ) in buildings.

Most parts of the studies on IAQ are focused on places were people spent most of their times: homes, work/schools. Comparatively with other facilities and types of buildings, fitness centers or gymnasiums have been the subject of limited study. However, adding to the fact that sport practitioners have an increased susceptibility to be exposed to air pollutants, the time spent by a person in a sport's facility has an important contribution to the integrated daily exposure and inhaled dose of air pollutants. In these types of buildings, specific conditions are joined to affect IAQ, such as building maintenance, building materials and type of ventilation, human occupancy and the activity practiced inside. Accumulation of pollutants due to poor ventilation, growing mold and dampness and dust resuspension are some of the IAQ problems identified in fitness centers.

Exercise in outdoor environments also pose some constraints. Besides the fact that outdoors, people are exposed to meteorological conditions, in urban environments it is not easy to find places to run or cycle avoiding traffic pollution. Furthermore, the incentive from European policies to promote active transportation allied to the increasing cost of fuels, brought the use of bicycles as a more popular mode of transportation in urban streets of the European cities and this behavior is becoming increasingly common. Within the question "*Do the benefits from practicing sports outweigh the risks?*" it is important to characterize peoples' exposure and dose while practice physical activity.

1.2 Exposure and Dose

This thesis is based on the assessment of human exposure and inhaled dose of air pollutants during physical activity. The exposure and the dose are both part of the environmental health paradigm of the health risk assessment (Sexton et al., 2006).

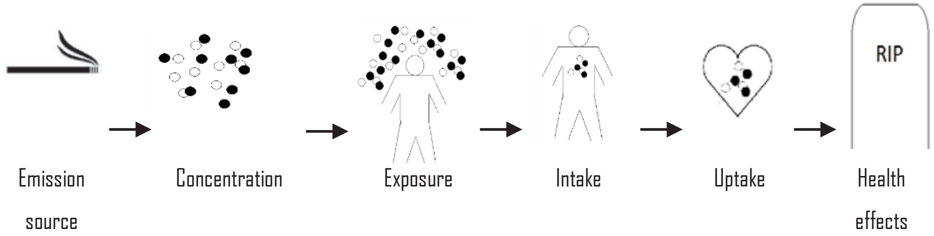


Figure 1.1 – Environmental health paradigm. Adapted from Nazaroff (2008).

Figure 1.1 shows that the chain of events starts with an emission source, which will contribute to the concentration of a mixture of pollutants in the air which people are exposed to. After that, the intake (hereafter referred as dose) can occur by inhalation, ingestion or dermal contact and the body can incorporate part of those pollutants in a process called uptake which can finally cause health effects. "Exposure" is as an event that occurs when a person comes in contact with the pollutant but it is not necessary that the person inhales or ingests the pollutant; "dose" is the amount of material absorbed or deposited in the body for an interval of time and is measured in units of mass (or mass per volume of body fluid in a biomarker measurement) (Monn, 2001).

In most cases, the evaluation of all the steps that are part of the environmental health paradigm is not an easy task. Time constraints, reduced budget, need of material and lack of human resources mean that the focus is applied only in one or two phases of the paradigm. In this work, assessments of pollutant exposure and dose were used to evaluate the health risk and complimentarily gave an important contribution to the evaluation of risk. With the exposure we can predict which pollutant has more burden and with the dose is possible to have a representative quantity of pollutant that interact with our body.

1.3 Outdoor and Indoor Air Quality

Historical facts led to the importance that air quality presents nowadays. In the earliest times, indoor and outdoor air quality was a topic of concern in Egypt (mummified lung tissues revealed that ancient societies suffered from anthracosis), Athens (the greek geographer Strabo described how toxic metallic emissions from smelter furnaces were discharged into the air) and Rome (the philosopher Seneca wrote a letter to a friend exposing the decision of leaving Rome due to the «*oppressive atmosphere of the city with clouds of ashes and poisonous fumes*») (Mosley 2010). In the last century, the concern in air quality focused on ambient outdoor air. The increasing number of industries and the economic growth since the end of the Second World War led to an increase of air pollutants; the London smog in 1952 trigger the first Clean Air Act in Europe, and therefore the political concern and regulation on atmospheric pollution (European Commission, 2014). IAQ complaints and Sick Building Syndrome (SBS) occurrences have steadily increased since 1973 when the Arab oil embargo forced building owners and operators to reduce energy costs in their buildings, in part by reducing indoor/outdoor air change rates (Hill 1991). Episodes of SBS still presently occurring (Apte et al., 2000; Seppanen and Fisk, 2004; Zhang et al., 2014), especially due to insufficient ventilation (Dimitroulopoulou, 2012; Fisk et al., 2009). In the seventies, the first major conference on IAQ was held in Copenhagen in 1978 with the purpose of studying the effects of the indoor environment on the human performance, comfort and health. Past more than thirty years since the Copenhagen conference, some of the discussed topics continue to be part of the topics of current research (Sundell et al., 2011).

In the 80's, global warming, climatic change and ozone depletion (Solomon 1999) were clear signs that governments should take a precautionary attitude towards air emissions by cars and industries.

After years of studies and research on outdoor air, in 2013 the International Agency for Research on Cancer (IARC) and the World Health Organization (WHO) declared outdoor air as a human carcinogenic from group 1 (IARC 2013). Particulate matter (PM) and ozone (O_3) are topic of concern in outdoor and indoor environments (Almeida et al., 2014a). PM vary in size and composition, depending upon its source and formation. PM10 were the most studied particles in the last decades but more focus is being placed in the fine fraction (PM2.5-PM1) and ultrafine particles (<PM1, UFP). Outdoors, particles come from anthropogenic sources (industries, traffic, dust resuspension, combustion sources) or natural events (Sahara dust events, volcanoes) (Almeida et al. 2006; Almeida et al. 2013a); the indoor particles are greatly influenced by the outdoor particles, but human activities also have a large impact on particles production and resuspension (Ferro et al. 2004; EPA 2007; Boor et al. 2013). The photochemical reaction of O_3 in densely populated areas of abundant sunshine is elevated due to increased emissions involving vapor phase organic compounds, nitrogen oxides,

carbon monoxide and sunlight (Weschler 2000). Because O_3 is a very strong oxidant, O_3 has the capacity to react with carbon double bonds in polyunsaturated fatty acids of fluids lining the lung, causing significant effects on pulmonary function and airway inflammation in individuals with pre-existing respiratory disease (Levy 2001) and on healthy young adults, as described in a more recent study (Kim et al., 2011).

Besides their importance in outdoor air, volatile organic compounds (VOC), carbon monoxide (CO) and carbon dioxide (CO_2) are more concerning indoors, mostly due to poor ventilation rates and poor conditions for pollutant dispersion. VOC are a mixture of chemical compounds; their main source outdoors being from gasoline vapor emitted by traffic (especially the BTEX mixture: benzene, toluene, ethylbenzene and the isomeric xylenes). Indoors, VOC are released by tobacco smoke, furniture, varnish, cleaning products and solvents (Shin and Jo 2012). These compounds are linked with airway and eye irritation (CCOHS 2004), impaired lung function (Cakmak et al., 2014), with two compounds of the BTEX mixture classified by IARC as carcinogenic to humans (benzene) and possibly carcinogenic to humans (ethylbenzene). CO is particularly dangerous because it is colorless and odorless. Because CO has 200 times more affinity with hemoglobin than oxygen (O_2), when presented at significantly high concentrations can be lethal (Kao and Nañagas 2005). If exposed to CO, a person can experience headache, nausea and fatigue (CCOHS 2004). Indoor sources of CO are related with combustion sources (stoves, heaters) and low ventilation rates or with contaminated air from outdoor air or garages. Indoors, CO_2 is a bioeffluent, released by occupants. To reach CO_2 levels warranting preoccupation, ventilation rates need to be very low, of 1L/person or less (Persily 1996). CO_2 causes dizziness, lack of attention, diminished of cognitive function and decline in productivity in general population (Gaihre et al., 2014; Ferreira and Cardoso, 2014).

Apart from chemical pollutants, indoor environment can also be contaminated by microorganisms. The domain of microorganisms includes mainly fungi, bacteria, their spores, toxins and volatile microbial organic compounds. Indoor concentrations of some of these organisms and agents are known or suspected to be elevated in damp indoor environments and may affect the health of people (WHO 2009a) with respiratory infections, allergies, asthma and impaired chronic respiratory disease (Douwes et al., 2003). Controls of moisture and ventilation rates are correct options to reduce and control the presence of microorganisms indoors (Nazaroff, 2013).

Ventilation is a key factor in the promotion of good indoor air quality (Mendell et al. 2013; Nazaroff, 2013; Canha et al., 2013; Gao et al., 2014; Almeida-Silva et al., 2014a). Ventilation is the process that promotes the entry and renewal of the air in spaces which has two primary purposes: i) to remove or dilute pollutants and odors of indoor air and ii) promote thermal comfort in indoor environments. Ventilation can be mechanical, natural or a mixture of both (hybrid ventilation), but it’s the effectiveness of the ventilation process that determines the efficiency to achieve the purposes of ventilation.

Alves et al. (2014) performed a study in a university gym and in a fronton, concluding that the use of climbing chalk contributes to high loads of particulate material, which is very rich in carbonate and magnesium. Similar results were obtained by Weinbruch et al. (2008). Braniš and Safránek (2011a) characterized the coarse PM in school gyms by X-ray spectrometry and showed the importance of ascertaining the composition of particles. Buonanno et al. (2013) developed a comprehensive study on particles and provided data on ventilation rates, an important issue to obtain a complete study regarding IAQ. Ice rinks are another sport facility that warrants the attention of IAQ control. Some studies and recommendations have been made to establish healthier spaces due to concerns of CO, NO_2 and particle concentrations (Pelham et al. 2002; Salonen et al. 2008) due to the presence of ice resurfacers powered by propane- or gasoline-fuelled engines that produce exhaust emissions. None of these authors assessed the metal concentrations of indoor particles sampled in fitness centers. Regarding microbiological contamination, Viegas et al. (2010, 2011) conducted a study in fitness centers to characterize fungal contamination, although scarce information is known about bacterial concentrations in this setting. The existent studies regarding sport facilities are with reference to scholar gyms, climbing halls and ice rinks. There is a lack of scientific information on regular fitness centers.

By reducing air pollution levels, countries can reduce the burden of diseases such as heart disease, lung cancer, and both chronic and acute respiratory diseases, including asthma (WHO, 2014a). Exposure to pollutants such as airborne particulate matter and ozone has been associated with increases in mortality and hospital admissions due to respiratory and cardiovascular disease (Brunekreef and Holgate, 2002; Almeida et al., 2014a). According to the Organization for Economic Cooperation and Development (OECD) the number of deaths due to outdoor air pollution fell by about 4% between 2005 and 2010. Fourteen of the thirty four OECD countries didn’t achieved progress, and Portugal is one of those countries, which recorded an increase of deaths from 3 623 to 3 842 (OECD 2014). The data states that vehicles accounts for an average of 50% of air pollution in OECD countries, indicating that traffic should be the focus to reduce air pollution.

1.4 Physical Activity

As the world develops, the types of diseases that affect the population shift from primarily infectious to primarily non-communicable diseases (NCD). Cardiovascular diseases account for 17.5 million people annually, followed by cancer (8.2 million), respiratory diseases (4 million), and diabetes (1.5

million); all of which can be prevented through physical activity (Warburton et al., 2006). Physical inactivity is one of the most important health challenges of the 21st century because of its influence on the NCD, contributing worldwide to 21% of ischemic heart disease, 11% of ischemic stroke, 14% of diabetes, 16% of colon cancer and 10% of breast cancer (WHO, 2009b). Thus, the benefits to health and human well-being derived from physical activity are recognized all over the world.

The Global Action Plan for the Prevention and Control of NCDs 2013-2020, which includes Portugal, aims to reduce the number of premature deaths from NCDs by 25% by 2025 (WHO, 2015). The WHO target is to reduce the prevalence of insufficient physical activity by 10% and therefore proposes strategies focusing on urban planning, transport policies and the creation of built and natural environments which support active transport and physical activity (WHO, 2013). Notwithstanding, poor air quality is one of several factors that cause people to make people apprehensive against performing physical activity (WHO, 2014b). The concern of the influence of air pollution during exercise came up with a special focus during the Beijing XXIX Olympic Games. At that time, efforts were made to reduce the pollution in the city and studies on athletes' performance during and after the event were conducted (Lippi et al. 2008; Salthammer 2008; Braniš and Vetvicka 2010; Wang T. et al. 2010; Wang SL et al. 2014). In addition to this emblematic event, previous studies on air quality have already been made in sport facilities (Lee et al., 1994; Yang et al., 2000; Pelham et al., 2002).

The exposure to air pollutants during exercise can cause decreases in lung and vascular function, increased airway and systemic oxidative stress (Weiss and Rundell, 2011; Rundell, 2012). In fact, athletes and the common individual can be at risk when exercising in polluted environments due to the fact that: 1) the increase in the minute ventilation (VĖ, L/min) increases proportionally the quantity of inhaled pollutants; 2) most part of the air is inhaled through the mouth, bypassing the normal nasal mechanisms for filtration of large particles in the upper respiratory system and; 3) the increased airflow carries gaseous pollutants deeper into the lungs (Carlisle and Sharp, 2001). Therefore, it is clear that the sport facilities have to be correctly planned and assessed, aiming to reduce the risk of exposure to air pollutants and enhancing the benefits of exercise. Studies on inhaled dose during physical activity are very scarce and no data was found on this topic during the review of current literature. In this thesis the VĖ of individuals undertaking typical fitness classes has been conducted for the first time.

The active transportation has the support of the WHO which encourages the creation of policies and investments in cleaner transport (WHO, 2014a). In London the levels of cycling doubled after the introduction of a congestion charge, but also with a significant investment in cycling infrastructure (de Nazelle et al., 2011); in Lisbon the need to comply with the limit values of ambient air pollutants lead to the creation of Low Emission Zones (LEZ) in order to reduce the pollution in specific streets

(CML, 2014); in Barcelona, the public bicycle sharing initiatives had more benefits than risks to health and reduced the CO_2 emissions (Rojas-Rueda et al., 2011); in Singapore, to encourage commuters to choose the most appropriate mode of transport, investments have been made in road infrastructure, public transport and traffic management schemes (road user charges and fiscal measures on car ownership); France introduced consumer-directed incentive measures in the form of bonus rebates for buyers of new vehicles with low CO_2 emissions; Seoul enacted a "Weekly No-Driving-Day Programme" that had a 30% participant rate, resulting in a 12% reduction of emissions and a 7% decrease in traffic (UNEP, 2009).

Few studies have taken into account that cyclists have an increased $\dot{VE}$ compared to other commuters influencing their inhaled dose of air pollutants. Daigle (2003) and his team studied for the first time this relation by assessing the deposition fraction of UPF during rest and exercise and concluded that deposition increases with decreasing particle size and increases with exercise. Zuurbier et al. (2009) assessed the differences of $\dot{VE}$ among cyclists, car and bus passengers, that in a following study were used to assess the inhaled dose for PM10, PM2.5, soot and particle number counts (PNC) between bus users, car users and cyclists (Zuurbier et al., 2010). Panis et al. (2010) also quantified the exposure and dose of car passengers and cyclicts to PM. Cole-Hunter et al. (2012) assessed the exposure and dose to UFP in two different cycle routes in Brisbane (Australia); Nyhan et al. (2014) also studied the dose among cyclists, pedestrians bus and train passengers. These four studies reported data on inhaled intake dose, but their sampling was limited to one, two or three periods of the day. Small sampling periods do not reflect the hourly variability of air pollutant concentrations and consequently do not reflect the exposure variability. The revision of literature also revealed that a small number of studies comprise more than three modes of transportation, mostly bicycle/walking, car and bus; the ones who embrace more transportation modes restricted their focus to one or two pollutants, such as PM2.5 (Adams et al., 2001), PM2.5 and CO (Kaur and Nieuwenhuijsen, 2009), PM2.5 and PM10 (Chan et al., 2002; Nyhan et al., 2014), UFP (Kaur et al. 2005) or VOC (McNabola et al., 2008). Recently in Lisbon, Baptista et al. (2015) studied the use of different transportation modes (conventional and electrical bikes, electrical vehicles, conventional vehicles and buses) to test the energy consumption and emission, distance and trip time on a specific route. The main focus of those studies is related to exposure to air pollutants on different cycle routes (lanes or paths) and their characteristics which influence people's exposure (Kendrick et al., 2011; MacNaughton et al. 2014). The benefits of active transportation are becoming more and more discussed to reduce traffic pollution, however this shift has influence on personal dose and it cannot be negligenciable.

1.5 Thesis Outline

This thesis comprises the study of personal exposure and inhaled dose during physical activity in three domains. Literature in this field presents the exposure to pollution during physical activity in a disintegrated form while the work developed to this thesis aims to study the personal exposure and the inhaled doses during exercise in a harmonized approach, addressing exposure indoors, outdoors and the complementary use that active transport add to exercise. Research provides better information which leads to better decision making. The main aim of this thesis is to give people the information that they need in order to avoid air pollutants and make better and healthier choices while practicing physical activity in indoor and outdoor environments. Figure 1.2 describes the framework to comprehend and overview this thesis, including the main findings in each chapter.

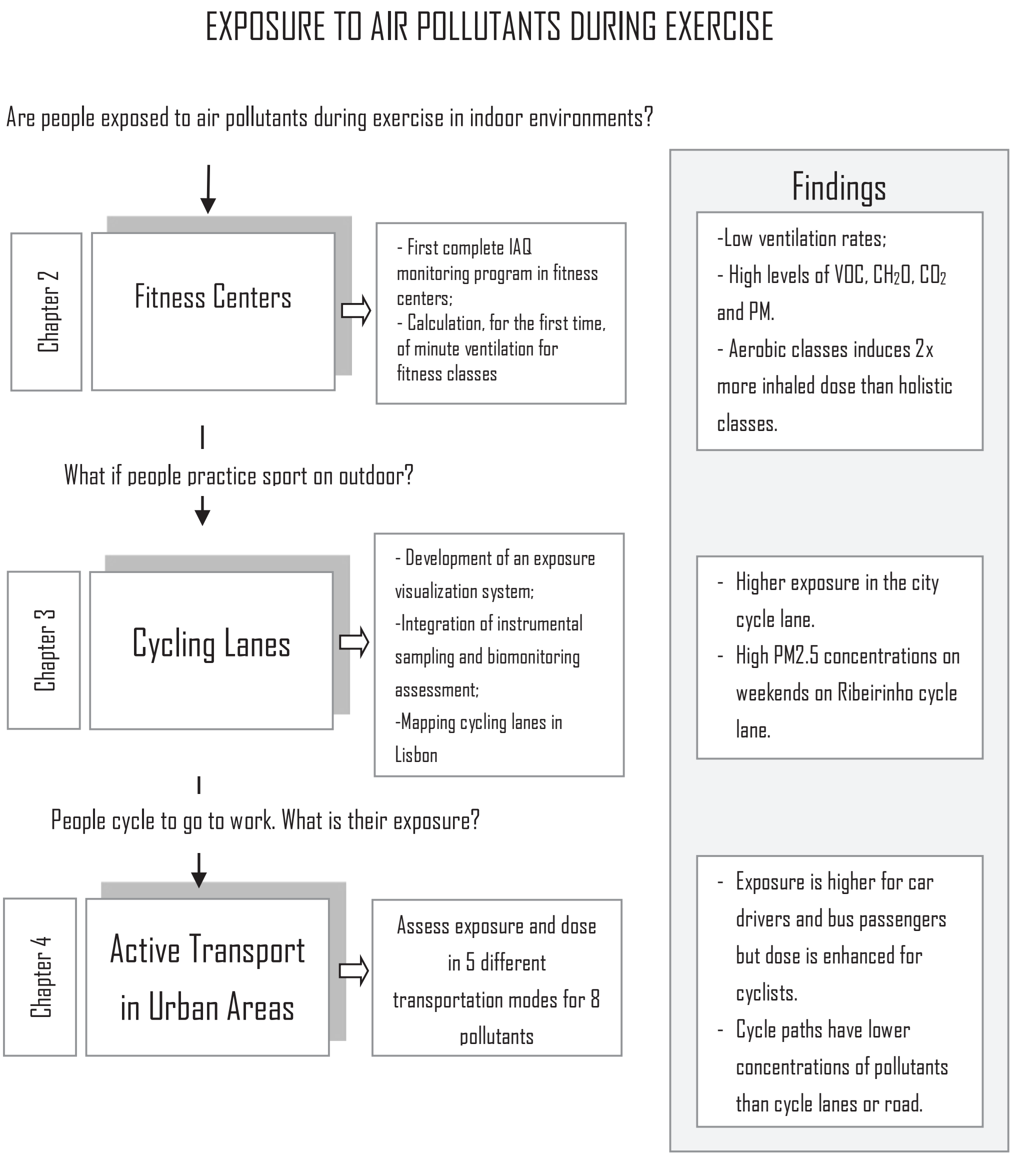
To achieve this goal, this thesis has the following objectives:

- Characterization of the IAQ in fitness centers;
- Identification of the principal sources of indoor air pollution in fitness centers;
- Assessment of the fungal and bacteria contamination in fitness centers;
- Estimation of the dose of inhaled pollutants in fitness centers;
- Assessment of the exposure and dose on three different cycle routes;
- Identification of pollutant sources which contribute to exposure during cycling;
- Comparison of the exposure and dose between bicycle and other transportation modes;
- Map the chemical elements and CO, CO_2, COV, O_3 and PM.

In Chapter 2, results obtained in an IAQ monitoring program developed in eleven fitness centers in Lisbon are presented. Additionally, a deeper IAQ assessment was performed in three of the eleven fitness centers considering daily patterns of the chemical pollutants and their possible sources. The microbiological assessment of fungi and bacteria was also addressed in these three sites, in order to have an IAQ evaluation as a whole. The estimation of the inhaled doses in fitness centers was performed with data collected from the IAQ monitoring program and with estimation in laboratory of the minute ventilation of a group of volunteers.

Chapter 3 focuses on the exposure and the dose to air pollutants during cycling using personal equipment and biomonitoring techniques. With the use of personal equipment it is possible to observe differences of particle concentration between peak and non-peak hours, weekends and weekdays. An exposure visualization system was created: combining GPS position with the measured concentrations enables the visualization of the hot spots in the cycle lanes. The biomonitoring technique was useful to measure elemental concentrations in the cycle lanes and to identify possible pollution sources. Elemental concentrations can give an overview on pollution sources on cycle paths and cycle routes, therefore giving information on where efforts should be implemented to correctly design cycle routes in order to reduce users' exposure to pollutants.

Chapter 4 describes the exposure and dose in different transportation modes along a main route in Lisbon. The selected transport modes were bus, metro, motorcycle, car and bicycle. With this approach the use of public or private transport with the use of active transport can be compared; pollutant maps for the assessed pollutants were created again in this chapter for the bicycle concentrations.



Are people exposed to air pollutants during exercise in indoor environments?

- First complete IAQ monitoring program in fitness centers;
- Calculation, for the first time, of minute ventilation for fitness classes

What if people practice sport on outdoor?

- Development of an exposure visualization system;
-Integration of instrumental sampling and biomonitoring assessment;
-Mapping cycling lanes in Lisbon

People cycle to go to work. What is their exposure?

Assess exposure and dose in 5 different transportation modes for 8 pollutants

-Low ventilation rates;
- High levels of VOC, CH_2O, CO_2 and PM.
- Aerobic classes induces 2x more inhaled dose than holistic classes.

- Higher exposure in the city cycle lane.
- High PM2.5 concentrations on weekends on Ribeirinho cycle lane.

- Exposure is higher for car drivers and bus passengers but dose is enhanced for cyclists.
- Cycle paths have lower concentrations of pollutants than cycle lanes or road.

Figure 1.2 – Thesis framework

2 Sport Practitioners Exposure to Indoor Aerosols

2.1 Exposure to indoor air pollutants during physical activity in fitness centers

Based on the article:
Exposure to indoor air pollutants during physical activity in fitness centers
C.A. Ramos, H. T. Wolterbeek, S. M. Almeida (2014)
Building and Environment, 82: 349-360

2.1.1 Abstract

Physical activity has become a social need among people and it has been clearly proved that exercise is a way to prevent all-cause and cardiovascular-related death, diabetes mellitus and obesity. However, athletes and the common individual can be at risk when they are practicing exercise in polluted environments. In 2012, a monitoring program was undertaken in 11 fitness centers from Lisbon where comfort parameters (temperature and humidity) and indoor air pollutants (PM10, PM2.5, CO_2, CO, CH_2O and VOC) were measured. Three gyms were selected to perform a deeper analysis consisting of longer measurement periods and more parameters, such as particle chemical composition and nanoparticle lung deposition. Measurements were performed during the occupation time, in the studios and in the bodybuilding room, in order to recognize daily patterns and to identify pollutant sources. The pollutants CO_2, VOC and CH_2O presented high concentrations exceeding the national limit values, while O_3 and CO did not present concerning levels. Pollutant continuous measurements demonstrated increased levels of particles when the spaces were occupied during classes. Results indicated that it is crucial to optimize the HVAC systems, ventilation rates and occupants behavior in order to reduce the exposure to air pollutants in fitness centers and to potentiate the benefits of sport activities.

2.1.2 Introduction

According to the WHO, physical inactivity was identified as the fourth greatest risk factor for mortality, accounting for 3.2 million deaths per year in the world (WHO 2012). Physical activity is an important factor for life quality and frequent practice of exercise, like walking or bicycling, presents great benefits for health (Warburton et al., 2006). A clean environment is also essential for human health and well-being. In Europe, the environmental aspect with most concern on human health is related to indoor and outdoor air pollution (EEA 2011; Almeida et al., 2014a). Considering these two aspects and in order to potentiate the benefits of physical activity, people who live in urban areas choose less polluted outdoor spots or fitness centers to avoid air pollution.

Fitness centers present specific characteristics that can affect the IAQ. Like in other indoor places, IAQ in gymnasiums is affected by building maintenance, building materials and type of ventilation, but what makes these places peculiar are the higher human occupancy and the type of activity developing inside. Fitness centers join all the conditions that promote the increase of CO_2 concentrations because occupants are the dominant source of indoor CO_2 and its production rate depends primarily on the number of people in the room and on their metabolic level (Apte et al., 2000). Occupancy also has influence in the PM10 concentrations (Ferro et al., 2004). Braniš et al. (2011) and colleagues observed a direct relation between the indoor concentrations of coarse PM and the number of children present in a scholar gymnasium. Also in school gyms, Buonanno et al. (2013) concluded that the high levels of coarse PM concentrations are produced by students' activity.

Therefore, there is an indubitable higher exposure to air pollutants in gymnasiums not only due to the characteristics of these places but also due to the changes in the respiratory parameters caused by the physical activity. However, despite the importance of healthy air in sport facilities, IAQ studies have been focused principally on schools (Pegas et al., 2010, 2011a; Canha et al., 2010, 2011; Almeida et al., 2011; Canha et al., 2012a,b; Canha et al. 2014a,b), elderly care centers (Almeida-Silva et al. 2014a,b; Viegas et al., 2014) homes (Osman et al., 2007) and offices (Bluyssen et al., 1996). Comparatively, IAQ monitoring programs carried out in sport facilities are very scarce. Aside from the studies of Buonanno et al., (2013) and Braniš et al., (2011a,b) performed in school gyms, only the exposure in ice rings are object of study since the 90's due to the high levels of CO, NO_2 and PM that are emitted by the ice resurface vehicles (Lee et al., 1994; Yang et al., 2000; Pelham et al., 2002) and exposure in swimming-pools due to the high concentrations of trihalomethanes (Dyck et al., 2011). Some other works were conducted in fitness centers but their focus was energy consumption or thermal comfort (Lam and Chan, 2001; Beusker et al., 2012; Revel and Arnesano, 2014).

This work aims to conduct a comprehensive characterization of IAQ in fitness centers and to identify the principal sources that compromise IAQ. This evaluation will be useful for epidemiological studies and to develop appropriate control strategies not only to minimize the adverse health effects on exercise practitioners, but also to potentiate the benefits of the physical activity.

2.1 3 Methodology

2.1.3.1 IAQ Assessment in 11 Fitness Centers

A monitoring program was undertaken in 11 fitness centers from Lisbon where comfort parameters (temperature and humidity) and indoor air pollutants (PM, CO_2, CO, CH_2O, VOC and O_3) were measured. Figure 2.1 shows the localization of the selected fitness centers and Table 2.1 presents their specific characteristics.

Figure 2.1 – Location of the studied fitness centers

Three direct reading apparatus were used: a Lighthouse Handled 3016 to measure PM5-10, PM2.5-5, PM1-2.5, PM0.5-1 and PM0.3-0.5, temperature and relative humidity, a Greywolf (IAQ 610,WolfSense Solutions, USA) to assess CO_2, CO, VOC and O_3 and a Formaldemeter (htV-M, PPM Technology, USA) to quantify the levels of CH_2O. All devices were calibrated according to the fabricators specifications.

Measurements were performed during the period of the late afternoon/night, in order to overlap with the more occupied period and with the worst case scenario. In each fitness center, measurements took place in the bodybuilding room (Bb) for 60 minutes, and in two studios (S1 and S2), during the time of a fitness class (varied between 45 – 60 minutes). Equipment was positioned at an elevation of 1.20m from the ground and 1.50m away from walls in order to avoid the influence on airborne particle dispersion (Jin et al., 2013; Holmberg and Li, 1998). The same air pollutants described previously were measured in the outdoor air. The sampling campaign was performed in October of 2012.

2.1.3.2 IAQ Assessment in 3 Selected Fitness Centers

After the previous analysis, three fitness centers were selected in order to perform a deeper IAQ assessment. In these sport facilities 1) daily continuous measurements of pollutants were performed in different spaces of the gymnasiums in order to recognize daily patterns and identify pollutant sources; 2) particles were sampled and element concentrations were measured; and 3) nanoparticle deposition in lungs were studied. The adopted methodology resulted in three campaigns of six days assessment, in each fitness center, that occurred from October to December 2012.

The selection of the fitness centers (G9, G10 and G11) was made based on the number of daily users, number of fitness classes during the day and type of ventilation. In G9, two studios and the Bb room were selected and monitored during two days in each space. In G10 and G11 only one studio was monitored (since the equipment's noise was incompatible with the classes practiced inside the other studios) resulting in four days of monitoring in the selected studio and two days in the bodybuilding room.

2.1.3.2.1 Continuous measurements of gases

The pollutants CO_2, CO, VOC and O_3 were measured continuously with the equipment GrayWolf. Data was registered in the same conditions as in chapter 2.1.3.1. In outdoor, parallel measurements of CO_2 and CO were performed (7545 IAQ-Calc, TSI, USA). Both devices were calibrated according with the fabricator specifications.

2.1.3.2.2 Particle sampling and measurement

Within the indoor areas of the selected fitness centers, particles were not only measured continuously with a Lighthouse Handled 3016, but were also sampled for subsequent PM10 chemical characterization with the medium volume sampler (MVS6, Leckel, Germany; flow rate 3.5 m^3/h,). Simultaneously, a Partisol 2000 (ThermoScientific, USA; flow rate 1 m^3/h) was used for outdoor PM10 chemical characterization. Teflon filters with 47mm were used to collect particles.

Table 2.1 – Main characteristics of the studied gymnasiums

	Year of construction	Location	Space	Volume (m^3)	Capacity (person)*	Floor type	Wall type	Ventilation system
G1	2009	Urban (residential area)	S1	337	20	Linoleum	Brickwork	Mixed
			S2	448	20			
			Bb	65	40			
G2	1997	Urban (street with intense road traffic)	S1	129	20	Floating Floor	Brickwork	Mechanical
			S2	266	30			
			Bb	2442	50			
G3	2003	Urban (residential area)	S1	394	25	Floating Floor	Brickwork Glass	Mechanical
			S2	394	25			
			Bb	990	40			
G4	1982	Urban (street with intense road traffic)	S1	146	15	Wood	Brickwork	Natural
			S2	136	15			
			Bb	87	10			
G5	2012	Urban (street with intense road traffic)	S1	219	30	Floating Floor	Brickwork	Mechanical
			S2	82	15			
			Bb	641	20			
G6	2012	Urban (residential area)	S1	395	35	Linoleum	Brickwork Glass	Mechanical
			S2	462	35			
			Bb	1509	50			
G7	1923	Urban (residential area)	S1	387	30	Wood/ Linoleum	Brickwork	Mixed
			S2	748	40			
			Bb	866	40			
G8	2012	Urban (street with intense road traffic)	S1	148	20	Linoleum	Brickwork Glass	Mechanical
			S2	306	30			
			Bb	1062	50			
G9	2010	Urban (street with intense road traffic)	S1	447	35	Floating Floor	Brickwork Glass	Mechanical
			S2	788	35			
			Bb	1948	60			
G10	2000	Urban (inside a city park)	S1	1156	40	Floating Floor	Brickwork Glass	Natural
			S2	1156	40			
			Bb	540	40	Linoleum		
G11	2005	Urban (residential area)	S1	745	35	Floating Floor	Brickwork Glass	Mechanical
			S2	745	35			
			Bb	1843	70			

* Maximum capacity. S1 – Studio 1; S2 – Studio 2; Bb – Bodybuilding room.

When the sampling was conducted in the studios, PM10 samplers (MVS6 and Partisol 2000) only worked during the occupied time, whereas in the Bb room, these devices worked continuously from the opening until the closure of the fitness centers.

The direct reading apparatus worked continuously, from the opening until the closure of the gymnasiums, and particle concentrations were registered every 60 seconds.

A correction factor (β) was applied to the PM indoor concentrations obtained by the Lighthouse Handled 3016. This correction factor was obtained by calculating the ratio between the concentrations obtained by the gravimetric method (considered as the reference method) and the concentrations measured by the Lighthouse 3016 (McNamara et al., 2011; Diapouli et al., 2008). The opening and closing of windows and the number of occupants were registered.

2.1.3.3 Elemental Composition of PM10

The filter loads were determined by gravimetry using a 0.1 μg-sensitivity balance in a clean laboratory (class 10 000) at a temperature of 20 ± 1ºC and a relative humidity of 50 ± 5% (EN12341:1998). Before being weighted, filters were equilibrated for 24 hours in the same room. Filters were weighted before and after sampling and the mass was obtained as the average of three measurements, when observed variations were less than 1%.

The chemical characterization of indoor and outdoor PM10 samples was performed by Instrumental Neutron Activation Analysis using the k_0 methodology (k_0-INAA) (Freitas et al., 2003, 2004; Almeida et al., 2013a).

For k_0-INAA, half of a filter was rolled up and put into a clean thin foil of aluminum and irradiated for 5h at a thermal neutron flux of 1.03 x 10^{13} cm^2/s in the Portuguese Research Reactor. After irradiation, the sample was removed from the aluminum foil and transferred to a polyethylene container. For each irradiated sample, two gamma spectra were measured during 7h with a hyperpure germanium detector: one spectrum 2-3 days after the irradiation and the other after 4 weeks (Almeida et al., 2012a,b). The accuracy of the analytical method was evaluated with the certified reference material NIST-1633a, Coal Fly Ash, revealing results with an agreement of ± 12% (Dung et al., 2010; Almeida et al., 2014b). During the sampling campaign, 6 blank filters were treated the same way as regular samples. All measured species were homogeneously distributed; therefore, concentrations were corrected by subtracting the filter blank contents.

2.1.3.4 Nanoparticle Deposition

Nanoparticles are described as having an increasing surface area with a decreasing particle size for the same amount of mass. Consequently, from the viewpoint of nanoparticle toxicity, the determination of nanoparticle surface area deposited in the human lung is very desirable (Almeida-Silva et al., 2014c) Therefore, in this study, a nanoparticle surface area monitor (NSAM 3550, TSI, USA) was used to measure the lung-deposited surface area of particles which is expressed as square micrometers of lung surface per cubic centimeter of inhaled air ($\mu m^2/cm^3$). This deposition corresponds to the tracheobronchial or alveolar regions of the human lung, according to the International Commission on Radiological Protection deposition model developed by the American

Conference of Governmental Industrial Hygienists (ICRP, 1994). This equipment worked continuously in the studios and in the Bb room and was installed at the same conditions as in chapter 2.1.3.1 but the data was registered every 10 seconds.

2.1.4. Results and Discussion

2.1.4.1 Part 1: IAQ in 11 Fitness Centers

The Portuguese legislation, Portaria no. 353-A/2013, defines indoor air limit values (LV) for the pollutants PM10, PM2.5, CO_2, CO, CH_2O, and VOC, as presented in table 2.2. Nevertheless, it was considered important to include O_3 in this table due to its impact on human health, reactivity with other pollutants, producing submicron particles that contribute to total particulate exposures, and indoor sources (Weschler, 2000). In figure 2.2, the results obtained in this work were compared with the LV based on a color scale where a) green corresponds to levels below 75% of the LV, b) yellow relates to concentrations between 75% of the LV and the limit value and c) red corresponds to values higher than the LV.

Table 2.3 presents the concentrations obtained in the monitoring program that was undertaken in 11 fitness centers from Lisbon. The average and the range values are presented together with the outdoor air measurements.

In G1, G3, G4, G7, and G11, PM10 concentrations exceeded the LV of 50 µg/m^3, representing 55% of the evaluated spaces. Except for the fitness centers G4 and G10, PM10 levels were higher in the outdoor than in the indoor. For PM2.5 only in the fitness center G11 all spaces were classified as green; exceedances of the LV of 25 µg/m^3 were found in G1, G3, and G10.

CO levels were always below the LV defined by the Portuguese legislation. CO is principally associated with infiltrations from the outdoors, garages and combustion processes that are principally related to HVAC systems and water heating (Wang et al., 2012). Higher concentrations of this pollutant were measured in the indoors of G1, G4, G5, G8 and G9. The observed differences between fitness centers may be explained by the localization of the air intakes of the ventilation systems and by the proximity of the gyms to high traffic roads which contributes to the contamination of the indoor air.

Table 2.2 – Limit values of indoor air pollutants defined by the Portuguese legislation, Portaria no. 353-A/2013

Pollutant	Limit value
PM10	50 µg/m^3 (*)
PM2.5	25 µg/m^3 (*)
CO_2	2250 mg/m^3 (**)
CO	10 mg/m^3 (*)
O_3	0.2 mg/m^3 (*)
CH_2O	0.1 mg/m^3 (*)
VOC	0.6 mg/m^3 (*)

(* based on the temporal maximum; ** based on the temporal average)

O_3 levels measured in the fitness centers were very low. A maximum concentration of 0.02 mg/m^3 was measured in G6, G9, G10 and G11. The main O_3 sources in the buildings are the printers (Lee et al., 2001; Destaillats et al., 2008), which are negligible in fitness centers. Outdoors, O_3 occurs as a secondary pollutant, principally as a result from traffic. Therefore, concentrations of this pollutant were always higher outdoor when compared with the indoor environment.

Indoors, the presence of CO_2 is principally associated with occupancy (Apte et al., 2000). In 54% of the studied fitness centers, the LV of 2250 mg/m^3 was exceeded in at least one of the spaces. CO_2 average concentration of all spaces was 2000 mg/m^3. G4 presented the highest average concentrations of CO_2 (4418 mg/m^3) while the maximum value was reached in G5 (5617 mg/m^3 in studio 2). It is not easy to properly characterize the CO_2 present indoors, since its concentration is a function of the occupation of the site, ventilation rates and metabolic activity of the occupants, with these parameters fluctuating with time (Pegas et al., 2011b). CO_2 levels suggested inefficient ventilation of the studied fitness centers.

Since VOC are emitted by consumer products or structures that exist mainly in the indoor environments, such as carpeting, furniture cleaners, paints, perfumes, lacquers and solvents, the concentrations of VOC are usually found to be higher indoors than outdoors (EPA, 2011a). In our study, exceedances of VOC were registered in 82% of the fitness centers and in 64% of the gymnasiums all the spaces presented concentrations higher than the LV.

The highest VOC average concentration was registered in G9 with 3.3 mg/m^3. CH_2O is a VOC, but given its importance due to the related health effects, it is usually assessed in an individualized form

(Hoskins, 2003). However, its indoor sources are also similar to the sources of VOC. In the majority of the cases, the indoor concentrations were higher than outdoors, except the cases of G7, G8, G9 and G10. The highest CH_2O concentrations were found in G4 studios (0.25 mg/m^3 in S1 and 0.21 mg/m^3 in S2) together with high concentrations of VOC that may be originated by the presence of alcohol base hand disinfectant distributed throughout this gymnasium.

Some fitness centers presented high values of some pollutants related to their design and construction. Table 2.3 shows that G5 presented high levels for CO (2.6 mg/m^3 in Bb) and furthermore elevated values for CH_2O (1.4 mg/m^3 in S2 and 1.5 mg/m^3 in Bb) and VOC (2.3 mg/m^3 in S1 and 2.2 mg/m^3 in Bb). The highest VOC and CH_2O concentrations registered in this recently open (2012) fitness center are probably associated with emissions from the new furniture, material and equipment: VOC concentrations analyzed in new apartments demonstrate a decreasing tendency in indoor VOC concentrations over the 24 month follow-up period (Shin and Jo, 2013). Moreover, G5 is located on the ground floor level of a major building, so its elevated CO levels may have resulted from the inappropriate location of the air admissions of the HVAC system, which are placed near the road and close to the pavement.

Table 2.3 – Pollutant concentrations measured in the 11 fitness centers

Gym		CO (mg/m^3)	CO$_2$ (mg/m^3)	PM10 (ug/m^3)	PM2.5 (ug/m^3)	PM1 (ug/m^3)	VOC (mg/m^3)	CH$_2$O (mg/m^3)	O$_3$ (mg/m^3)	T (ºC)	RH (%)
G1	S1	1.5 [1.0-1.7]	2624 [2276-2978]	77 [60-105]	19 [12-31]	8.9 [4.8-16]	-	0.2	0.01 [0-0.02]	21 [19-22]	72 [64-82]
	S2	0.6 [0.3-1.3]	1911 [1511-2682]	54 [41-88]	17 [15-23]	12 [10-15]	-	0.17	0.01 [0-0.01]	21 [21-22]	64 [61-70]
	Bb	1.3 [1.0-1.6]	2542 [2148-2992]	61 [47-74]	17 [14-20]	11 [8.4-13]	-	0.23	0.01 [0-0.01]	22 [22-23]	64 [62-67]
	Out	0.4	861	-	-	-	-	0.19	0.02	18	56
G2	S1	0.89 [0.40-1.40]	1181 [988-1373]	31 [24-39]	10 [10-11]	3.5 [3.04-3.8]	0	0.04	0	17 [17-16]	45 [47-44]
	S2	0.087 [0.00-0.30]	1665 [1564-1860]	47 [34-103]	12 [11-14]	4.4 [3.7-4.8]	0	0.08	0	18 [18-18]	51 [50-52]
	Bb	1.7 [1.6-1.8]	1430 [1363-1557]	33 [29-37]	8.9 [9.2-8.8]	2.8 [2.7-2.8]	0.45 [0.04-0.89]	0.04	0	15 [14-16]	55 [51-55]
	Out	-	-	26 [24-28]	11 [11-11]	3.6 [3.5-3.6]	-	-	-		-
G3	S1	0.31 [0.20-0.40]	1789 [987-2299]	101 [45-153]	23 [16-27]	5.2 [4.3-5.9]	1.2 [0.92-1.4]	0.04	0	18 [17-19]	53 [48-57]
	S2	0	1993 [1813-2299]	89 [63-143]	23 [31-19]	5.6 [5.1-6.9]	1.02 [0.99-1.1]	0.04	0	20 [19-20]	53 [53-53]
	Bb	0.78 [0.40-1.2]	1069 [952-1619]	65 [52-76]	20 [18-21]	4.7 [4.6-4.9]	1.15 [0.94-1.44]	0.04	0	16 [16-17]	50 [49-51]
	Out	1.9 [1.8-2.0]	524 [456-597]	49 [42-55]	11 [12-10]	3.4 [3.5-3.2]	0.87 [0.38-1.09]	0.03	0.01 [0.01-0.02]	12 [11-13]	68 [54-76]
G4	S1	2.6 [2.4-2.7]	2431 [2022-2675]	43 [29-67]	8.9 [8.5-9.7]	2.18 [2.1-2.3]	1.9 [1.7-2.3]	0.25	0	15 [14-16]	73 [67-75]
	S2	1.8 [1.7-2.0]	2042 [1122-2986]	35 [26-50]	9.2 [8.1-10]	2.5 [2.2-2.7]	1.5 [1.4-1.7]	0.21	0	18 [16-19]	56 [54-60]
	Bb	2.2 [1.9-2.4]	4418 [3880-5021]	43 [34-52]	11 [9.4-12]	3.7 [4.2-2.7]	1.7 [1.6-1.9]	0.13	0	20 [19-21]	61 [57-65]
	Out	1.2 [1.1-1.5]	896 [859-905]	51 [39-82]	11 [10-12]	3.4 [3.2-3.5]	0.65 [0.61-0.69]	0.06	0.01 [0.01-0.02]	16 [14-17]	45 [41-50]

Table 2.3 (cont.) – Pollutant concentrations measured in the 11 fitness centers

Gym		CO (mg/m^3)	CO$_2$ (mg/m^3)	PM10 (ug/m^3)	PM2.5 (ug/m^3)	PM1 (ug/m^3)	VOC (mg/m^3)	CH$_2$O (mg/m^3)	O$_3$ (mg/m^3)	T (ºC)	RH (%)
G5	S1	1.8 [1.5-2.2]	2401 [2077-2640]	49 [47-52]	18 [18-18]	6.8 [6.7-6.9]	2.3 [2.1-2.5]	0.10	0	18 [17-18]	77 [74-81]
	S2	1.8 [1.5-2.1]	4109 [2573-5617]	42 [34-54]	6.6 [7.2-6.1]	16 [15-18]	1.8 [1.3-2.2]	1.4	0.01	19 [18-21]	19 [18-20]
	Bb	2.6 [2.4-2.8]	3139 [2945-3341]	37 [31-44]	11 [10-11]	3.4 [3.3-3.5]	2.2 [1.6-2.5]	1.5	0	17 [16-17]	86 [84-90]
	Out	0.66 [0-2.3]	809 [784-835]	37 [34-44]	18 [16-20]	5.2 [3.8-6.8]	0.93 [0.87-1]	0.05	0.02 [0.01-0.04]	13 [11-15]	60 [53-67]
G6	S1	1.3 [1.2-1.4]	1550 [1363-1720]	17 [9-45]	5.8 [4.9-7.7]	3.5 [3.2-4]	2.03 [1.9-2.1]	0.11	0.02 [0.01-0.03]	17 [18-20]	56 [54-58]
	S2	1.01 [0.60-1.2]	3484 [2336-3932]	44 [21-68]	11 [7.4-13]	4.3 [3.7-4.7]	1.7 [1.5-1.9]	0.08	0	19 [18-20]	76 [69-79]
	Bb	1.2 [1.4-1.1]	1414 [1136-1708]	26 [21-37]	7.3 [7-8.6]	4.5 [4.3-4.8]	2 [1.8-2.2]	0.09	0.01 [0-0.02]	19 [18-20]	56 [54-58]
	Out	1.7 [1-2.1]	942 [832-999]	26 [24-28]	11 [11-11]	3.8 [3.7-4.04]	1.6 [1.5-1.7]	0.08	0.03 [0.01-0.04]	12 [11-12]	52 [47-57]
G7	S1	0	1732 [381-2835]	-	-	-	0.50 [0.06-0.57]	0.01	0.19 [0.07-0.82]	17 [16-17]	51 [61-40]
	S2	0	2751 [2078-3330]	84 [21-105]	11 [4.4-13.7]	3.3 [2.2-3.7]	0.57 [0.34-0.74]	0.01	0	16 [16-16]	68 [59-76]
	Bb	0	1660 [1811-1562]	55 [42-68]	15 [12-17]	5.5 [5.9-4.9]	0.37 [0.33-0.43]	0.01	0.01 [0-0-0.2]	17 [16-18]	53 [51-58]
	Out	0.93 [0.0-2.0]	-	11 [9.4-13]	7 [6.3-7.9]	5.4 [5-5.7]	0.34 [0.31-0.37]	0.01	1.7 [0.18-2.9]	8 [6-10]	44 [38-51]
G8	S1	0.018 [0.0-0.20]	3078 [1173-5964]	50 [19-83]	9.2 [6.3-13]	3 [2.6-4.1]	0	0.01	0	18 [17-19]	80 [64-95]
	S2	0.29 [0.20-0.40]	4234 [3803-4694]	56 [43-67]	10 [9.1-11]	3.2 [2.9-3.4]	0	0.01	0	20 [20-21]	74 [70-79]
	Bb	0	1193 [1045-1381]	29 [23-33]	5.7 [5.1-6.8]	2 [1.6-2.8]	0	0.01	0	17 [16-18]	64 [61-66]
	Out	0	-	7.6 [1.7-21]	3.7 [1.7-9.1]	2.1 [1.03-6.7]	0	0.01	0	10 [10-12]	66 [65-68]

Table 2.3 (cont.) – Pollutant concentrations measured in the 11 fitness centers

Gym		CO (mg/m^3)	CO_2 (mg/m^3)	PM10 (ug/m^3)	PM2.5 (ug/m^3)	PM1 (ug/m^3)	VOC (mg/m^3)	CH_2O (mg/m^3)	O_3 (mg/m^3)	T (ºC)	RH (%)
G9	S1	0	1339 [810-1774]	34 [12-49]	7.7 [3.9-14]	4 [2.2-6.5]	2.2 [2-2.5]	0.01	0.01 [0-0.01]	18 [18-19]	73 [71-74]
	S2	0.080 [0.0-0.2]	1266 [860-1735]	17 [4.9-49]	4.1 [1.9-6.9]	2.3 [1.2-3.9]	3.3 [2.9-3.8]	0.02	0.02 [0.01-0.02]	18 [17-19]	71 [67-77]
	Bb	0.10 [0.10-0.10]	2210 [669-3590]	24 [6.8-61]	5.3 [3.2-11]	2.5 [1.5-5.1]	1.9 [1.8-2.2]	0	0.01 [0.01-0.02]	19 [18-20]	70 [66-77]
	Out	0	753 [734-845]	18 [13-23]	4.0 [3.9-4.1]	2.4 [2.4-2.4]	0.65 [0.5-1.2]	0.06	0.06 [0.05-0.09]	22 [22-23]	40 [38-44]
G10	S1	0.28 [0.10-0.50]	1549 [1139-2149]	15 [2.8-25]	12 [10-16]	5.4 [4.7-6.2]	1.8 [1.2-2.3]	0.06	0.02 [0-0.05]	25 [24-25]	43 [41-46]
	S2	0.15 [0.10-0.20]	1277 [984-1482]	3.5 [1.8-8.6]	12 [25-43]	7.3 [1-13]	1.04 [0.95-1.17]	0.06	0.02 [0.01-0.03]	24 [23-25]	44 [40-49]
	Bb	0	1479 [755-2510]	14 [13-18]	14 [13-15.5]	8.1 [7.7-8.8]	1.03 [0.88-1.1]	0.03	0.02 [0.01-0.03]	24 [24-24]	52 [50-54]
	Out	2.3 [1.5-2.9]	899 [748-770]	50 [23-115]	8.8 [6.4-12]	3.8 [3.1-6.7]	2.1 [1.8-2.8]	0.09	0.05 [0.02-0.07]	23 [23-24]	35 [34-36]
G11	S1	0.51 [0.20-0.60]	1116 [673-1652]	79 [57-126]	4 [1-19]	1.5 [3.4-0.7]	1.9 [1.7-2.2]	0.10	0.02 [0.01-0.05]	21 [20-21]	73 [62-85]
	S2	0.53 [0.30-0.80]	1188 [635-1906]	48 [35-71]	1.2 [0.90-2.4]	0.9 [0.74-1.3]	1.8 [1.7-2.02]	0.10	0.02 [0-0.03]	20 [19-21]	76 [74-84]
	Bb	0.68 [0.40-1.10]	1467 [665-2552]	90 [25-71]	7.3 [6.1-8.9]	4.8 [4.1-5.7]	2.5 [2.4-2.5]	0.12	0.01 [0-0-02]	21 [20-21]	67 [66-71]
	Out	1.6 [1.2-2.0]	-	48 [19-108]	15 [12-34]	6.9 [6.4-16]	1.3 [1.2-1.4]	-	0.06 [0.02-0.09]	21 [20-22]	44 [40-47]

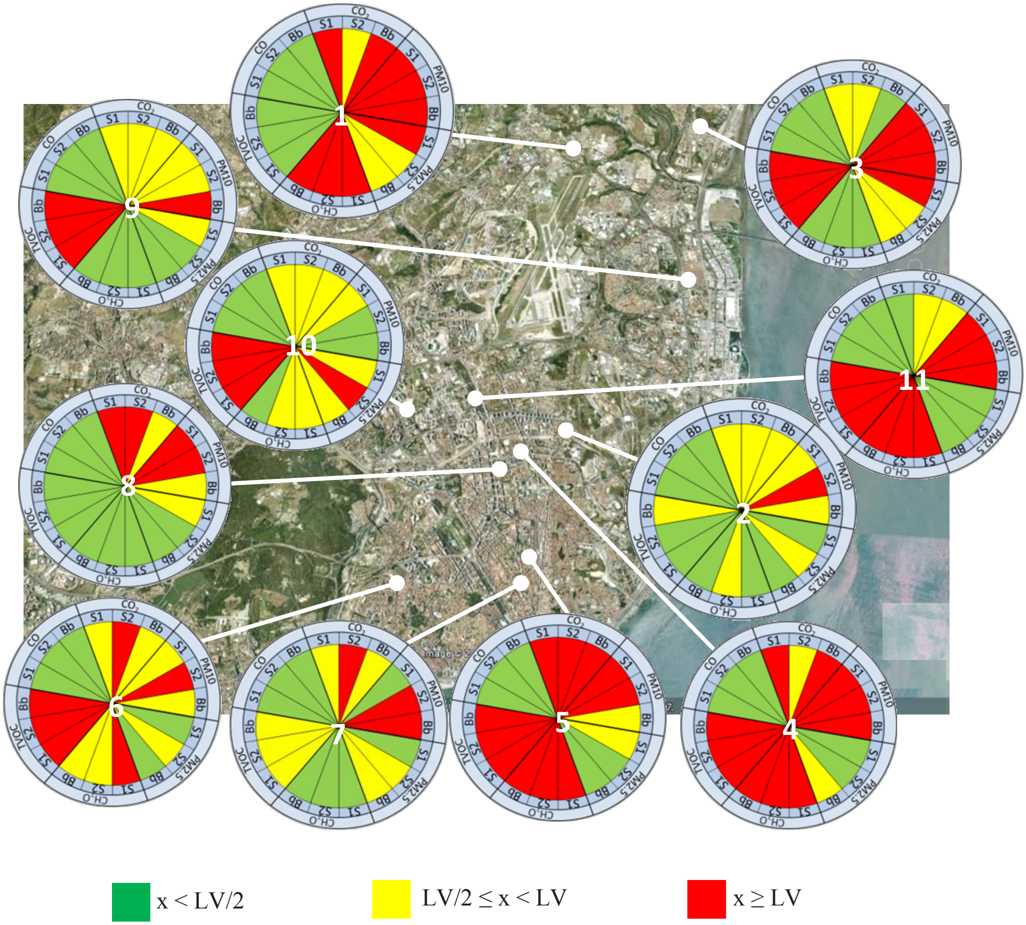


Figure 2.2 – Classification of the fitness centers according to the Portuguese legislation for IAQ (Portaria no. 353-A/2013). S1 and S2 – studios; Bb – bodybuilding room.

2.1.4.1.1 Ventilation rates

Ventilation rates were calculated using the build-up method developed by Hanninen (2012) which is based on the use of CO_2 as a tracer gas. CO_2 represents an advantage comparing with other tracers since it is emitted by occupants and it is inert. This method is based on the curve fit of CO_2 concentrations and requires inputs of: the indoor and outdoor CO_2 concentrations, the number of occupants and the volume of the space (Canha et al. 2013). Air exchange rates (AER) and ventilation rates (VR) were calculated for all the fitness centers. However, these parameters were only estimated for studios because the Bb rooms did not present the required constant number of occupants necessary to run the build-up method.

Table 2.4 presents the AER and the VR and shows that AER varied between 1.4 h^{-1} and 4.4 h^{-1} and VR ranged between 8.9 and 51.5 lps/person. Since the Portuguese legislation (Portaria no. 353-A/2013) defines VR between 13.6 and 27.2 lps/person for fitness centers, the results indicated G1, G2 and G4 did not meet the Portuguese legislation criteria. According to the main national standards in Europe (but despite the lack of unanimity), the AER of 0.5 h^{-1} is defined as a threshold below which associations with poor IAQ may occur (Dimitroulopoulou, 2012). In this study, all fitness centers presented a higher AER.

Table 2.4 – Air exchange rates (h^{-1}) and ventilation rates (lps) in the 11 fitness centers

Gym	Air exchange rate (h^{-1})	Ventilation rate (lps/person)
G1	1.6	8.9
G2	2.1	11.4
G3	3.1	43.1
G4	1.4	10.2
G5	2.3	14.0
G6	3.5	15.3
G7	-	-
G8	-	-
G9	4.4	29.3
G10	1.6	46.7
G11	2.3	51.5

2.1.4.2 Part 2: IAQ Assessment in Three Fitness Centers

Three fitness centers (G9, G10 and G11) were selected in order to perform a deeper IAQ assessment considering longer measurement periods and more parameters.

2.1.4.2.1 Continuous measurements of gases

Figure 2.3 presents the CO_2 concentrations measured in the three fitness centers. A similar trend was observed in all gyms which was characterized by an increase of CO_2 levels in the studios during the occupied period. However, results showed that CO_2 concentrations were influenced not only by the number of people inside the room but also by their metabolic activity during the fitness classes. Figure 2.4 shows the CO_2 growth curve for the same room but in two different fitness classes: Yoga (mind class) and Body Attack (cardio class). Besides the greater number of occupants in the Yoga class (24 people in Yoga versus 20 people in Body Attack), CO_2 concentrations were significantly lower than in Body Attack class. The average CO_2 concentration was 959 mg/m^3 for Yoga and 1774 mg/m^3 for Body Attack. Additionally, the slope of the CO_2 build-up phase in the Body Attack class was higher reflecting a quick growth in the production of this pollutant. In Yoga class, this increase was not observed.

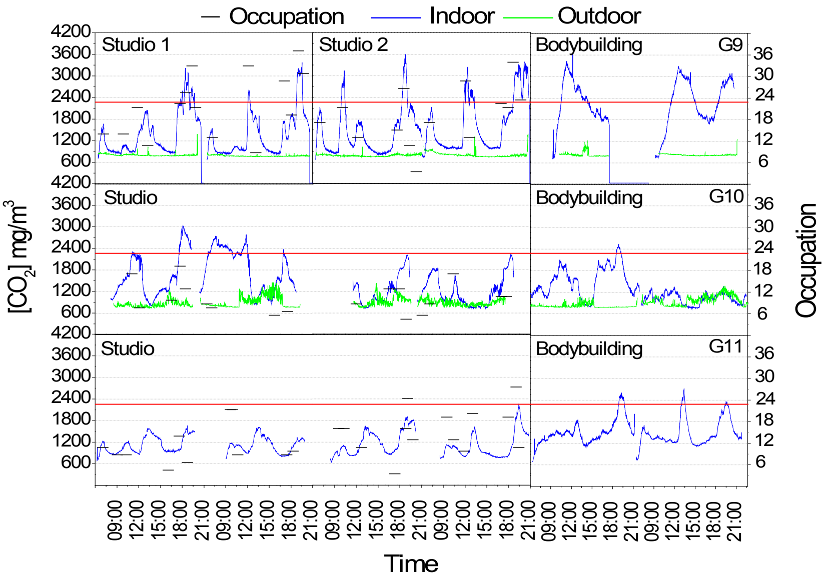


Figure 2.3 – Temporal variation of CO_2 concentration in the 3 fitness centers (values in mg/m^3) and human occupation inside the sites. The horizontal line corresponds to the CO_2 LV defined by the Portuguese legislation.

Inside the bodybuilding rooms, CO_2 concentrations also reflected the degree of occupancy. In G11, the highest CO_2 levels were associated with the cycling classes which occurred inside the bodybuilding room behind a folding screen. Despite not causing toxicity to humans at the registered concentrations (Persily, 1997) CO_2 is a good indicator of IAQ and can influence the human perception of the spaces. Moreover, the performance of people is affected by the concentrations of this gas. Previous studies showed that changes in CO_2 concentrations were associated with statistically significant and meaningful reductions in decision-making performance (Satish et al., 2012).

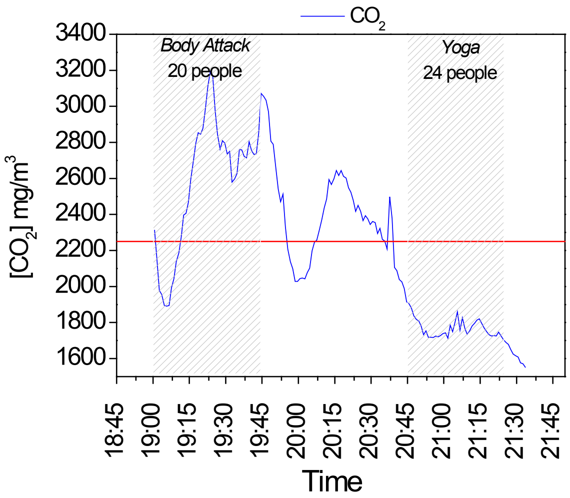


Figure 2.4 – Growth curve of CO_2 in fitness classes associated with different metabolic rates (values in mg/m^3). Shading represents the duration of the classes and the horizontal line corresponds to the CO_2 LV defined by the Portuguese legislation.

Figure 2.5 shows that CO concentrations in the three fitness centers were below the LV (10 mg/m^3) defined by the Portuguese legislation (Portaria no. 353-A/2013). In G9, CO concentration increased in the late afternoon/night which is the period with more entrances and exits in the gymnasium. The air intake for this fitness center is located near the garage, and this fact can explain the highest contamination of indoor air by the traffic during this period. In G10, CO concentrations were higher during the morning due to the presence of trucks, which unload material for an annex building. In G11, the rises in CO levels were also traffic-related and enhanced by the surroundings, since the gymnasium building was walled by other buildings with a height greater than eight floors causing a canyon effect which lead to a lack of pollutant dispersion (Zhou and Levy, 2008). At the registered concentrations, CO does not present harmful health effects to humans, although this pollutant can connect with hemoglobin, replacing the O_2 which in turns reaches the tissues in smaller concentrations (Kao and Nañagas, 2005).

In fitness center G9, the VOC concentrations exceeded the limit value of 0.6 mg/m^3 most of the time (figure 2.6). Figure 2.7 shows that cleaning procedures highly contributed for the increase of VOC concentrations.

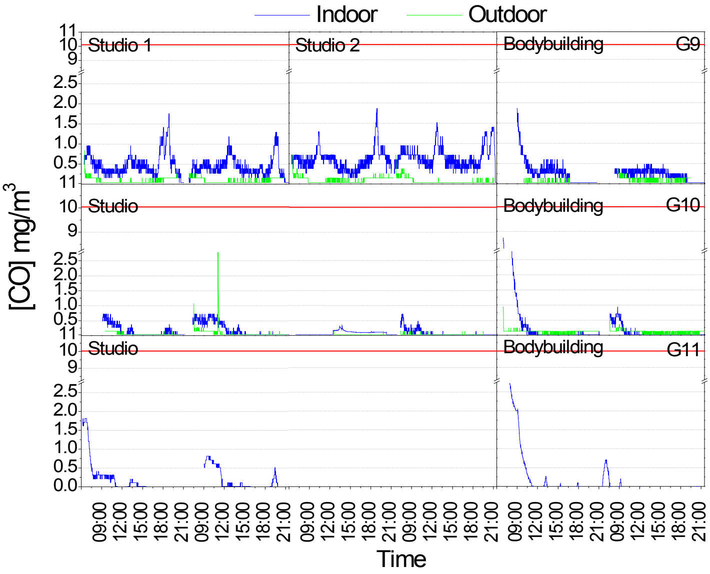


Figure 2.5 – Temporal variation of CO concentration in the 3 fitness centers (values in mg/m^3). The horizontal line corresponds to the CO LV defined by the Portuguese legislation.

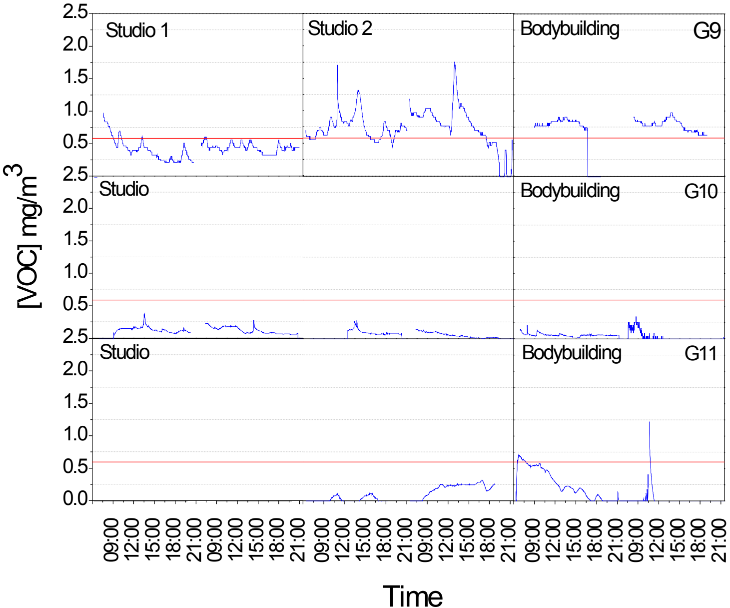


Figure 2.6 – Temporal variation of VOC concentration in the 3 fitness centers (values in mg/m^3). The horizontal line corresponds to the VOC LV defined by the Portuguese legislation.

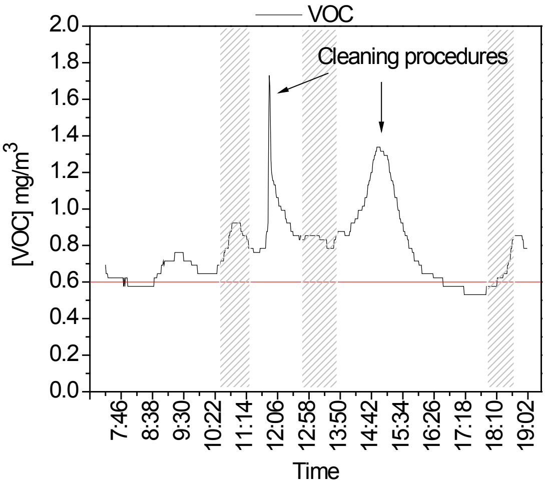


Figure 2.7 – Temporal variation of VOC concentration for a selected period in G9 (values in mg/m^3). Shading represents the duration of the classes and the horizontal line corresponds to the VOC LV defined by the Portuguese legislation.

2.1.4.2.2 Levels of particulate matter

Figure 2.8 presents the temporal distribution of PM, measured in five ranges (PM0.3-0.5, PM0.5-1, PM1-2.5, PM2.5-5 and PM5-10) in the selected fitness centers. In G9 and G10, the PM10 and PM2.5 Portuguese LV of 50 µg/m^3 and 25 µg/m^3, respectively, were exceeded.

The maximum concentrations in G9 for PM10 and PM2.5 were measured in studio 1 (109 µg/m^3 and 30 µg/m^3, respectively). In G10, the maximum PM10 concentrations were 157 µg/m^3 in the studio and 190 µg/m^3 in the bodybuilding room. The maximal PM2.5 value measured in this fitness center was 37.4 µg/m^3 in studio 1.

Results showed that, in the studios, the highest PM concentrations were coincident with the period of fitness classes, revealing a relation between PM concentration and the resuspension of dust caused by the practitioners of physical activity. In scholar gyms, previous studies showed that dust resuspension influenced by students' activity is the major source of coarse particles (Braniš et al., 2011; Buonanno et al., 2013).

The highest concentrations measured in the studios of G10 occurred principally during the cleaning operations performed during the afternoon (approximately at 14:00). Cleaning operations have already been identified as one important source for indoor particle resuspension. Corsi et al., (2008) showed that the resuspension caused by vacuum cleaning can increase PM10 concentrations more than 17 µg/m^3 above the average concentration. Concentrations in the G10 studio increased 8 times in the first day of sampling and 6.5 times in the third day of sampling when compared with the average PM10 concentrations in the space.

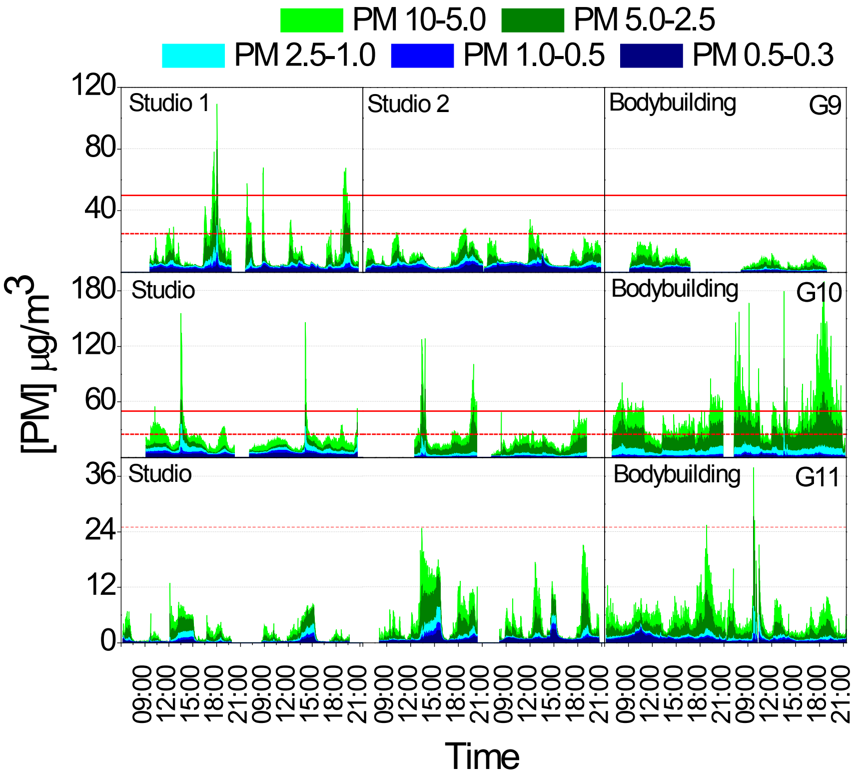


Figure 2.8 – Temporal variation of PM concentration in the 3 fitness centers (values in µg/m^3). The horizontal lines correspond to the PM10 LV defined by the Portuguese legislation (50 µg/m^3) and PM2.5 (25 µg/m^3).

G10 is the only fitness center that opens the windows to ventilate the spaces and this fact was reflected in its highest levels of coarse particles. This gym is placed inside a city park where natural sources of particles, such as soil and pollens, are dominant and contribute principally for the coarse fraction. Canha presented the same conclusion between coarse fraction, natural ventilation and grove vicinity (Canha et al., 2014c).

Figure 2.9 presents the comparison between the indoor and outdoor PM10 total mass concentrations measured by gravimetry. While G9 and G11 presented significantly higher outdoor PM10 concentrations, in G10 the ratio between indoor and outdoor concentrations was closed to 1 or even higher than 1. These results can be explained by the fact that in G9 and G11 the coarser outdoor particles are retained in the filters, presented in the air treatment units from both buildings, whereas in G10 outdoor air enters in the spaces by the windows without any filtration.

Table 2.5 – Indoor and outdoor average element concentrations in the fitness centers G9, G10 and G11 (values in ng/m^3)

	G9		G10		G11		Total	
	I	O	I	O	I	O	I	O
As	0.068 ± 0.032	0.59 ± 0.59	0.25 ± 0.032	0.29 ± 0.041	0.055 ± 0.055	0.41 ± 0.31	0.10 ± 0.088	0.48 ± 0.42
Co	0.06 ± 0.01	0.30 ± 0.22	0.11 ± 0.029	0.14 ± 0.045	0.057 ± 0.049	0.14 ± 0.040	0.076 ± 0.037	0.21 ± 0.16
Cr	1.40 ± 0.86	5.4 ± 3.9	2.47 ± 0.56	3.0 ± 2.1	0.57 ± 0.43	5.5 ± 2.5	1.60 ± 0.98	4.8 ± 2.8
Fe	161 ± 117	1250 ± 1593	375 ± 162	530 ± 350	56 ± 53	771 ± 570	215 ± 190	850 ± 954
K	55±34	480 ± 290	190 ± 22	150 ± 38	51 ± 19	242 ± 257	103 ± 73	291 ± 250
La	0.071 ± 0.0077	0.16 ± 0.14	0.10 ± 0.045	0.16 ± 0.063	0.011 ± 0.010	0.12 ± 0.09	0.064 ± 0.050	0.10 ± 0.09
Na	202 ± 94	640 ± 375	1350 ± 930	1650 ± 760	74 ± 70	691 ± 167	542 ± 770	1020 ± 682
Sb	0.62 ± 0.47	2.61 ± 2.33	0.74 ± 0.16	2.9 ± 1.4	0.19 ± 0.12	2.2 ± 1.7	0.46 ± 0.36	2.5 ± 1.8
Sc	0.0087 ± 0.071	0.014 ± 0.013	0.015 ± 0.0072	0.023 ± 0.0085	<dl	0.0073 ± 0.0099	0.01 ± 0.008	0.02 ± 0.01
Zn	9.7 ± 2.7	74 ± 54	19 ± 11	27 ± 19	5.8 ± 4.7	29 ± 20	12.0 ± 9.0	46 ± 42

Table 2.5 shows the indoor and outdoor concentrations for the chemical elements As, Co, Cr, Fe, K, La, Na, Sb, Sc and Zn measured in the PM10 filters. The outdoor concentrations of these elements were significantly higher than the indoor, except for G10 where significant differences were not observed. This gym has natural ventilation and, consequently, higher contributions of the outdoor elements generated by traffic (As, Sb, Zn), soil (Co, Fe, La and Sc) and sea (Na) were registered (Almeida et al., 2008; Almeida et al., 2009; Almeida et al., 2013b; Freitas et al., 2005). Results showed that, besides the higher outdoor As, Co, Cr, Fe, K and Zn concentrations in G9 and G11, the air filtration by their air treatment units allowed the retention of particles and, therefore, the capture of these elements.

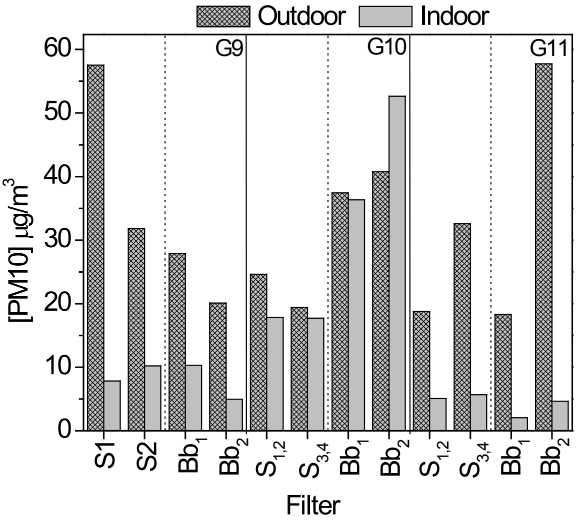


Figure 2.9 – PM10 concentrations measured indoor and outdoor of the fitness centers (values in µg/m³). (S1 – Studio1; S2 – Studio 2; $S_{1,2}$ – First and second day of sampling; $S_{3,4}$ – third and fourth day of sampling; Bb_1 – First day of sampling in the bodybuilding; Bb_2 – Second day of sampling in the bodybuilding)

The crustal enrichment factor method has been used as an attempt to evaluate the strength of the crustal and non-crustal origin of the elements. Enrichment Factors (EF), using Fe as a crustal reference element (EF_{Fe}), were calculated based on equation 1 and using soil composition (Mason and Moore, 1982):

$$EF_{Fe} = \frac{\left(\frac{[x]}{[Fe]}\right)_{PM}}{\left(\frac{[x]}{[Fe]}\right)_{soil}} \qquad \text{(equation 1)}$$

Elements with EF_{Fe} values that approach unity can be considered predominantly from soil, whereas if the evaluated element has EF values higher than 10, its provenance is asserted mainly to local, regional and/or long transportation phenomena from other natural and/or anthropogenic sources (Farinha et al., 2004). The EF_{Fe}, presented in figure 2.10 , indicate that, both in indoor and outdoor, the elements Sc, La, Co, K, Fe and Cr were associated with soil emissions ($EF_{Fe}<10$) while As, Sb and Zn were related to anthropogenic emissions ($EF_{Fe}>10$).

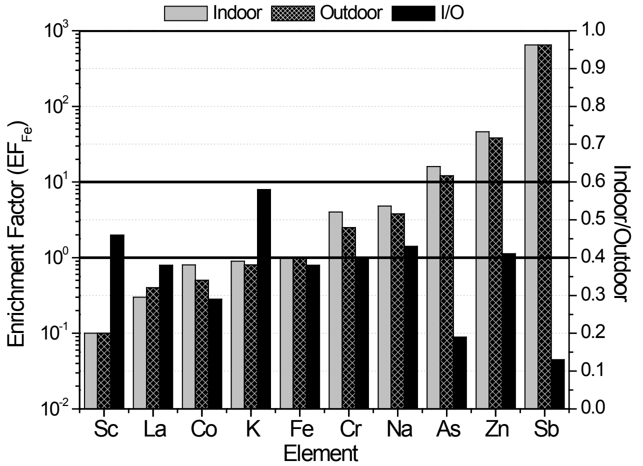


Figure 2.10 – Enrichment factor using Fe as a reference element and Mason and Moore (1982) soil composition and ratio indoor/outdoor.

2.1.4.2.3 Nanoparticle lung deposition

The estimated total deposited alveolar area and the total deposited surface area were calculated for a lung surface area of 80m², which is the defined area for an adult. Table 2.6 shows that the minimum value for the deposited alveolar area was reached in G11 with 13µm²/cm³ and the maximum was registered in G10 with 39 µm²/cm³. As these measurements were performed for the first time in fitness centers, levels were compared with studies performed in other indoor environments. In schools, Buonanno et al. (2012) registered higher alveolar area levels deposits which ranged between 35 µm²/cm³ and 150 µm²/cm³. In elderly care centers, Almeida-Silva et al. (2014c) found values between 10 µm²/cm³ and 46µm²/cm³ and in houses, Gomes et al. (2012) recorded an average value of 29 ± 1.0 µm²/cm³ and Ntziachristos et al. (2007) registered an average value of 45 ± 26 µm²/cm³.

Table 2.6 – Average deposited area and total deposited surface area in the fitness centers G9, G10 and G11.

Fitness Center	Average Deposited Area (µm²/cm³)	Total Deposited Surface Area (µm²)
G9	28.61 ± 25.40	1.93×10^7
G10	39.17 ± 15.95	2.37×10^7
G11	13.47 ± 6.12	7.99×10^8

2.1.5 Conclusions

This work conducted a comprehensive characterization of a vast array of indoor pollutants in 11 fitness centers and identified sources that compromise IAQ.

The high CO_2 levels registered within this study and the calculated ventilation rates indicated that, in general, the fitness centers have inefficient ventilation, considering the type of activity that is preconized indoors. This fact influences the human perception of the space and gives the feeling of discomfort during the practice of sports. Taking into account that VOC spikes were observed during cleaning activities and that cleaning products are recognized as risk factors for respiratory health, low emitting agents and “green” practices should be adopted. The levels of particles were highly influenced by the intense indoor activities and by the type of ventilation. Results showed that the location of the air intakes and the efficiency of the air filtration are essential for the maintenance of a good IAQ.

Taking into account the unique characteristics of the fitness centers – intense indoor activities, large number of people who are more susceptible to air pollutants during exercise, insufficient ventilation and relatively small room sizes – there is a need to better assess the exposure and inhaled doses by gyms practitioners in order to minimize adverse health effects and to potentiate the benefits of the physical activity.

2.2 Characterizing the fungal and bacterial microflora and concentrations in fitness centers

Based on the article:
Characterizing the fungal and bacterial microflora and concentrations in fitness centers
C.A. Ramos, C. Viegas, S. Cabo Verde, H. T. Wolterbeek, S. M. Almeida
Indoor and Built Environment, DOI: 10.1177/1420326X15587954

2.2.3 Abstract

Fitness centers are special places which gather conditions for microbiological proliferation. Moisture due to perspiration and water condensation, marked human presence, elevated physical activity and contact between the occupants and surfaces are circumstances that meet in this types of buildings. Exposure to microbial contaminants is clinically associated with respiratory disorders and people who work out in polluted environments are more susceptible to contaminants. This work studied the indoor air contamination in three gymnasiums in the city of Lisbon. The sampling was performed at two periods of the day: at the opening (morning) and closure (night) of the gymnasiums. The airborne bacterial and fungal populations were sampled by impaction directly onto Tryptic Soy Agar (for bacteria) and Malt Extract Agar (for fungi) plates, using a Merck MAS-100 air sampler. An increase in bacterial concentrations in the night compared with the morning was verified but the same behavior was not found for fungal concentrations. Gram-negative catalase positive cocci were the dominant morphological type of bacteria within indoor air samples of all gymnasiums. In this study, 21 genera/species of fungal colonies were identified. *Chrysosporium* sp., *Chrysonilia* sp., *Neoscytalidium hialinum*, *Sepedonium* sp. and *Penicillium* sp. were the most prevalent species identified in the morning, while *Cladosporium* sp., *Penicillium* sp., *Chrysosporium* sp., *Acremonium* sp. and *Chrysonilia* sp. were more prevalent in the night. Bacteria were associated with human presence while fungi were linked with outdoor contamination. A well designed sanitation and maintenance program of the gymnasiums will promote a heathier space for indoor physical activity.

2.2.4 Introduction

Within indoor air, there is a complex mixture of viable and non-viable particles. The non-viable include inorganic particles, such as metals and other chemical compounds, and organic non-reactive material. The viable components are those who are capable of growing under favorable conditions, such as bacteria, fungi and all other microorganisms. Bioaerosols are normally defined as “particles with biological origin suspended in the air”, which can cause health effects, especially in the upper airways (Bünger et al., 2000; Heldal et al., 2003; Eduard and Halstensen, 2009).

The indoor microbial pollution involves hundreds of species of bacteria and fungi growing inside buildings when specific conditions are available. The main factors that influence microbial growth in a building are moisture, temperature and nutrient availability. The ventilation rate for air renewal is also a crucial factor for the control of microbial growth. In fitness centers, moisture due to perspiration and water condensation, marked human presence, elevated physical activity that promotes the resuspension of dust from the ground and contact between the occupants and surfaces (pavement, fitness equipment) are conditions that promote the microbial growth. Fungi are ubiquitous microorganisms that proliferate in more diverse environments due to their lower water activity (a_w) than bacteria. Bacteria require an a_w above 0.80, while fungi present minimum a_w of approximately 0.70 (Beuchat et al., 2013). Moreover, fungi are less selective in what concerns the substrate and consequently are able to grow on a diverse range of surfaces (wood, wall paper, etc.). Combined with these growth conditions joins the fact of the existence of fungal spore colonies that are easily released into the air through aerial hyphae, while in the case of bacteria, this process is not easy to promote due to its gelatinous colonies.

Exposure to microbial contaminants is clinically associated with respiratory symptoms, allergies, asthma, and immune reactions (WHO, 2009a) depending upon the nature of the microbiological agent and the host´s immune status. Some species of gram-negative bacteria are of most concern when present in indoor air because they are producers of endotoxins that can cause respiratory symptoms, including non-allergic asthma (WHO, 2009a). Gram-positive bacteria represent the largest group present in the atmosphere due to their greater resistance and survival abilities (Fang et al., 2007; Shaffer and Lightart, 1997). Fungi species among *Aspergillus, Penicillium* and *Fusarium* genera are producers of mycotoxins which can enter the human body by inhalation, dermal and oral contact, thereby causing different reactions in the host organism (Jarvies and Miller, 2005).

The reasons why athletes and the common individual that practice sport present a higher risk of contact with bioaerosols and pollution were previously described (Carlisle and Sharp 2001). The aim of this work was to assess indoor air contamination in three gymnasiums, by fungal identification and bacteria characterization in order to estimate the potential biological hazards during sporting practice in fitness centers.

2.2.5 Methodology

2.2.5.1 Sampling Sites

The three gymnasiums selected to perform the deeper IAQ assessment, presented in chapter 2.1 (G9, G10 and G11), were used to assess the microbial load. Inside the fitness centers, sampling sites were chosen: the studios and the bodybuilding rooms. In G10 and G11, only the studio with the most practicing fitness classes was monitored, whereas in G9, two studios were evaluated. As described in table 2.7, all fitness centers have identical location besides having a different surrounding.

All fitness centers have mechanical ventilation, however G10 preferentially uses natural ventilation over mechanical as it was observed that it was often switched off. The sampling campaigns were performed between October and December of 2012.

2.2.5.2 Air Sampling

Samples were collected in two periods of the day – in the morning (at the opening of the gymnasium) and at night (at the closure) – in order to recognize the differences before and after occupancy. Air samples were collected at the center of the studied room, at ground level.

Air sampling was conducted using a microbial air sampler (MAS-100, Merck Millipore, Germany) that collected, by impaction, 250L of air in each plate, with a flow rate of 100L/min. Two different culture medias were used in order to provide to the microorganisms the most suitable nutrients for their growth: Malt Extract Agar (MEA) (supplemented with 0.1g/L chloramphenicol), used for fungi, and Tryptic-Soy Agar (TSA), used for bacteria. TSA is a general agar medium used for culturing many kinds of non-fastidious and moderately fastidious microorganisms (Nunes et al., 2013).

The sampling was also performed outdoors to compare the results between the indoor and the outdoor environments. The samples were sealed with parafilm and transported to the laboratory in a cooler bag. Air sample culture plates were incubated at 30°C between 5 to 7 days (Memmert oven, Germany). A total of 48 petri dishes with bacterial colonies and 48 petri dishes with fungal colonies were analyzed. The colony counts were corrected using the positive hole correction table MAS-100, provided by the supplier. The microbiological concentrations were expressed in colonies forming units per cubic meter (CFU/m^3).

Table 2.7 – Main characteristics of the studied sites of the gymnasiums

Gym	Code	Sampling Days	Capacity (person) [b]	Cleaning Operations
G9	G9S1	1 day	35	In the middle of morning and afternoon In the closure time
	G9S2	1 day	35	
	$G9Bb_1$	2 days	60	In the closure time
	$G9Bb_2$			
G10	$G10S_1$	2 days	40	In the middle of the day In the closure time
	$G10S_2$			
	$G10Bb_1$	2 days	40	In the closure time
	$G10Bb_2$			
G11	$G11S_1$	2 days	35	In the middle of the morning and afternoon In the closure time
	$G11S_2$			
	$G11Bb_1$	2 days	100	In the closure time
	$G11Bb_2$			

[a] S1 – Studio 1, S2 – Studio 2; Bb – Bodybuilding. [b] Maximum capacity.

A Greywolf (IAQ 610,WolfSense Solutions, USA) was used to continuously monitor the comfort parameters (temperature, relative humidity and CO_2) inside the rooms during the sampling days, from the opening to the closure of the gymnasiums. Outdoor meteorological data was obtained from Aeroporto weather station located in the center of Lisbon (38°46' N, 9°08' W), which data is available online (Russia's Weather server). MAS-100 and Greywolf were calibrated according to fabricant specifications.

2.2.5.3 Microbial Characterization

Fungal colonies were grouped by macroscopic colony characteristics (e.g. color, shape and elevation). For fungal identification, microscopic mounts were performed using tease mount or Scotch tape mount and lactophenol cotton blue mount procedures. Morphological identification was achieved through macro and microscopic characteristics as noted by de Hoog et al. (2000).

The obtained bacterial isolates were characterized based on their macroscopic traits (e.g. pigmentation, texture, and shape), microscopic morphology (cellular morphology, and presence/ absence of endospores) and biochemical characteristics (gram staining, catalase and oxidase activities). For the morphological characterization, bacteria were isolated in TSA medium and incubated at 30 ºC for 24 h. The isolates were grouped into morphological types based on their characteristics. The definition of the morphological types was based on the Bergey's Manual of Determinative Bacteriology (Holt, 1994). The frequency of each morphological type was calculated based on the number of isolates obtained and on their characters.

2.2.5.4 National Guidelines for Bioaressols

In Portugal, a recent legislation established new limit values for microbiological contamination in indoor environments (Portaria no. 353-A/2013), replacing the previous diploma (Decreto-Lei no. 79/2006). In the previous legislation, a critical limit of 500 CFU/m^3 was defined as the threshold for bacteria and fungi concentrations. Currently, the legal compliance is different concerning the type of microorganism. For fungi, indoor concentrations should be less than outdoor concentrations; and for bacteria, the indoor concentration should not exceed the outdoor concentration by 350 CFU/m^3. However, when these situations are not fulfilled, there is a second opportunity to satisfy the legal requirements according to Table 2.8 and Table 2.9.

Table 2.8 – Portuguese legal compliance for microbiological parameters according to Portaria no. 353-A/2013.

	Fungi	Bacteria
1st requirement	• [indoor] < [outdoor]	• [indoor] + 350 CFU/m^3 < [outdoor]
2nd requirement (to be applied when the 1st requirement is not fullfield)	• No visible fungal growth on surfaces; • Species should be evaluated according table 3	• [indoor] + 350 CFU/m^3 > [outdoor] and [CO_2] < 1800mg/m^3; • Ratio between gram negative bacteria and total bacteria should be less than 0.5.

Once the critical limit of 500 CFU/m^3 was applied by other authors and guidelines (ACGIH 1989; Reynolds et al. 1990; The Government of Hog Kong 2003) and because the sampling campaigns were performed when the previous legislation was in force, the legal compliance will approach the old and the new requirements.

Table 2.9 – Fungal conformity based on the species according to Portaria no. 353-A/2013.

Species		Specific Condition of Conformity
Common species	*Cladosporium spp* *Penicillium spp* *Aspergillus spp* *Alternaria spp* *Eurotium spp* *Paecilomyces spp* *Wallemia spp..*	Mixture of species: ≤ 500 CFU/m^3
Non-common species	*Acremonium spp* *Chrysonilia spp* *Tricothecium spp* *Curvularia spp* *Nigrospora spp*	One specie: < 50 CFU/m^3 Mixture of species: < 150 CFU/m^3
Pathogenic species	*Chryptococcus neoformans* *Histoplasma capsulatum* *Blastomyces dermatitidis* *Coccidioides immitis*	Absence of any species
Toxigenic species	*Stachybotrys chartarum* *Aspergillus versicolor* *Aspergillus flavus* *Aspergillus ochraceus* *Aspergillus terreus* *Aspergillus fumigatus* *Fusarium moniliforme* *Fusarium culmorum* *Trichoderma viride*	One specie: < 12 CFU/m^3 (Several colonies per plate)

2.2.5.5 Statistical analysis

The Origin7.5® software was used to compute graphical figures and the Statistica® software was used to calculate the statistical tests.

2.2.6 Results and Discussion

2.2.6.1 Comfort Parameters

According to the comfort criteria defined by the ISO 7730:2005, the temperature should range between 23°C and 26°C and the relative humidity should vary between 30% and 70%. Table 2.10 presents the temperature and relative humidity measured in the three fitness centers during the sampling campaigns. Temperature varied between 10ºC and 27ºC with the greatest humidity levels recorded/observed in G9 (80%), exceeding the comfort criteria defined by ISO 7730:2005. The highest values for these parameters were recorded during occupancy of the spaces (Ramos, 2013).

CO_2 concentration was used not only as an indicator of ventilation efficiency, comfort and excess of occupancy and but also to evaluate the microbiological compliance according the Portuguese legislation. Table 2.10 depicts the variation of indoor CO_2 concentrations measured during the sampling campaigns. CO_2 varied between 398 mg/m^3 and 3590 mg/m^3 and showed a strong correlation between high occupancy and HVAC systems (Ramos et al., 2014). Higher CO_2 concentrations were observed during periods of physical activities within the studios.

Table 2.10 – Comfort parameters (temperature, relative humidity) and CO_2 measured in the fitness centers.

Fitness Center	Sampling Site	Temperature (ºC)		Relative Humidity (%RH)		CO_2 (mg/m^3)	
		$\bar{x} \pm \sigma$	range	$\bar{x} \pm \sigma$	range	$\bar{x} \pm \sigma$	range
G9	Studio 1	18 ± 0.88	16 – 21	80 ± 4.1	73 – 93	1147 ± 502	577 – 3350
	Studio 2	19 ± 1.01	16 – 22	78 ± 5.2	71 – 94	1315 ± 591	613 – 3584
	Bodybuilding	19 ± 0.35	18 – 20	72 ± 2.6	67 – 86	1882 ± 553	611 – 3590
G10	Studio	23 ± 1.7	19 – 26	58 ± 7.07	43 – 72	1185 ± 587	398 – 3005
	Bodybuilding	21 ± 1.3	17 – 23	59 ± 7.8	46 – 81	1015 ± 219	709 – 2510
G11	Studio	20 ± 1.1	15 – 27	69 ± 5.2	56 – 85	1122 ± 289	635 – 2195
	Bodybuilding	20 ± 1.1	10 – 22	57 ± 5.6	49 – 99	1456 ± 355	655 – 2685

2.2.6.2 Total Bacteria and Fungi Concentrations

Figures 2.11 and 2.12 illustrate the indoor and outdoor concentrations of bacteria and fungi in the fitness centers.

Bacterial concentrations exceed the outdoor concentrations by 350 CFU/m^3 during the night period in the studio of G10 in the second day of sampling (556 CFU/m^3 indoor and 56 CFU/m^3 outdoor) and in the bodybuilding room of G11 in the first day of sampling (824 CFU/m^3 indoor and 60 CFU.m^{-3} outdoor). In the above situations, the critical limit of 500 CFU/m^3 was also exceeded. Results showed that at the end of the day, the bacterial load was significantly higher indoors than outdoors, indicating the importance of the occupation for the bacterial development.

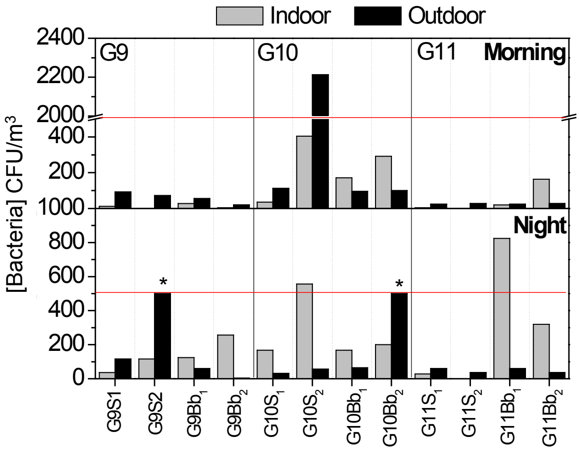


Figure 2.11 – Concentrations of airborne bacteria measured indoors and outdoors of analyzed fitness centers. Results provided for each sampling site and sampling period. The horizontal line establishes the critical limit of 500 CFU/m^3. The * indicates that the number of colonies were countless and therefore a concentration above 500 CFU/m^3 was assumed.

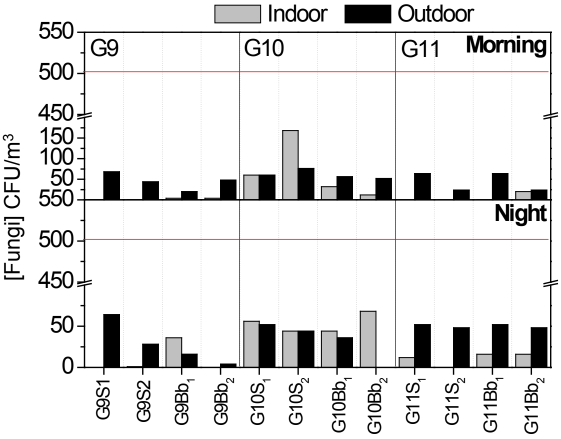


Figure 2.12 – Concentrations of airborne fungi measured indoors and outdoors of analyzed fitness centers. Results provided for each sampling site and sampling period. The horizontal line establishes the critical limit of 500 CFU/m^3.

For fungal concentrations, indoor concentrations were greater than outdoor concentrations in G10 in six measurements (sampling performed in the studio in the morning period and in all sampling performed at the end of the day) and in G9 in the bodybuilding room at night. Regarding the old guidelines, the critical limit value of 500 CFU/m^3 was not exceeded in any situation. Results show that the highest concentrations were registered in G10 that uses natural ventilation, a phenomena also observed by Frankel et al.. In G9 and G11, outdoor particles are retained in the filters, placed in the air treatment units from both buildings, whereas in G10, outdoor air enters through the window spaces without any filtration. Moreover, in G9 and G11, the mechanical ventilation is more efficient in promoting pollutant dilution (Canha et al., 2013).

In general, there was an increase in bacterial load at night that was not observed with fungi. This suggests that the bacteria are more associated with human occupancy than fungi. The presence of bacteria indoors might be associated with deposited dust (Hospodsky et al., 2012), skin cells and hair (Clark et al., 1973). These results are in agreement with the ones presented by Dacarro et al. (2003) that studied the microbial load in universities and school gyms during physical education classes.

2.2.6.3 Identification of Fungal Species

The identification of fungal species is very important for the study of fungal contamination since it allows the differentiation between benign and harmful species (Hoog et al., 2000; Rao et al., 1996; Kemp et al., 2002).

As represented in figure 2.13, indoors, *Chrysosporium* sp. *re*presented 56% of the fungal genera in the morning, decreasing it's presence to 10% in the night, while *Cladosporium* sp. clearly emerged in the night time with a prevalence of approximately 51%. *Chrysonilia* sp. decreased it's prevalence from 27% in the morning to 5.8 % in the closing time; with the same behavior registered outdoors, decreasing from 35% to 18%. *Penicillium* sp. increase indoors between the two studied periods (1.9% to 10%), although this increase was also found outdoors (10% to 16%). *Acremonium* sp. was only identified indoors at the end of the day (7.5%). In fact, the most prevalent fungal genera found in our study coincided with other studies. *Cladosporium* sp. was widely found as the dominant genera inside buildings in many works (Jafta et al., 2012; Baxi et al., 2003; Celtik et al., 2011, Soleimani et al., 2013, Oliveira et al. 2009). Regarding sport facilities, a study conducted in a sports hall in China indicated that the dominate genera's indoors were *Cladosporium* sp., *Penicillium* sp., *Aspergillus* sp. and *Alternaria* sp., making up 95% of the total observed genera (Xie et al., 2009). Viegas et al. (2010) described *Cladosporium* sp. as the principal isolated genera in a gymnasium, followed by *Penicillium* sp., *Aspergillus* sp., *Mucor* sp., *Phoma* sp. and *Crysonilia* sp.. In a study conducted in Barcelona houses, *Cladosporium* sp. was found to have the greatest indoor concentrations during autumn (de Ana et al., 2006), with the same trend found in infant bedrooms in the USA (Ren et al., 2001). Species of *Cladosporium* sp. are largely distributed, commonly encountered on all kinds of plants and on debris and are frequently isolated from soil, food, paint, textiles and other organic matter (Bensch et al., 2012), which can justify the existence of high prevalence of this fungi indoors in the end of the day because of the passage of people throughout the day who bring debris from outdoors.

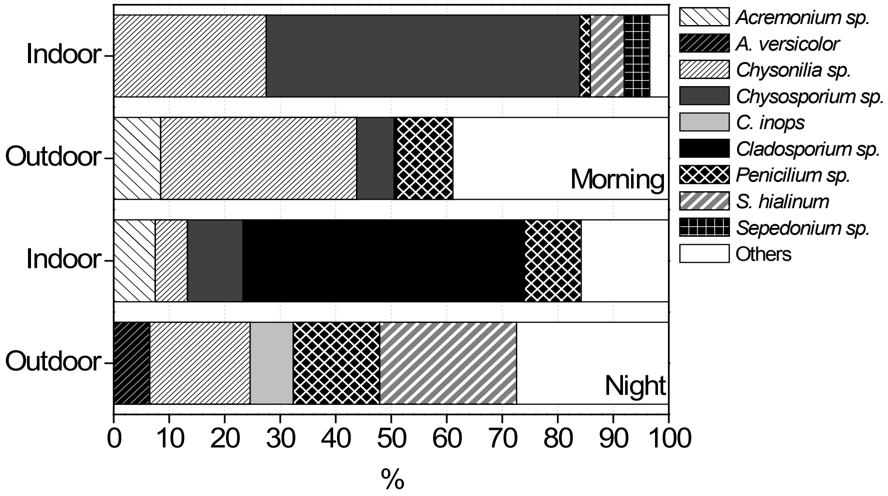


Figure 2.13 – Distribution of fungal species indoor and outdoor in the two periods of sampling (morning and night).

A total of 22 genera and 27 species of fungal colonies were identified in this study. In Table 2.11, the fungal species found indoors and outdoors, discriminated by fitness center, are presented for the two periods of sampling. Significant statistical differences were found in relation to the indoor and outdoor concentrations, for G9 and G11 (Wilcoxon Matched Pairs test, G9, *p*=0.03; G10, *p*=0.89; G11, *p*=0.01), although no significant statistical differences were found between the indoor concentrations among gymnasiums, (Mann-Whitney test, *p*=1 for all tests). In G9, two fungal species were identified (*Penicillium* sp. and one belonging to *A. fumigatus* complex), while in G10, twelve different genera and three species were found (*Acremonium* sp., *Chrysonilia* sp., *Chrysosporium* sp.*, Cladosporium* sp., *Penicillium* sp.*, Geotrichum* sp.*, Mucor* sp., *Neoscytalidium* sp., *N. dimidiatum, N. hialinum, Rhodotorula* sp.*, Sepedonium* sp., *Syncephalastrum recemosum, Scopulariopsis* sp.*, Phoma* sp.). In G11, seven species were identified (one belonging to *A. ustus* complex*, Alternaria* sp., *Aureobasidium* sp., *Geotrichum* sp*., Penicillium* sp., *S. brevicaulis, N. hialinum*). As observed, the fungal load in G10 was higher than in the other fitness centers. This can be explained by the fact that in G9 and G11, the fungi that come from outdoors are retained in the filters, whereas in G10, outdoor air enters in the rooms by the windows without any filtration. As emphasized in the studies of Frankel et al. (2012) and Kemp et al. (2002), outdoor air is the main source of indoor fungi in healthy buildings.

Toxic species were found in G9 and G11 indoors, such as *Aspergillus* genus, namely belonging to *A. fumigatus* complex (Land et al., 1989; Orcioulo et al., 2007; dos Santos et al., 2002; El-Shanawany et al., 2005; Fischer et al., 1999), which is considered an indicator of moisture-damaged in buildings (Samson et al., 1994), and another belonging to *A. ustus* complex. Fungal identification revealed one potentially dangerous situation (defined according table 2.9) in the G10 studio associated with the presence of *Chrysonilia* sp. at 72 CFU/m^3. *Chrysonilia* sp. is considered a non-common species and is known to induce asthma (Francuz et al., 2010; Monzón et al., 2009; Tarlo et al., 1996). In all the assessed gymnasiums, no signal of fungal growth was detected on the walls, furniture or in other materials.

Concerning the colonies concentration found indoor, there was an increase between the morning and night that resulted in 7 new species in G10 and 6 new species in G11. Some of these new isolates (*A. ustus* complex*, Acremonium* sp.*, Alternaria* sp.*, Aureobasidium* sp.*, Cladosporium* sp.*, Geotrichum* sp.*, Mucor* sp.*, Neoscytalidium* sp.*, N. hialinum, Phoma* sp. *and S. brevicaulis*) can produce toxic compounds (metabolites or mycotoxins), though few of their metabolites have been shown to be produced in natural indoor environments (Jarvies and Miller, 2005).

2.2.6.4 Bacteria Characterization

Phenotypic characterization of the most prevalent isolates, collected by impaction in TSA medium, allowed for the identification of seven morphological groups, as summarized in table 2.12. Observing bacterial morphology and the Gram reaction usually constitutes the first stage of identification and is very useful for the preliminary identification of bacterial species. The traditional methods that employ observation of either single cell morphology or colony characteristics remain reliable parameters for the identification of bacterial species and still have significant taxonomic value (Tshikhudo et al., 2013).

Table 2.11 – Distribution of fungal species indoor and outdoor in the two periods of sampling (morning and night). In bold are identified the five most prevalent fungal species in the morning (M) and in the night (N), both indoor and outdoor. Results in CFU/m^3.

Colonies	G9				G10				G11			
	Indoor		Outdoor		Indoor		Outdoor		Indoor		Outdoor	
	M	N	M	N	M	N	M	N	M	N	M	N
A. flavus complex								4				
A. fumigatus complex	4		11	4							8	8
A. niger complex			4				4					
Circumdati complex												8
A. ustus complex										4		
A. versicolor complex				20								
Acremonium sp.						**36**	20	8			40	
Alternaria sp.								24		4	16	
Aureobasidium sp.				12						4	16	8
Botrytis sp.							4					
Chrysonilia sp.			**35**	**12**	**72**	**28**	**56**	**32**			**24**	**24**
Chrysosporium sp.			**4**		**148**	**48**	28					4
C.inops			4	16				8				
Cladosporium sp.			8			**244**						4
Eurotium sp.							4					
Fusarium poae							8					
Geotrichum sp.				16		12	8			4		
Mucor sp.				4		8		16				
Neoscytalidium sp.						8	12					
N. dimidiatum						4	4					
N. hialinum				8	16	4	**28**	**32**		**8**		**56**
Paecilomyces sp.											24	
Penicillium sp.	**4**	**9**			**4**	**24**	**20**	**4**	**20**	**16**	**48**	**88**
Phoma sp.						4		4				
Rhodotorula sp.				4	4	8	12					
Scedosporium sp.			4				32					
Scopulariopsis sp.					4							
S. brevicaulis										4		
Sepedonium sp.					12							
Syncephalastrum racemosum					12							
TOTAL	8	9	70	96	272	428	240	132	20	44	176	200

Therefore, and despite being described by several authors as old fashioned, bacterial morphological characterization can provide valuable insights into individual microbial diversity, derived from either genetic or reversible changes (Sousa et al. 2013). Several morphotypes have been identified in bacteria related to chronic and acute infections, and specific phenotypic traits are important clinical features (Proctor et al., 2004; Hogardt and Heesemann, 2000; Hilmi et al., 2013).

According to the national legislation, when indoor concentrations exceed the outdoor concentrations by 350 CFU/m^3, the ratio between the Gram-negative and the total bacteria should be less than 0.5. In the second day of sampling in G10 studio 2, the concentration of Gram-negative bacteria was calculated at 540 CFU/m^3 in a total of 556 CFU/m^3, resulting in a ratio of 0.9. In G11 bodybuilding room 1, the concentration of Gram-negative bacteria was calculated at 632 CFU/m^3 in a total of 824 CFU/m^3, giving a ratio of 0.7. Resultantly, both locations failed to comply with the national legal compliance.

Table 2.12 – Frequencies of the isolated morphological groups (%)

Morphological type	G9		G10		G11	
	I	O	I	O	I	O
Gram-positive, catalase-positive cocci	3.2	5.1	0.11		1.7	
Gram-negative, catalase-positive cocci	25	58	30	55	30	38
Gram-negative, catalase-negative cocci	1.4				48	38
Non-spore forming, Gram-positive, catalase-positive bacilli	0.13	3.4				
Non-spore forming, Gram-positive, catalase-negative bacilli	2		0.16			6.3
Gram-negative, oxidase-positive bacilli		8.5	0.05	5.8		
Gram-negative, oxidase-negative bacilli	1.2	25		20	20	19

Our results indicated that Gram-negative, catalase-positive cocci were the most prevalent airborne bacterial morphological type indoors (25% in G9, 30% in G10 and 30% in G11) and outdoors (55% in G9, 30% in G10 and 38% in G11) of all fitness centers. In a study of culturable airborne bacteria by the US Environmental Protection Agency in the Building Assessment Survey and Evaluation (BASE) (Tsai and Macher, 2005), Gram-negative cocci were also found to be present within office buildings. The main source of Gram-negative bacteria is from settled dust (Bouillard et al., 2005), brought into fitness centers by users, with the concentration of indoor particles effected by the levels of human occupancy (Hospodsky et al., 2012; Canha et al., 2014a; Almeida-Silva et al., 2014b). Contamination can also be caused from outdoor particles due to the high prevalence of Gram-negative cocci. The second most prevalent bacterial phenotype was the Gram-positive, catalase positive cocci, appearing indoors in all the three studied gymnasiums. Several studies indicated that this phenotype is the most prevalent morphological type indoors (Nunes et al., 2013; Bouillard et al., 2005; di Giulio et al., 2010; Aydogdu et al., 2010). This phenotype includes species such as *Staphylococcus* and *Micrococcus*, which are abundant on human skin and on mucous membranes (Aydogdu et al., 2010;

Fox et al., 2005). Our results were similar with those found by Bouillard et al. (2005) in healthy office buildings once Gram-positive catalase negative cocci were not identified. G9 presented the highest morphological diversity when compared the others fitness centers. As bacteria are strongly linked with levels of human occupancy, this result can be related to the higher occupancy of G9 and the need of more effective sanitation. This difference is/can be attributed to the higher levels of human occupancy within this gymnasium, as there is a strong correlation between human occupancy and bacterial diversity, revealing the need for more effective sanitation.

2.2.7 Conclusions

The indoor microflora is a complex mixture that varies according to the activities being undertaken, human occupancy levels, ventilation systems and physical parameters such as temperature and humidity. This work studied the microbiological load present in three fitness centers in the city of Lisbon, with results showing the existence of critical situations due to the presence of dangerous and toxic fungal species indoors. It was found that natural ventilation used in G10 had an influence on indoor fungal concentrations as no physical barrier exists to filter the outdoor air. For bacteria, nonconformities were recorded in G10 and G11. An increase of indoor bacterial concentration was observed during the evening that was not observed for fungal concentrations, thereby demonstrating the effect of human occupancy on bacterial load.

2.3 Estimating the inhaled dose of pollutants during indoor physical activity

Based on the article:
Estimating the inhaled dose of pollutants during indoor physical activity
C.A. Ramos, J.F. Reis, T. Almeida, F. Alves, H.T. Wolterbeek, S.M. Almeida
Science of the Total Environment, 527–528:111–118

2.3.1 Abstract

It is undeniable that many benefits come from physical activity. People exercise in fitness centers to improve their health and well-being, prevent disease and to increase physical attractiveness. However, these facilities join conditions that cause poor indoor air quality. Moreover, increased inhalation rates during exercise have influence on inhaled doses of air pollution. This chapter aims to calculate the inhaled dose of air pollutants during exercise, by estimating minute ventilation of participants and measuring air pollutant concentrations in fitness centers. Firstly, the 20 participants performed an incremental test on a treadmill, where heart rate and minute ventilation were measured simultaneously to develop individual exponential regression equations. Secondly, heart rate was measured during fitness classes and minute ventilation was estimated based on the calculated regression coefficients. Finally, the inhaled dose of air pollutants was calculated using the estimated minute ventilation and the concentrations of the pollutants measured in a monitoring program performed in 63 fitness classes. Estimated inhaled doses were higher in aerobic classes than in holistic classes. The main difference was registered for PM10 inhaled dose that presented an average ratio between aerobic and holistic classes greater than four. Minute ventilation and PM10 concentrations in aerobic classes were, on average, 2.0 times higher than in holistic classes. Results showed that inhalation of pollutants are increased during heavy exercise, demonstrating the need to maintain high indoor air quality in fitness centers. This chapter illustrates the importance of inclusion minute ventilation data when comparing inhaled doses of air pollution between different population groups. This work has estimated for the first time the minute ventilation for different fitness classes. Also constitutes an important contribution for the assessment of inhaled dose in future studies to be performed in fitness centers.

2.3.2 Introduction

Approximately 3.2 million deaths each year are attributable to insufficient physical activity and it is the fourth leading risk factor for death worldwide (WHO, 2014c). The benefits that come from physical activity are indubitable (Warburton et al., 2006) and contribute to improve people's health, reducing cardiovascular diseases (Myers, 2003; Patel et al., 2013) and diabetes (Brown, et al., 2014; Weisser, 2014), preventing several types of cancer and recovering from it (Foucaut, 2014; Keimling et al., 2014; Behrens et al., 2014; Gotte et al., 2013; Gonçalves et al., 2014; Buffart et al., 2014), improving musculoskeletal status and disability (Laskowski and Lexell, 2012) and finally potentiating physical attractiveness, well-being (Duda et al., 2014) and social experiences (Pila et al., 2014). The rates of physical activity are different across countries and regions, gender, age and socioeconomic status. According to the Eurobarometer Sport and Physical Activity Report (2014), citizens in the northern part of the EU are more physically active than the southern, with the lowest levels of participation found clustered in the southern EU Member States. Men are more likely to exercise or play sports than women, with the amount of regular activity tending to decrease with age.

The practice of sport in fitness centers in 2009 registered a slight increase in the number of memberships of health or fitness centers (Eurobarometer, 2014). An IAQ monitoring program performed in eleven fitness centers from Lisbon, Portugal, indicated concerning levels of VOC, CH_2O, CO_2 and PM (Ramos et al., 2014). Moreover, toxigenic fungal species were found present within the same fitness centers (Ramos et al., 2015a). PM has also been identified as a concerning pollutant in fitness centers by Braniš and Safránek (2011a) and Buonanno et al. (2013).

Data on IAQ in fitness centers demonstrate the importance of studying exposure to pollutants during physical activity in order to minimize adverse health effects and potentiate the benefits of physical activity. Only few have taken into account that sport practitioners have an increased VĖ compared to elders, office workers or children influencing their inhaled dose of air pollutants (Almeida-Silva et al., 2015). For increased health benefits, adults should practice moderate-intensity aerobic physical activity to 300 minutes/week (WHO, 2010). This represents 5 hours/week and with this information, the time spent during exercise reveals great importance to the relative daily dose, due to the increased VĖ in this activity than in others that take more time (e.g. sleep) (Dons et al. 2011).

The study of the interaction between person and pollutant involves several steps. The inhaled dose is one of the principal steps in the chain of events since dose received by an individual directly influences the impacts on health. This work joined environmental researchers and exercise physiologists to assess the inhaled dose of pollutants during fitness center's classes.

2.3.3 Methodology

The inhalation dose of pollutants during the fitness classes was estimated by using the methodology described in figure 2.14.

2.3.3.1 Determination of VĖ During Fitness Classes

Since VĖ has never been measured before for fitness class users, this work estimated VĖ for aerobic and holistic classes that represent the majority of the programs offered in fitness centers. VĖ is difficult to measure in field studies due to some constraints such as discomfort for the user and the need for an elevated number of instruments to perform the evaluation of a representative number of individuals. However, VĖ can be estimated by measuring heart rate (HR) in fitness classes because HR is easily measured and is a good predictor of VĖ (Mermier et al., 1993; Zuurbier et al., 2009). Once HR is mainly influenced by oxygen consumption and the correlation between oxygen consumption and VĖ is high, HR and VĖ are expected to be strongly associated.

A questionnaire was applied to the participants about their physical status and healthy behaviours (smoking status, hours of physical activity per week, cardiac and respiratory diseases, orthopedic problems). On test days, the subjects were instructed to report to the laboratory or the fitness center in a rested state, having completed no strenuous exercise or consumed alcohol within the previous 24h, and having abstained from food and caffeine for the preceding 3h.

2.3.3.1.1 Studied population

Ten men and ten women participated in this study and signed a free and informed agreement.

Table 2.13 presents the descriptive statistics of the studied population. The age of the volunteers varied between 18 and 38 years old, representative of principal users of fitness centers. All the subjects were physically active, but not involved in an exercise training program at the time of data collection. The weekly exercise volume is as stated in table 2.13. The average body mass index (BMI) and weight are similar to the average Portuguese BMI.

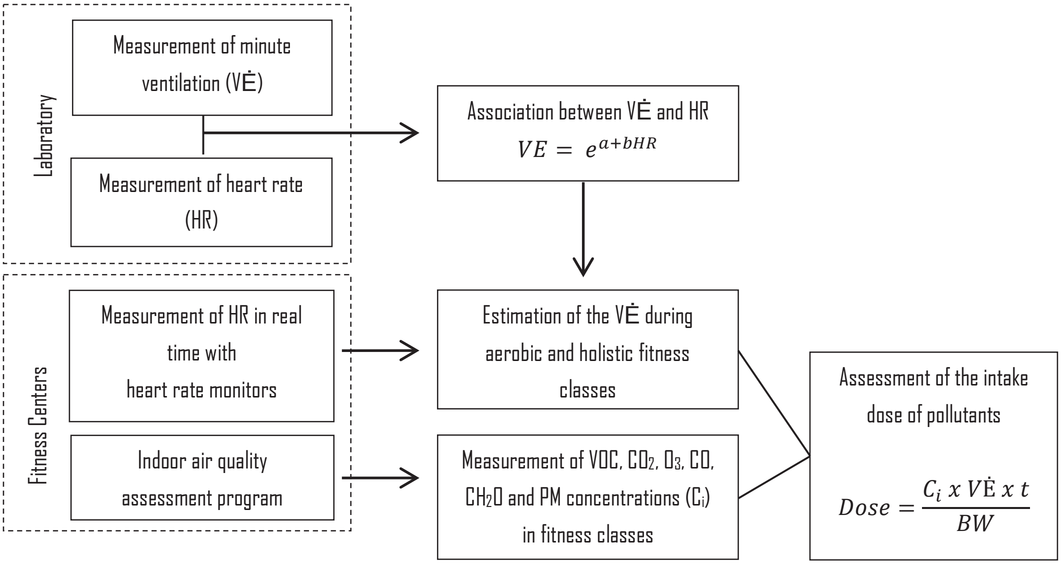


Figure 2.14 – Diagram of the methodology (BW - body mass in kg; t - duration of the fitness class)

Table 2.13 – Descriptive statistics of the participants

	Men (n=10)	Women (n=10)
Age (years) $\bar{x}$ (range)	24 (18-29)	28 (23-38)
Physical Activity (hr/week)	<1h – 20% 1h – 3h – 20% 3h – 6h – 40% >6h – 20%	<1h – 20% 1h – 3h – 40% 3h – 6h – 20% >6h – 20%
BMI (kg/m^2) $\bar{x}$ (range)	24 (18 – 27)	22 (20 – 27)
Weight (kg) $\bar{x}$ (range)	73 (48 – 85)	61 (59 – 75)

2.3.3.1.2 Estimation of the association between VĖ and HR in laboratory

VĖ levels of the participants were estimated using HR measurements, because direct measurements of VĖ are not recommended as it would influence VĖ. Therefore, participants performed an incremental test on the treadmill in order to establish the individual relation between HR and VĖ during the exercise. After a warm-up period of 3 minutes at 5km/h, the subjects performed an incremental test with 1km/h increments each minute, until exhaustion. Throughout the test, the respiratory and pulmonary gas-exchange variables were measured using a breath-by-breath portable gas analyzer (Cortex Metamax 3B, Leipzig, Germany). Before each test, the O_2 and CO_2 analysis systems were calibrated in accordance with manufacturer's guidelines against known concentrations of cylinder gases (15% oxygen, 5% carbon dioxide) while the turbine flowmeter was calibrated using a 3-l syringe. HR was also monitored throughout the tests (Polar, Kempele, Finland). Breath-by-breath VO_2 and HR data were averaged using 10sec bins. After the simultaneous measurement of $\dot{VE}$ and HR in the laboratory, exponential regression equations between these parameters were calculated. The effect of gender, age and BMI on individual slopes and intercepts was analyzed.

2.3.3.1.3 Estimation of the $\dot{VE}$ for the holistic and aerobic fitness classes

The participants performed, in group, two fitness classes – a holistic class and an aerobic class – of 45 minutes each, guided by a professional fitness instructor. The duration and types of classes were chosen for being representative of classes usually found at any fitness center. Both were composed of 3 stages: 1) starting warm-up period, 2) main conditioning period and, 3) final period of active recovery and, for the holistic class, meditation.

The holistic class is inspired by Pilates, Yoga and Tai Chi movements where the training of flexibility and strength were the primary goals. The types of exercises practiced were related to balance, stability and flexibility with a series of stretches and strength poses that provides a quiet and harmonious state of mind. The aerobic class is characterized by jumps, fast and vigorous movements of legs and arms, and other body weight strength exercises. The participants perform cardiovascular and core strength training, providing an effort much more demanding than in the holistic class.

During the fitness classes HR was recorded with a heart rate monitor from Polar Team^2 Pro (Polar, Kempele, Finland). The transmitter in the waistband recorded the data and communicated with the Polar Team^2 Base Station (Polar, Kempele, Finland), which allowed the visualization of the data in real time. Immediately after each class, the subjects classified it with the Borg Scale Perceived Exertion Rate (RPE) from 1 (very easy) to 10 (maximal exertion) (Foster et al., 2001). The recorded HR were calculated into $\dot{VE}$ levels using the regression coefficients.

2.3.3.2 IAQ Monitoring Programme

The monitoring program was carried out during 63 fitness classes in Lisbon and the methodology is already described on chapter 2.1.3.1.

2.3.3.3 Statistical Analysis

Origin 8.0® was used to compute the individuals' regression equations. The statistical tests were performed with Statistica 12® software.

2.3.3 Results and Discussion

2.3.3.1 Estimation of the VĖ in fitness classes

Exponential regression equations between HR and VĖ were calculated for the 20 volunteers. Regression lines of all individuals discriminated by gender are presented in figure 2.15. In table 2.14 are presented the distribution of the regression coefficients for men, women and for the total group. The correlation between HR and VĖ was high (mean r^2=0.90) suggesting, as expected, that HR is a good predictor of VĖ.

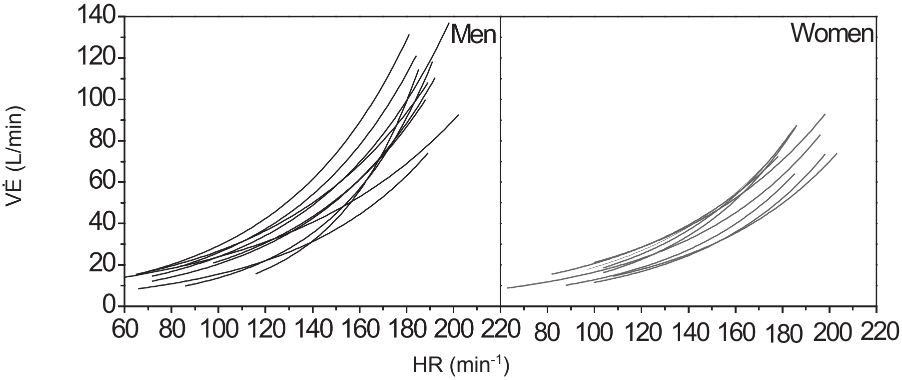


Figure 2.15 – Individual fitted regression lines of HR (beats per minute) and VĖ (litre per minute) discriminated by men and women.

Zuurbier et al. (2009) stated that the use of a general equation results in a good prediction of the mean VĖ. Although the author emphasized that, if the focus of the study is at the individual level, an overall equation of the whole group may result in substantial differences in estimated VĖ at the individual level. In the same way, Mermier et al. (1993) indicated that it is necessary to calculate individual regression equations due to the individual variability among HR and VĖ. Results showed small differences in slopes and intercepts between individuals of the same gender and lower intercept in women than in men.

During the fitness classes, HR of the volunteers was measured in order to estimate the VĖ using the individual relation between HR and VĖ derived from the laboratory test. Figure 2.16 presents the HR in the fitness classes. In both classes men presented lower HR (98 ± 17 min^{-1} in the holistic and 120 ± 17 min^{-1} in the aerobic) than women (106 ± 20 min^{-1} in the holistic and 146 ± 21 min^{-1} in the aerobic). Table 2.14 presents the HR measured in both fitness classes and the estimated VĖ. The results are displayed by genders and for each individual.

In the holistic class that involves stages of localized force, stability, balance and breath control an average VĖ of 27 ± 8 L/min for men and 25 ± 10 L/min for women were estimated. In the aerobic class, characterized by intense cardio and aerobic movements, the average VĖ was 37 ± 12 L/min for men and 57 ± 20 L/min for women. The Mann Whitney test showed no statistical differences between genders for the VĖ (*p*=1, CI=95%). Significant differences were found between the holistic and the aerobic fitness classes for women (Wilcoxon matched pairs test, *p*=0.0005, CI=95%). RPE is a subjective way of measuring physical activity intensity level, based on the personal experiences on physical sensations during physical activity, including increased HR, breathing rate, sweating and muscle fatigue (CDC, 2011). Men evaluated the holistic class as moderate level (2.9) while women considerate it as somewhat hard (4.5). Both genders classified the aerobic class as hard (5.4 for men and 6.0 for women).

Until now VĖ has never been measured for different fitness class users. The measurement of VĖ during fitness activity has two principal limitations: 1) the number of volunteers that have to use uncomfortable equipment/mask during a fitness class and 2) the influence that the use of this mask can have on the VĖ. Therefore, on large studies, it could be efficient to use an established relation between HR and VĖ for a sample of individuals and to apply the regression coefficients in field studies where HR can be easily measured by a heart rate monitor.

Table 2.14 – Collected data and results of the estimation of minute ventilation

		Treadmill Protocol		Fitted exponential equation			Measured fitness class		Estimated for fitness class		Perceived Exertion Rate	
							Holistic	Aerobic	Holistic	Aerobic	Holistic	Aerobic
	Individuals	HR (min^{-1})	VE (L/min)	r^2	Intercept – a	Slope – b	HR (min^{-1})	HR (min^{-1})	VĖ (L/min)	VĖ (L/min)		
Men	a	152 ± 33	60 ± 37	0.97	0.26 ± 0.14	$0.02 \pm 7.5E^{-4}$	91 ± 18	116 ± 20	8.5 ± 3.0	14 ± 5.8	3	6
	b	152 ± 21	53 ± 31	0.97	-0.56 ± 0.13	$0.03 \pm 7.9E^{-4}$	88 ± 16	105 ± 21	8.7 ± 3.5	16 ± 11	5	8
	c	146 ± 30	55 ± 28	0.96	1.34 ± 0.09	$0.02 \pm 5.2E^{-4}$	110 ± 16	141 ± 17	37 ± 11	68 ± 21	4	4
	d	133 ± 47	44 ± 27	0.93	1.84 ± 0.09	$0.01 \pm 5.0E^{-4}$	107 ± 20	136± 18	19 ± 3.6	25 ± 4.5	2	5
	e	134 ± 35	58 ± 34	0.96	1.42 ± 0.09	$0.02 \pm 5.4E^{-4}$	121 ± 19	163 ± 19	50 ± 18	-	3	5
	f	138 ± 38	52 ± 33	0.97	1.82 ± 0.08	$0.02 \pm 4.5E^{-4}$	92 ± 13	111 ± 13	40 ± 9.5	59 ± 15	4	8
	g	132 ± 41	53 ± 33	0.89	1.71 ± 0.14	$0.02 \pm 8.2\ E^{-4}$	77 ± 14	86 ± 16	26 ± 6.8	33 ± 9.8	3	6
	h	146 ± 41	68 ± 42	0.98	1.39 ± 0.06	$0.02 \pm 3.8E^{-4}$	96 ± 18	131 ± 18	29 ± 9.2	59 ± 22	2	7
	i	139 ± 37	37 ± 23	0.86	0.98 ± 0.19	$0.02 \pm 1E^{-3}$	101 ± 19	124 ± 19	21 ± 7.6	34 ± 13	2	4
	j	128 ± 36	65 ± 43	0.65	1.53 ± 0.32	$0.02 \pm 2E^{-3}$	96 ± 14	87 ± 6.9	33 ± 8.5	26 ± 3.6	1	1
	Group	**140 ± 36**	**55 ± 33**	**0.91**	**1.17 ± 0.13**	$\mathbf{0.02 \pm 7.7E^{-4}}$	**98 ± 17**	**120 ± 17**	**27 ± 8**	**37 ± 12**	**2.9±1.2**	**5.4±2.1**

Table 2.14 (cont.) – Collected data and results of the estimation of minute ventilation

	Treadmill Protocol		Fitted exponential equation			Measured fitness class		Estimated for fitness class		Perceived Exertion Rate	
						Aerobic	Holistic	Aerobic	Holistic	Holistic	Aerobic
Individuals	HR (min^{-1})	VE (L/min)	r^2	Intercept – a	Slope – b	HR (min^{-1})	HR (min^{-1})	VĖ (L/min)	VĖ (L/min)		
A	127 ± 32	37 ± 19	0.96	1.32 ± 0.07	0.02 ± 5.11E^{-4}	82 ± 9.7	110 ± 16	20 ± 3.9	36 ± 12	4	3
B	144 ± 23	49 ± 20	0.81	1.16 ± 0.19	0.02 ± 12.5E^{-4}	106 ± 27	154 ± 21	30 ± 14	75 ± 26	3	4
C	160 ± 31	41 ± 21	0.84	0.84 ± 0.23	0.02 ± 12.4E^{-4}	120 ± 18	158 ± 19	27 ± 9.4	59 ± 21	2	5
D	141 ± 45	38 ± 25	0.96	1.12 ± 0.12	0.02 ± 6.6E^{-4}	96 ± 21	149 ± 26	23 ± 9.8	68 ± 30	4	4
E	146 ± 31	41 ± 23	0.81	1.54 ± 0.21	0.015 ± 12E^{-4}	97 ± 18	134 ± 19	21 ± 5.3	36 ± 9.8	5	8
F	146 ± 32	38 ± 17	0.77	1.23 ± 0.21	0.02 ± 11.5E^{-4}	122 ± 23	166 ± 23	25 ± 11	59 ± 23	7	8
G	156 ± 27	48 ± 26	0.96	0.90 ± 0.12	0.02 ± 7.1E^{-4}	92 ± 22	130 ± 23	18 ± 7.4	39 ± 16	2	5
H	147 ± 34	51 ± 23	0.94	1.53 ± 0.10	0.02 ± 58E^{-4}	113 ± 20	158 ± 19	-	116 ± 35	7	8
I	144 ± 23	38 ± 17	0.77	0.67 ± 0.27	0.02 ± 16.8E^{-4}	103 ± 22	133 ± 28	17 ± 7.1	32 ± 15	6	7
J	146 ± 31	33 ± 20	0.97	0.56 ± 0.1	0.02 ± 5.34E^{-4}	130 ± 22	162 ± 18	26 ± 9.9	48 ± 15	5	8
Group	**146 ± 31**	**41 ± 21**	**0.88**	**0.99 ± 0.17**	**0.02 ± 1.02E^{-3}**	**106 ± 20**	**146 ± 21**	**25 ± 10**	**57 ± 20**	**4.5±1.8**	**6±2.0**
All	143 ± 33	48 ± 27	0.9	1.09 ± 0.15	0.02 ± 8.96E^{-4}	102 ± 19	133 ± 19	26 ± 8.6	47 ± 16	3.7±1.8	5.7±2

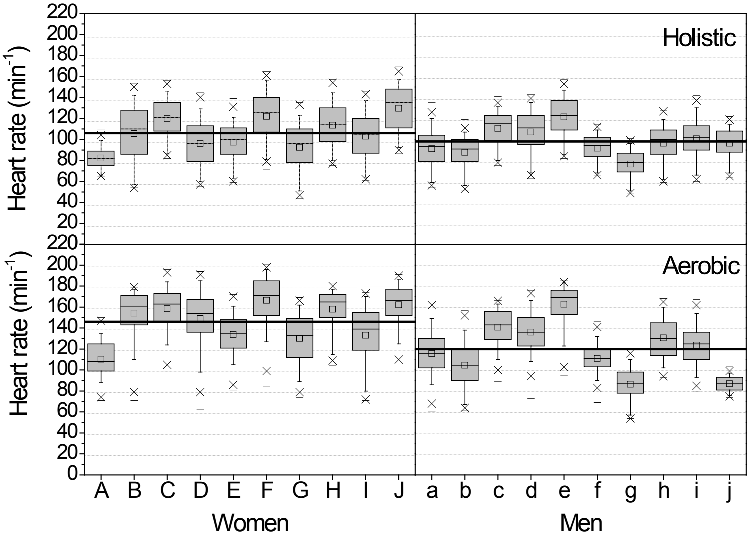


Figure 2.16 – Box plot of the heart rate of the individuals (men and women) in the fitness classes (holistic and aerobic) and average values in the groups (black line). Graphs present the minimum and maximum (-), 1st and 99th percentile (x), 25th, 50th and 75th percentile (box range), 5th and 95th percentile (box whisker) and mean (□).

2.3.3.2 Indoor air quality assessment

A total of 63 fitness classes were classified according to their typology as aerobic or holistic. The aerobic class category included all fitness classes which stimulate power, strength, vigorous and fast movements; cycling classes were excluded from this collection. Holistic classes included all classes that comprise meditation, stability and flexibility movements. IAQ was assessed in the 63 fitness classes, with pollutants' concentrations measured during the activities presented in table 2.15.

Concentrations measured in the fitness classes were compared with the Portuguese legislation, Portaria no. 353-A/2013 that defines the indoor air limit values for the pollutants PM10, PM2.5, CO_2, CO and VOC. CO never exceeded the limit value of 10 mg/m^3. For all the other pollutants, the Portuguese IAQ limit concentrations were sometimes exceeded. VOC exceeded the reference value of 0.6 mg/m^3 in both classes. This pollutant is emitted by consumer products or structures that exist in the indoor environments, such as carpeting, furniture cleaners, paints, perfumes, lacquers and solvents (Almeida-Silva et al. 2014c).

Table 2.15 – Statistical data of IAQ pollutants measured during different types of fitness classes

Pollutant	Class	n	Mean ± stdev	min	P_5	P_{25}	P_{50}	P_{75}	P_{95}	Max	IAQ Guidelines*
tVOC (mg/m^3)	Aerobic	45	0.48 ± 0.42	0.010	0.050	0.17	0.46	0.66	1.1	2.5	0.6
	Holistic	5	0.38 ± 0.23	0.16	0.17	0.18	0.31	0.37	0.85	0.85	
CH_2O (mg/m^3)	Aerobic	13	0.07 ± 0.06	0.010	0.010	0.038	0.050	0.075	0.19	0.25	0.1
	Holistic	0	-	-	-	-	-	-	-	-	
CO_2 (mg/m^3)	Aerobic	45	1682 ± 599	208	984	1327	1582	1815	2846	5964	2250
	Holistic	5	1662 ± 376	831	986	1522	1738	1878	2173	2406	
O_3 (mg/m^3)	Aerobic	13	0.01 ± 0.054	0.005	0.005	0.005	0.005	0.005	0.020	1.9	-
	Holistic	0	-	-	-	-	-	-	-	-	
CO (mg/m^3)	Aerobic	41	0.64 ± 0.49	0.005	0.12	0.30	0.57	0.83	1.6	2.7	10
	Holistic	5	0.29 ± 0.12	0.12	0.12	0.23	0.23	0.34	0.6	0.69	
PM0.5 (µg/m^3)	Aerobic	45	2.9 ± 1.7	0.52	0.76	1.6	2.6	4.02	6.2	9.3	-
	Holistic	5	2.4 ± 0.86	1.6	1.7	1.9	1.9	2.4	4.3	4.4	
PM1 (µg/m^3)	Aerobic	45	4.6 ± 2.5	0.700	1.5	2.9	4.3	5.7	9.8	15	-
	Holistic	5	3.6 ± 1.3	2.9	2.8	2.9	3.03	3.8	6.5	6.9	
PM2.5 (µg/m^3)	Aerobic	45	8.3 ± 4.5	0.89	2.5	5.4	7.3	10	16	34	25
	Holistic	5	5.5 ± 2.2	3.5	3.9	4.1	4.4	5.6	11	12	
PM5 (µg/m^3)	Aerobic	45	18 ± 10	1.4	4.9	10	15	23	38	89	-
	Holistic	5	10 ± 6.3	4.2	5	5.9	7	9.2	26	30	
PM10 (µg/m^3)	Aerobic	45	31± 23	1.8	7.9	16	24	40	76	153	50
	Holistic	5	14 ± 8.9	4.2	6.2	7.9	10	15	36	42	

*Portaria no. 353-A/2013

For PM10, PM2.5 and CO_2 the maximum value exceeds the guidelines only in the aerobic classes. These higher concentrations during aerobic classes were expected since CO_2 concentration is a function of the site occupation, ventilation rates and metabolic activity of the occupants (Ramos et al., 2014; Pegas et al., 2011b) and particles are highly dependent on the activity pattern of the room that can promote resuspension of particles (Canha et al., 2010, 2012; Braniš and Safránek, 2011a).

CH_2O and O_3 were only measured in aerobic classes due to failing in the equipment. In the aerobic classes, CH_2O showed the maximum value (0.25 mg/m^3) which is above the guideline. CH_2O is a VOC, but given its importance due to the related health effects, is usually assessed in an individualized form. However, its indoor sources are also similar to the sources of VOC. O_3 is not regulated by the Portuguese legislation.

IAQ also varies according to the characteristics of each fitness center such as ventilation system, occupancy, maintenance, age, etc. However, even in the same fitness classroom the pollutant concentrations can also vary according to the type of activity that is performed inside it. Figure 2.17 shows that PM10 concentrations measured in the same classroom and on the same day were higher during the aerobic class (average 45 µg/m^3) than in the holistic class (average 33 µg/m^3) due to the greater resuspension caused by the first activity. Ramos et al. (2014) found that in studios, higher PM concentrations coincided with the period of fitness classes, revealing a relation between PM concentration and the resuspension of dust caused by the practitioners of physical activity. The same behaviour can be observed for CO_2, found to be highly produced in an aerobic classes (Ramos et al., 2014).

2.3.3.3 Estimation of inhaled dose

The inhaled dose was calculated by using the VĖ estimated for men and women (table 2.14) and the air pollutants' concentrations measured during the fitness classes (table 2.15). Table 2.16 presents the estimated inhaled dose discriminated by gender and type of activity considering the minimum, mean and the maximum pollutant concentrations measured during the classes.

Higher inhalation doses were determined for the aerobic classes as a consequence of the high VĖ associated with this activity. For men, the average aerobic/holistic ratio of the inhaled dose was 1.4 (maximum ratios calculated for PM10 and CO with values equal to 4.0 and 3.9, respectively) while for women the average aerobic/holistic ratio was 1.8 (maximum ratios calculated for PM10 and CO with the values 5.1 and 5.0, respectively). The average ratio (women/men) of 1.5 was obtained for inhaled doses, meaning that women inhales 1.5 times higher doses than men. These results points to the importance of studying the specific VĖ associated with each activity and gender in the calculation of the inhaled dose.

Inhalation of air pollutants is influenced by VĖ, but deposition in lungs of air pollutants is also influenced by the amount of nasal and oral breathing and by the depth of inhalation (Zuurbier et al., 2009). More oral breathing and deeper inhalation occur during exercise in fitness classes, both leading to higher deposition of pollutants in lungs. In the present study, we have not been able to measure oral and nasal breathing separately, nor have we measured the depth of inhalation.

Studies evaluating the inhaled dose of pollutants during exercise and physical activity are very scarce. As far as we know, this parameter has only been assessed for cyclists during commuting. For cyclists, Nyhan et al. (2014) calculate inhaled doses of 0.95 µg/kg for PM2.5 and 1.2 µg/kg for PM10. Panis et al. (2010) calculated average inhaled doses of 0.93 µg/kg for PM2.5 and 2.1 µg/kg for PM10 (values adapted to the body weight of the present study and for 45 minutes in order to allow comparisons). These values are comparable with the dose estimated in our study for aerobic (0.30 µg/kg for PM2.5 and 1.2 µg/kg for PM10).

Table 2.4 – Estimated inhaled dose of pollutants in holistic and aerobic class

	Men			Women		
	min (µg/kg)	Mean (µg/kg)	Max (µg/kg)	min (µg/kg)	Mean (µg/kg)	Max (µg/kg)
			Holistic			
tVOC	2.7	6.3	14	3.1	7.2	16
CO_2	13838	27670	40046	15947	31887	46149
O_3	0.00	-	0.00	0.00	-	0.00
CO	1.9	4.8	11	2.2	5.6	13
CH_2O	0.000	-	0.000	0.000	-	0.000
PM0.5	0.027	0.040	0.074	0.031	0.046	0.085
PM1.0	0.043	0.062	0.12	0.050	0.071	0.13
PM2.5	0.057	0.091	0.202	0.066	0.105	0.23
PM5.0	0.070	0.16	0.49	0.081	0.18	0.57
PM10	0.070	0.23	0.70	0.081	0.26	0.81
			Aerobic			
tVOC	0.29	14	74	0.44	21	108
CO_2	6166	49787	176469	9070	73235	259580
O_3	0.304	1.6	-	0.45	2.4	-
CO	0.00	19	80	0.000	28	117.516
CH_2O	0.29	2.0	7.4	0.44	2.975	11
PM0.5	0.015	0.086	0.28	0.023	0.13	0.41
PM1.0	0.021	0.14	0.45	0.030	0.20	0.66
PM2.5	0.026	0.24	1.0	0.039	0.36	1.5
PM5.0	0.043	0.53	2.6	0.063	0.77	3.9
PM10	0.052	0.92	4.5	0.077	1.4	6.7

The incremental test on the treadmill showed that HR and VĖ are highly correlated as has also been reported before (Zuurbier et al., 2009). HR is not only influenced by exercise, but also by coffee, drugs, time of the day and temperature. These factors probably did not play an important role in this study. Differences in regression equations were observed between genders. According to Harms (2006), the reproductive hormones estrogen and progesterone can influence ventilation and pulmonary function during exercise. This author suggests that during heavy exercise, women demonstrate greater expiratory flow limitation and an increased effort to breath. Figure 2.17 shows that VĖ varied during the classes and between classes. In aerobic classes VĖ levels (55 L/min) were on average 2.1 times higher than in holistic classes (26 L/min).

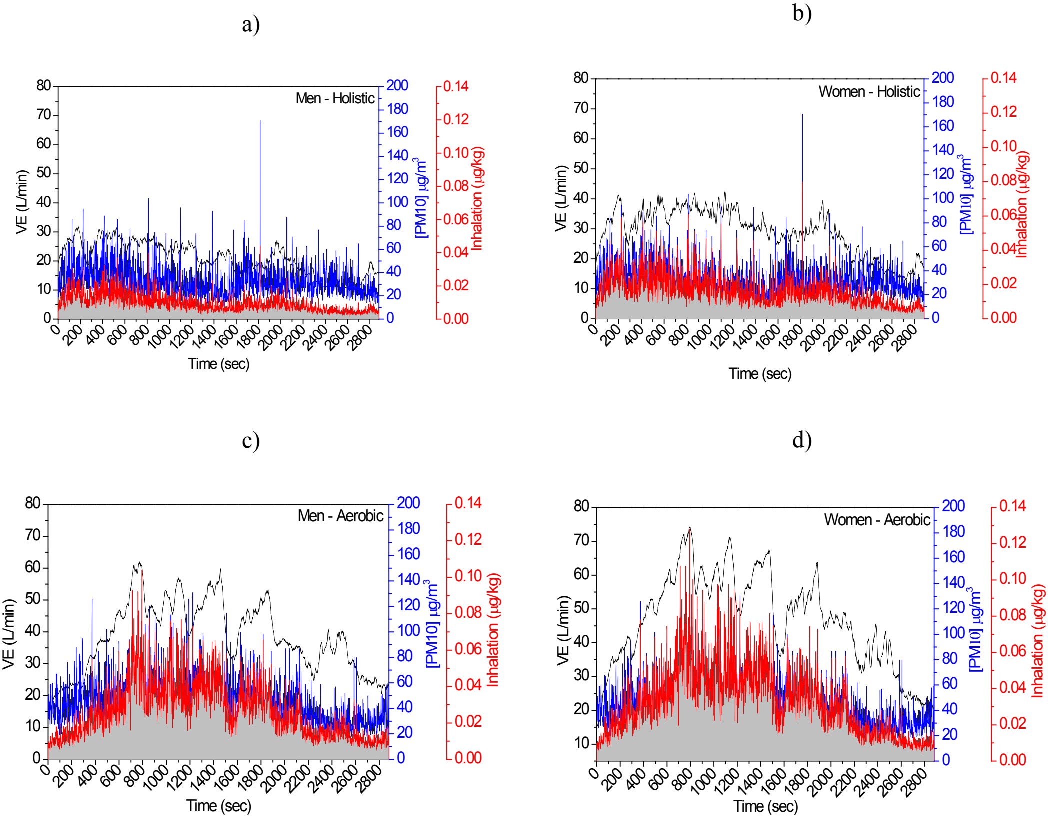


Figure 2.17 – VĖ and PM10 concentration and inhaled dose during holistic (a and b) and aerobic (c and d) fitness classes, for men and women. The grey area represents the total inhaled dose of PM10.

Besides the inhaled dose, it is important to consider the uptake fraction of pollutants. The solubility of the gaseous pollutants determines the absorption and the diffusion rates and particle size influence deposition fraction in the lungs (Bigazzi and Figliozzi, 2014). Smaller particles enhanced the deposition and hygroscopic aerosols will absorb water vapor from the lungs, thus growing in size and consequently changing their deposition properties (Winkler-Heil et al., 2014). Some of the analyzed pollutants have chemical characteristics that pose some concerns, individually or in synergistic actions. In a study conducted by Mautz (2003) it was found that O_3 is capable to boost CH_2O damage, therefore the inhaled estimated dose in this study for these pollutants was low (2 µg/kg), it is important to take into account the combined effects. CH_2O is a highly soluble gas that may be absorbed in the upper respiratory tract at resting VĖ values and O_3 is an insoluble gas, which penetrate deep into the lungs with the main cellular target the terminal bronchioles and proximal alveoli. Its uptake is amplified for higher tidal volumes (volume of air in the lungs in one breath, L) than in breathing frequency (number of breaths per minute, breaths/min) (Mautz, 2003), thus even differences in the respiratory pattern cause different uptake dose.

CO_2 was the gas that presented the higher inhaled dose but this pollutant is a human product of respiration. Studies showed that inhalation of high concentrations of CO_2 (50000 mg/m^3) during physical activity has no effect on psychomotor performance (Vercruyssen et al., 2007).

2.3.4 Conclusions

Assessing the inhaled dose of air pollutants in fitness centers is the key determinant of the impacts that these pollutants can have on the health of the sport practitioners. Differences between $\dot{VE}$ and pollutant concentrations between different classrooms influence the inhaled dose of air pollution and therefore an accurate assessment of these two parameters is fundamental.

Study the dose is also a complex challenge. The inhaled dose is affected by several physiological and chemical mechanisms. Also the differences in the definitions used among the researchers lead to complex and tricky comparisons between the studies. Knowing that trained or recreational athletes are at special risk when they are practicing exercise in polluted environments, this study assessed by the first time air pollutants' inhaled dose by users of fitness centers during the physical activity. $\dot{VE}$ for different fitness classes were estimated based on the measurement of HR. Results generated within the present work can be used to calculate inhaled dose in future studies with a similar population.

From the results can be concluded that the magnitude of differences in pollutant concentrations depend not only on the fitness center, but also on the type of activity that occur inside the classrooms. During aerobic classes the concentrations of particles and CO_2 were higher than in holistic classes. Also the $\dot{VE}$ is higher during aerobic classes and consequently inhaled dose is on average 2.1 times higher in this activity compared with the holistic classes. This study illustrates the importance of inclusion $\dot{VE}$ data in comparing inhaled doses for different population groups. Sport activity in fitness centers has undeniable positive effects on the health of the sport practitioners. The results presented in this chapter should be seen as an opportunity to improve environmental conditions in gymnasiums in order to potentiate their positive impacts.

3 Cycling in Urban Areas

3.1 Exposure Assessment of a Cyclist to Particles and Chemical Elements

Based on the article:
Exposure assessment of a cyclist to particles and chemical elements
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submitted to *Environmental Science and Pollution Research*

3.1.1 Abstract

Cycle paths can be used as a route for active transportation or simply to cycle for physical activity and leisure. However, exposure to air pollutants can be enhanced while cycling, in urban environments, due to the proximity to vehicular emissions and elevated breathing rates. The objective of this work was to assess the exposure of a cyclist to particles and to chemical elements by combining direct reading equipment and biomonitoring techniques. PM10 and PM2.5 were measured during 60 travels, performed on three cycle paths located in Lisbon throughout weekdays and weekends and during rush hours and off-peak hours. Lichens were exposed along cycle paths for 3 months and their element contents measured by Instrumental Neutron Activation Analysis using the k0 methodology (k0-INAA). Results of this study indicated that using a bicycle commute route of lower traffic intensity and avoiding rush hours or other times with elevated vehicular congestion facilitate a significant reduction in exposure to pollutants. The implementation of cycle paths in cities is important to stimulate physical activity and active transportation; however it is essential to consider ambient air and pollutant sources to create safer infrastructures.

3.1.2. Introduction

The increased mobility of people with recurrent use of private cars, even for short distances, transformed road traffic to being a major contributor to the poor air quality in urban areas. In cities, traffic-generated emissions were estimated to account for more than 50% of the total emissions of PM

(Han and Naeher 2006) that are of central importance for atmospheric chemistry and physics, biosphere, lithosphere, hydrosphere, climate and public health.

Evidence linking PM exposure to adverse human health impacts, especially associated with respiratory and cardiovascular mortality and morbidity, is well reported in literature (Nyhan et al. 2013; Almeida et al. 2014a). In 2013 the International Agency for Research on Cancer (IARC) from the World Health Organization (WHO) declared PM as a human carcinogenic from group 1 (IARC 2013).

PM effects have been seen at very low levels of exposure and there is no evidence of a safe level of exposure or a threshold below which no adverse health effects occur. This is due to the fact that PM is a complex mixture of microscopic particles enriched with different chemicals, including heavy metals, derived from both anthropogenic and natural sources. From a mechanistic perspective, it is highly plausible that the chemical composition of PM would better predict health effects than other characteristics such as PM mass or size. This is consistent with the large number of laboratory studies that demonstrated compositional variability in PM toxicity and epidemiological studies that portray the regional heterogeneity in PM-related health effects (Zanobetti et al., 2009; Bell et al., 2008).

There is thus a new paradigm in society: the sustainable mobility and the demand for new transport policies that can be the alternative to individual transport. In southern European countries, which presents the lowest physical activity rates (Eurobarometer 2014), cycling can represent an important alternative, not only to encourage healthy behaviors and promote physical activity, but also to promote active transportation and reduce pollutant emissions. However, besides policies that increase active travel which are likely to generate large individual health benefits, depending on the conditions of policy implementation, risk tradeoffs are possible for some individuals, who shift to active travel, by increasing inhalation of air pollutants and exposure to traffic injuries. Cycle lanes and paths have been built to promote active transportation and physical activity and to bring some order and safety for cyclists and car users. However, route selection is very important to decrease cyclist exposure to air pollutants. While PM inhalation may affect those with pre-existing condition, the healthy population is not immune to the effects of PM inhalation, especially during exercise in active transportation. This population is susceptible to pulmonary inflammation, decreased lung function, increased risk of asthma, vascular endothelial dysfunction, mild elevations in pulmonary artery pressure and diminished exercise performance (Cutrufello et al. 2012). Car drivers in urban environments are exposed to higher pollutant levels than cyclists, however cyclists are especially at risk (Carlisle and Sharp 2001) resulting in enhanced inhaled doses for cyclists (Rank et al. 2001).

The assessment of human exposure to particles concentration and chemical composition in cycle routes requires the combination of different methods. Instrumental techniques are commonly used to measure atmospheric particles concentration; however, the instrumental monitoring methodology performed with stationary sampling stations is limited to a few number of sampling equipment and do not represent the human exposure while cycling.

Portable equipment to measure PM levels combined with GPS data is thus essential to assess cyclers' exposure to particles concentrations. Several works assessed the exposure of cyclists to PM concentrations using this methodology (Boogaard et al. 2009; Berghmans et al. 2009; Int Panis et al. 2010; Cole-Hunter et al. 2012; Elen et al. 2013). However, as far as we know, no work attempts to assess the exposure to chemical elements probably due to the inherent technical difficulties. Firstly, on-line methodologies to measure the element composition of particles while cycling are not available. Secondly, the short period of a travel does not allow the sampling of enough mass of particles, by personal samplers, for subsequent chemical analysis, in order to calculate the average exposure to elements for each route.

Biomonitoring techniques can be regarded as a complementary technique to assess the distribution of chemical elements spatially (Almeida et al. 2012c). Lichens and mosses are believed to be the best biomonitors of several atmospheric pollutants, including chemical elements, gases and dioxins. This belief is rooted on two characteristics of these organisms: 1) they acquire nutrients virtually exclusively from atmospheric deposition; and 2) they have a simple physiology which makes them relatively passive accumulators (Bargagli 1998). Compared to conventional instrumental monitoring, biomonitoring offers advantages that are difficult to surpass: 1) the ability to perform high-density sampling at virtually any desired spatial scale, and 2) the ability to measure a wide range of pollutants simultaneously. This is achieved at comparatively low costs and man-power, since biomonitors are energetically self-sustainable, require no maintenance and are not attractive targets of vandalism. The successful implementation and the usefulness of atmospheric biomonitoring are reflected 1) in the large number of biomonitoring surveys performed throughout the world at international (Harmens et al. 2010), national (Freitas et al. 2000), regional (Almeida et al. 2012c; Lage et al., 2014) and indoor levels (Canha et al. 2012b, 2014), 2) its widespread use in the identification and characterization of emission sources (Marques et al. 2008) and 3) more recently its application in the realm of human epidemiology (Sarmento et al. 2008; Wolterbeek and Verburg 2004).

The aim of this work was i) to evaluate the personal exposure to PM10 and PM2.5 on three different cycle routes located in Lisbon, ii) to estimate the inhaled dose of PM during the selected paths, iii) to identify pollutant sources, iv) and to map PM2.5, PM10 and chemical elements concentrations. This work will contribute to apportion sources of pollutants that affect cyclists, to identify the best areas to build new cycling infra-structures, and to support cyclists in the selection of the safer paths and/or periods to cycle.

3.1.3 Methodology

3.1.3.1 Area of Study

The present study was developed in Lisbon, the capital city of Portugal. In Lisbon, traffic is the main source of atmospheric pollution (Almeida et al. 2009a,b). Due to the geographic position of Lisbon – on the extreme southwest of Europe – and to the dominant western wind regime, influenced by the presence of the semi-permanent Azores high-pressure and the Icelandic low-pressure systems over the North Atlantic Ocean, high levels of pollutants should be expected. The transport of maritime air mass is usually associated with cleaner air masses from the Atlantic Ocean and with better dispersion conditions of pollutants coming from the industrial areas (Almeida et al. 2013b). Nevertheless, under adverse meteorological conditions, low dispersion conditions and thermal inversions, high concentrations of air pollutants are registered. Moreover, in Lisbon, natural PM sources cause a number of PM exceedances. Prior studies in Lisbon have shown that natural mineral particulate sources such as high-dust Saharan air mass intrusions interfere with the monitoring of the incidence of anthropogenic emissions on ambient air PM levels (Almeida et al., 2008).

In the last years Lisbon increased the incentive to the use of bicycles as an active mode of transportation, or as a complement of public transportation by including changes in the Portuguese Road Code and changes in the municipality regulations for cyclists (Barreto 2013). From a total of 90km of cycling paths in Lisbon, three paths separated from vehicle traffic, each with different characteristics, were selected for this study (Figure 3.1).

- *Cidade* (11.8km) starts in Gare do Oriente and finishes in Benfica. This path goes through the city, intersecting other cycle paths, roads, city parks and green corridors;
- *Ribeirinho* (17.8km) starts in Parque das Nações and go along with Tagus river until Belém;
- *Monsanto* (5.6km) is placed inside a florestal zone.

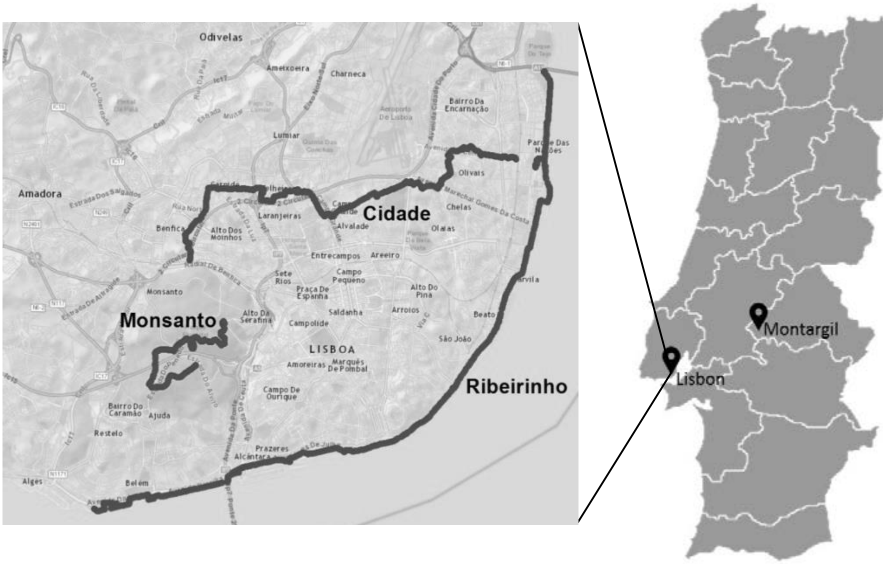


Figure 3.1 – Location of the three cycle paths in Lisbon (*Cidade, Monsanto* and *Ribeirinho*) and original location of the transplanted lichens (Montargil)

3.1.3.2. Personal Sampling

Air pollution measurements were performed during rush hours (8h – 10h) and off peak hours (11h – 13h) along the three pre-designated cycling paths in Lisbon. Each route was monitored 20 times: 10 trips were performed during weekends and 10 trips were performed during weekdays. This methodology resulted in a total of 60 sampling campaigns and lasted from May to August 2014. PM10 and PM2.5 measurements were obtained with two personal samplers SidePak AM510 (TSI, EUA). The position of the cyclist was recorded with a Garmin Etrex20 GPS (Garmin, USA). Concentrations and location were recorded each 10s. All the campaigns were performed on non-rainy days.

3.1.3.3. Biomonitoring with Lichens

Samples of the lichen *Flavoparmelia caperata* with their substrate olive bark were collected from olive trees at about 1.5m above the soil in Montargil (39◦03’24’’ N and 8◦10’36’’ W) on the 3rd April 2014. Montargil is a Portuguese rural area that presents low levels of air contamination (Almeida et al. 2012c). Before exposure, a random selection of 10 lichens was separated to serve as reference blank samples. After 4 days, a total of 53 samples with an average of 3.3g of lichen in each sample, still attached to their substrate olive bark were suspended in trees, fixed to a nylon rope, at about 1.5m above the soil, along the three cycle lanes with an interval distance of 1.5km. Lichens were exposed for 3 months, from April 7th to July 21st 2014.

3.1.3.4 Assessment of the Cell Membrane Integrity in Lichen

After exposure, lichen samples were firstly cleaned from dust, leaf debris, fungus contamination, and degraded material in the laboratory. To judge lichen vitality, exposed transplants were soaked in demineralized water, after which water conductivity was determined. The procedure followed three steps: 1) lichen material was cleaned and rinsed rapidly with demineralized water, 2–3 times for 5 seconds each; 2) after drying, approximately 1g of lichen was weighed and immersed in 100 ml demineralized water for 1h; and 3) after removal of the lichens, electric conductivity of the solution was measured with a Conductometer Metrohm 712. Blanks were made by repeating the same procedure without immersing the lichens, with their conductivity subtracted from the conductivity of the sample solution.

3.1.3.5. Element Concentrations by k_0-INAA

The determination of elemental concentrations in lichens was performed by k_0-INAA in a Portuguese Research Reactor (Freitas et al., 2000; Almeida et al., 2013a). Lichens were freeze-dried, ground in a Teflon ball mill, encapsulated, with capsules containing 150–180mg of the samples and irradiated for 5h at a thermal neutron flux of 1.03×10^{13} cm^2/s. After irradiation, two spectra were obtained with a germanium detector: 1) Samples were measured 3 days after irradiation for a period of 3.5 h; and 2) the same samples were measured again 4 weeks after the irradiation during a period of 3.5h. Irradiations allowed for determination of the elements As, Br, Ce, Co, Cr, Eu, Fe, Ga, K, La, Na, Rb, Sb, Sc, Se, Sm, U, W, and Zn.

The accuracy of the analytical method was evaluated with the certified reference material IAEA-336 which was prepared identically to the samples and co-irradiated with them (Dung et al., 2010; Almeida et al., 2014b). During the sampling campaign, 9 blank samples were treated the same way as regular samples. All measured species were homogeneously distributed; therefore, concentrations were corrected by subtracting the blank contents.

3.1.3.6. Statistical and Data Analysis

Graphical images were produced with Origin® 7.5 and ArcGIS® 10 was used to create the pollutant maps. Statistica® 12 was used to perform statistical analysis.

3.1.4 Results and Discussion

3.1.4.1 Quality Control

3.1.4.1.1 Quality control of k_0-INAA results

The quality control of k_0-INAA results were obtained by using the reference material IAEA-336 and by calculating the Zeta Score according to equation 2:

$$\zeta = \frac{x_{lab} - x_{ref}}{\sqrt{u_{lab}^2 + u_{ref}^2}} \qquad \text{(equation 2)}$$

in which x_{lab} is the mass fraction of the measured result of the element in the reference material, x_{ref} is the certified/indicative mass fraction, u_{lab} is the combined standard uncertainty of the measured result and u_{ref} is the combined standard uncertainty of the certified value. The results were interpreted according the following classes: $|\zeta| \leq 2$, considered as a satisfactory level; $2 < |\zeta| < 3$, classified as a questionable level and $|\zeta| \geq 3$, which is an unsatisfactory level (ISO 17043). Figure 3.2 indicates that the obtained results were satisfactory and did not differ significantly from the certified ones.

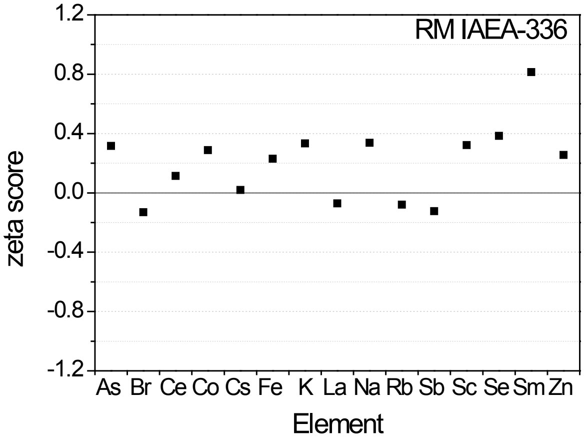


Figure 3.2 – Control chart showing the Zeta-score obtained for the certified reference material IAEA-336 analyzed by k_0-INAA.

3.1.4.1.2 Personal monitors

Prior to the sampling campaign, the SidePak AM510 worked in parallel with a Gent sampler which collects PM2.5 and PM2.5-10 samples in polycarbonate filters for gravimetric analysis (considered as reference method). The equipment worked continuously for 8h during a 5 day period. Figure 3.3 shows that the SidePak measured the highest PM2.5 concentrations (SidePak/Gent = 1.2, r^2 =0.69), which is consistent with work developed by Zhu et al. (2015), while PM10 concentrations measured by SidePak were lower than the ones measured by the Gent sampler (SidePak/Gent = 0.61, r^2 =0.78). Considering that the light-scattering properties of PM vary substantially with particle size and composition, the SidePak measurements were calibrated, based on the reference methodology, to the specific aerosol being sampled (McNamara et al. 2011; Diapouli et al. 2008).

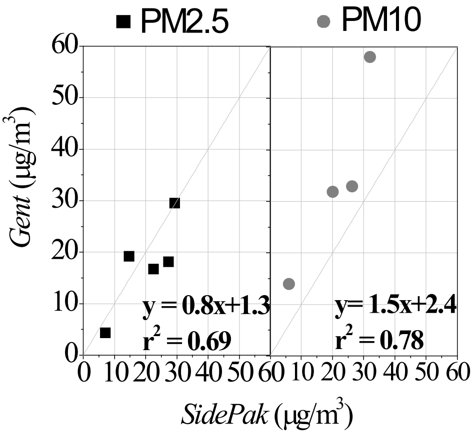


Figure 3.3 – Control chart showing the comparison between PM2.5 and PM10 measured by Side Pack and gravimetry (values in µg/m^3).

3.1.4.2. Particle Exposure and Dose

3.1.4.2.1 Exposure

Traffic-related air pollution exposure tends to be higher during travel because of the proximity to other vehicles. Exposure differences can vary considerably by the localization of the path, local traffic and period of the day.

Figure 3.4a shows that *Ribeirinho* cycle path recorded the highest PM10 mean concentration for the studied periods, both in weekdays 39.1 ± 23.5µg/m^3 at 8h and 34.8 ± 17.5 µg/m^3 at 11h) and weekends (43.4 ± 29.1 µg/m^3 at 8:00 and 40.7 ± 27.7 µg/m^3 at 11:00). Mann-Whitney test showed significantly higher concentrations at 8h and during weekends.

Cidade cycle path registered the maximum PM10 concentrations of 653.4 µg/m^3 and 635.4 µg/m^3 in very specific points, influenced by heavy traffic. However, exposure on this cycle path was lower than in *Ribeirinho*, mainly because this path crosses some urban green areas. The PM10 mean concentrations measured in this path during the weekdays was 26.7 ± 24.7 µg/m^3 at 8h and 25.4 ± 26.7 µg/m^3 at 11h and during the weekend was 18.8 ± 11.8 µg/m^3 at 8h and 18.5 ± 18.3 µg/m^3 at 11h.

Mann-Whitney test showed that concentrations were significantly higher during the weekdays and during the rush hours.

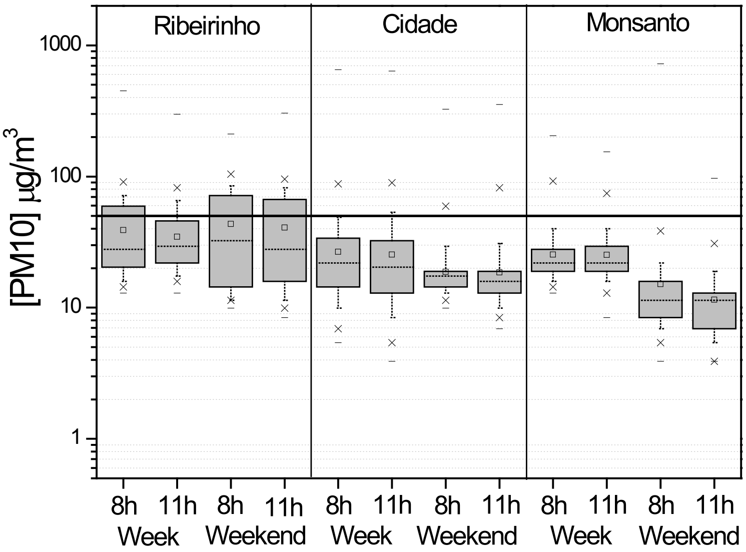
Monsanto cycle path recorded the lowest PM10 mean concentrations both in weekdays (25.4 ± 13.8 µg/m^3 at 8h and 25.2 ± 11.7 µg/m^3 at 11h) and weekends (15.1 ± 38.6 µg/m^3 at 8h and 11.5 ± 6.8 µg/m^3 at 11h). This result was expected due to the fact that the path is located in a forested area of Lisbon. Mann-Whitney test showed that concentrations were significantly higher during the weekdays and at 8h during the weekend.

Figure 3.4b shows that *Ribeirinho* path registered the highest PM2.5 concentrations, both in weekdays (11.8 ± 13.4 µg/m^3 at 8h and 10.1 ± 10.6 µg/m^3 at 11h) and especially during the weekends (19.8 ± 21.7 µg/m^3 at 8h and 18.9 ± 18.1 µg/m^3 at 11h). Significantly higher concentrations were registered during the weekends and at rush hours only during the weekdays.

The mean PM2.5 concentrations obtained in *Cidade* and *Monsanto* were 7.5 ± 11.6 µg/m^3 and 7.1 ± 13.0 µg/m^3, respectively. During weekends, the *Monsanto* cycle path registered the lowest PM2.5 concentrations among the studied days, periods and paths (6.3 ± 5.6 µg/m^3).

In Figure 3.5a and 3.5b, measured PM concentrations were coupled with the GPS position and then projected in the entire routes. The generated pollutant maps were very suitable to detect hotspots of high pollutant concentrations. The interpolation with Natural Neighbor technique was applied to obtain a continuous representation of data. The PM10 and PM2.5 classes were created according with the European Directive for ambient air quality (2008/50/CE).

a)



b)

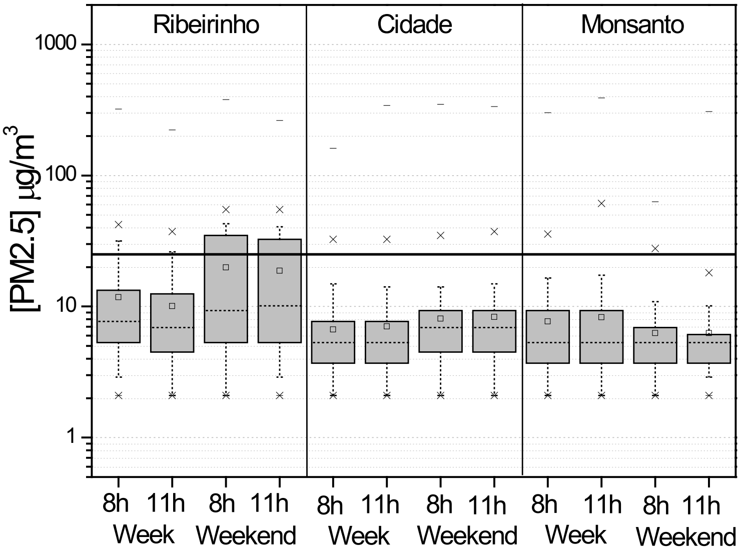
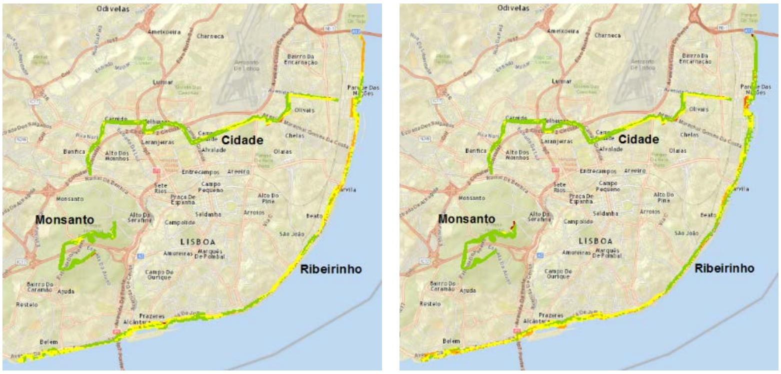


Figure 3.4 – Box-plot of PM10 and PM2.5 concentrations measured in the cycle paths (values in μg.m^{-3}). Graphs present the minimum and maximum (-), 1st and 99th percentile (x), 25th, 50th and 75th percentile (box range), 5th and 95th percentile (box whisker) and mean (□). The black line indicates the legal limit value for PM10 (50μg/m^3) for PM2.5 (25μg/m^3).

i) PM10, weekday, 8h

ii) PM10, weekend, 8h

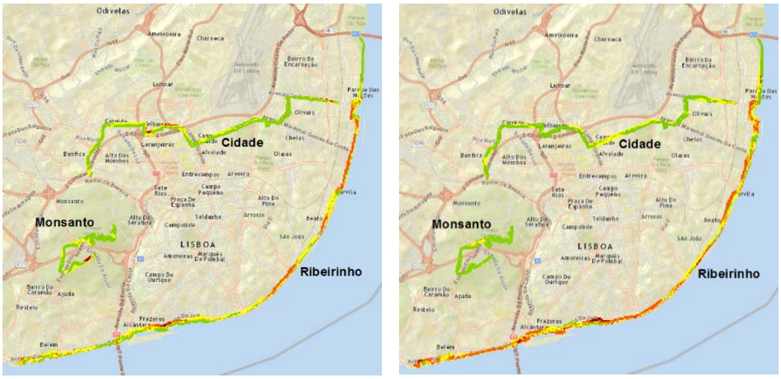


iii) PM10, weekday, 11h

iv) PM10, weekend, 11h

Figure 3.5a – Pollutant maps of PM10 concentrations along cycle paths, for weekday and weekends, at 8h and 11h.

The generated pollutant maps were very suitable to detect hotspots of high pollutant concentrations. Higher concentrations of PM2.5 and PM10 were clearly observed on the *Ribeirinho* cycle path. In these hotspots the limit value established by the Directive 2008/50/CE for PM10 and PM2.5 were exceeded. Two major contributing factors justify the highest concentrations in this cycle path principally during the weekends. Firstly, this cycle path goes side by side with a traffic road that leads to the downtown of Lisbon, which is used substantially for leisure and tourism. The circulation in this road is forbidden (in defined zones) for Euro 2 vehicles on weekdays. Thus on weekends, when the restriction is not applied, road traffic increases, leading to an increase in particle concentrations. Secondly, the Lisbon harbour can play an important role in high levels of PM presented in figure 3.5. Several harbour activities can potentiate negative environmental impacts, especially on air quality levels. Besides the direct emissions from ships, the operations of loading, unloading and transport of dusty materials in harbours contributes highly to the emission of atmospheric particulate matter (Almeida et al. 2012a). In Greece, researchers found evidence to support that port activities affect the city's air quality with PM2.5 levels observed to be higher in the port area compared to the city center (Tolis et al. 2015; Tolis et al. 2014).


i) PM2.5, weekday, 8h
ii) PM2.5, weekend, 8h

iii) PM2.5, weekday, 11h
iv) PM2.5,weekend, 11h

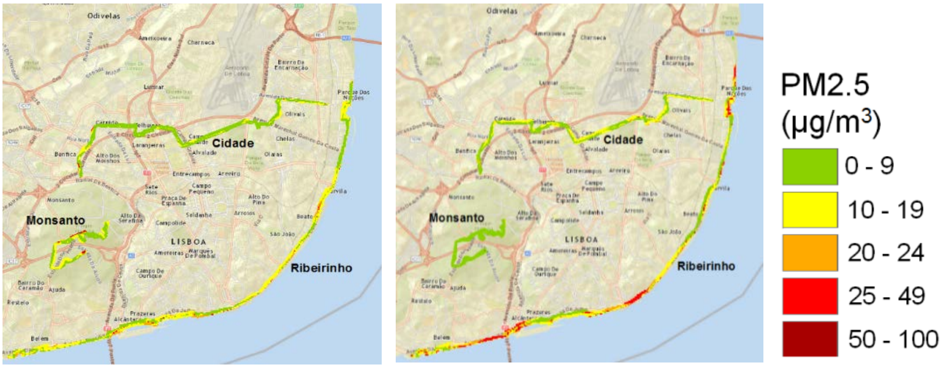


Figure 3.5b – Pollutant maps of PM2.5 concentrations along cycle paths, for weekday and weekends, at 8h and 11h.

3.1.4.2.2 Dose

Exposure occurs when an individual comes in contact with a pollutant while dose is the amount of material absorbed or deposited in the body for an interval of time (Monn 2001). Increased physical effort leads to elevated inhalation rates, thus higher inhaled doses and subsequent higher lung deposition of air pollution per unit time spent in commute. The determination of the dose goes beyond examining only exposure concentrations by including the VĖ and travel duration to compare intake dose per trip. Equation 3 was applied to estimate the average inhaled dose per km for each cycle path:

$$Dose(ug/Km) = \frac{C_i \, x \, V\dot{E} \, x \, t}{Km} \qquad \text{(equation 3)}$$

where:
Ci is the average concentration of the pollutants measured in one trip;
t is the time spent in a trip (min);
VĖ is the minute ventilation (L/min);
Km is the length of the cycle path (km).

The VĖ for cycling activity has been measured by several authors (Zuurbier et al. 2009, 2010; Int Panis et al. 2010; Cole-Hunter et al. 2012; Nyhan et al. 2014). However, due to differences on velocity, heart rate, fitness status of the individual, type of bicycle, road and terrain the results differ from work to work. The present work used the values recommended by the Environmental Protection Agency for VĖ estimated for high intensity activities (>6 MET) which is the rate considered for cycling on 'self-selected pace' (EPA, 2011b; Bigazzi and Figliozzi 2014) (table 3.1).

Table 3.1 – VĖ defined by EPA (2011b)

Age group	VĖ high intensity activities (L/min)
21-31	53.9
31-41	54.3
41-51	57.3
51-61	58.4
Mean	55.98

The mean time spent in each cycle path was calculated and 85 min for *Ribeirinho* path, 75 min for *Cidade* path and 26 min for *Monsanto* path.

Figure 3.6 presents the calculated inhaled doses for the cycle paths. *Ribeirinho* cycle path presented the highest values of inhaled doses for PM10, on weekdays (10.5 µg/km at 8h and 9.3 µg/km at 11h) and weekend (11.6 µg/m at 8h and 10.9 µg/km at 11h), as a result of higher PM concentrations and longer time cycling associated with this path. *Cidade* cycle path showed higher doses for PM10 on weekdays (9.5 µg/km at 8h and 9.0 µg/km at 11h) but higher PM2.5 doses on weekends (2.9 µg/km at 8h and 3.0 µg/km at 11h). *Monsanto* presented the lowest doses for PM10 (6.6 µg/km at 8h and 11h at weekdays; and 3.9 µg/km at 8h and 3.0 µg/km at 11h weekends) and for PM2.5 (2.0 µg/km at 8h and 2.2 µg/km at 11h at weekdays and 1.6 µg/km at 8h and 11h on weekends).

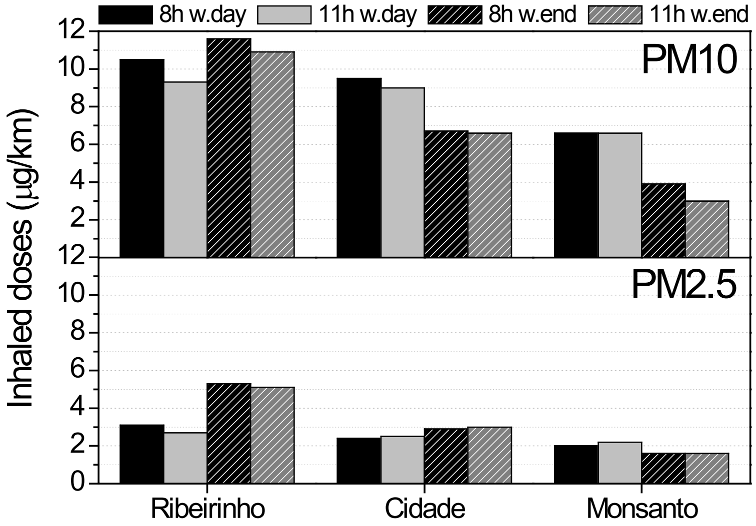


Figure 3.6 – Inhaled doses of PM2.5 and PM10 on the cycle paths for the studied periods (8h and 11h) on weekdays (w.day) and weekends (w.end).

3.1.4.3 Biomonitoring

3.1.4.3.1 Electric conductivity

Electric conductivity is an indicator of lichen vitality. This parameter is affected by environmental stressors, especially environmental pollution and meteorological conditions, to which the lichen is exposed in the place where it was transplanted. Changes in membrane permeability and the loss of electrolytes have been shown to negative correlate with the presence of gaseous pollutants in the air, such as SO_2, O_3 and NO_2 (Godinho, 2010). Figure 3.7 presents the conductivity levels measured after the exposure and the difference between the conductivity measured before and after exposure. Results show that the *Ribeirinho* cycle path was the one with highest levels of conductivity (mean 25.03 $mSm^{-1}g^{-1}$), followed by *Cidade* (mean 15.76 $mSm^{-1}g^{-1}$) and *Monsanto* that presented the lowest levels (mean 9.48 $mSm^{-1}g^{-1}$). The difference between conductivity levels before exposure and after exposure was always positive in *Ribeirinho* path, while in Monsanto the diference was negative or very low, except for lichen with reference M10 that was exposed near a traffic area located in the end of the path. This result indicates higher pollution levels in *Ribeirinho* path which is in agreement with results obtained for the instrumental measurements.

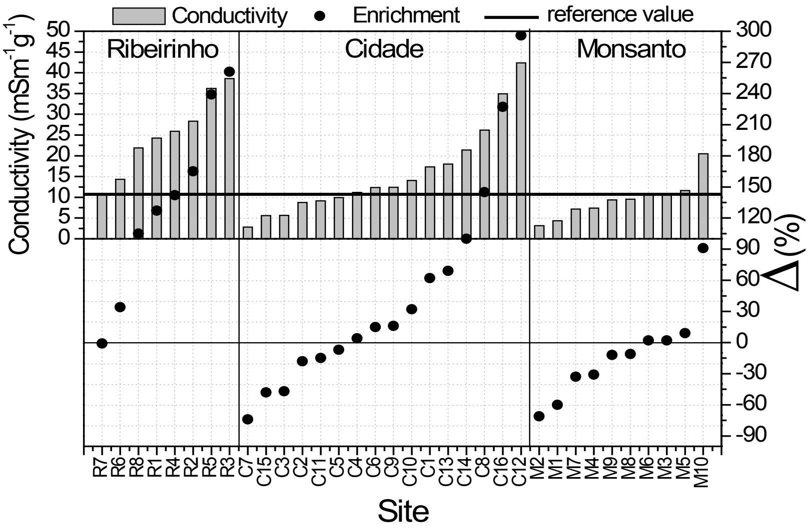


Figure 3.7 – Lichens conductivity measured after exposure (values in mSm^{-1}g^{-1}) and difference between conductivity measured before and after exposure (values in %).

3.1.4.3.1 Element mapping and sources

Pollutant sources were identified by means of principal component factor analysis (PCA) using STATISTICA® 12 software. This was performed by utilizing the orthogonal transformation method with Varimax normalized and retention of principal components whose eigenvalues were greater than unity. Factor loadings indicated the correlation of each pollutant species with each component which were also related to the source emission composition. Only the elements quantified in more than 70% of the samples were retained for PCA analysis. To evaluate the strength of the crustal and non-crustal origin of the elements, the crustal enrichment factor method has been used. EF using Sc as a crustal reference element (EF_{Sc}) were calculated based on equation 4 (Mason and Moore, 1982) and using soil composition:

$$EF_{Sc} = \frac{\left(\frac{[x]}{[Sc]}\right)_{lichen}}{\left(\frac{[x]}{[Sc]}\right)_{soil}} \qquad \text{(equation 4)}$$

Given the local variation in soil composition, $EF_{Sc} > 10$ suggests that a significant fraction of the element was contributed by noncrustal sources. Figure 3.8 presents the crustal EF in relation to the element Sc (EF_{Sc}) for exposed lichens.

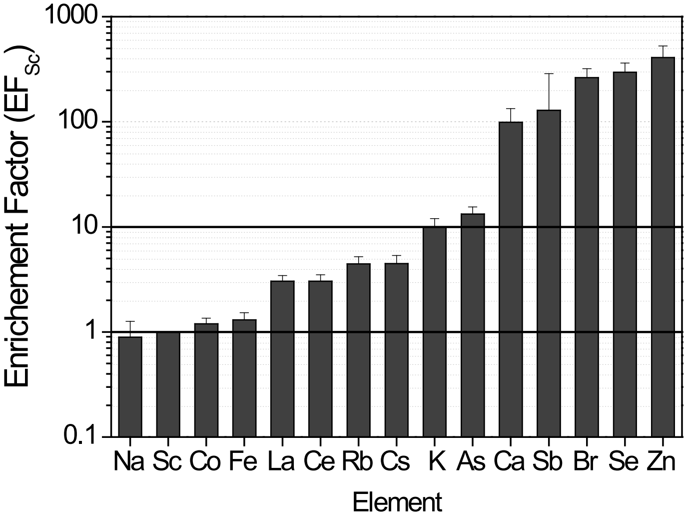


Figure 3.8 – Crustal enrichment factors in relation to the element Sc (EF_{Sc}) for exposed lichens.

GIS provided spatial linkage that enabled the integration of measures of proximity, connectivity, density and other environmental factors (Saelens et al. 2003). The application of GIS on environmental studies has proven to be effective on the identification of hotspots (Lage et al. 2014). Figure 3.9 shows the spatial distribution of the elemental contents measured in the transplanted lichens.

Four main chemical profiles were identified, which accounted for 85% of the total variance (table 3.2). The first factor represented the crustal contribution defined by typical soil elements, such as Cs, Fe, La, Rb and Sc. Results from EF_{Sc} confirmed that Sc, Cs, Fe, La and Rb had a crustal origin. K also presented some association with this factor. Canha et al. (2012c) associated insoluble K with soil sources but this element can also be related to physiological components of lichens (Canha et al., 2014d). These elements presented the highest concentrations on *Cidade* path, probably due to the resuspension of dust in areas of greater traffic (Av. Brasil and Campo Grande), and in *Monsanto* path due to the contribution of unpaved surfaces. The second factor composed with As, Ce and Co might be related with industrial sources located in the north of Lisbon and in the southern of Tagus river (Almeida et al., 2009a).

The third factor, correlated with Sb and Zn, is associated with road traffic, mainly from the abrasion of tires and breaks (Almeida et al., 2007). The highest concentrations of Sb and Zn were found in the paths located near roads with greatest amounts of traffic and near the river, indicating a possible contribution from ships to the increased concentration of these elements. Ca is also associated with this factor, with calcareous rocks used for sidewalk coating (Almeida et al., 2013b) and cement production the main sources in Lisbon. Enrichment factors for Zn, Sb and Ca suggested a significant fraction of these elements originated from noncrustal sources. The fourth factor represented the marine spray, as indicated by the high Na and Br factor loadings. Na concentrations were higher along the *Ribeirinho* path which indicates an association between this element and the sea. Br can have double origin, from sea spray or from combustion process (Calvo et al. 2013). EF suggested the origin of Br from noncrustal sources.

Table 3.2 – Varimax normalized rotated factor loading PCA to exposed lichens

	PC 1 Soil	PC 2 Industry	PC 3 Traffic	PC 4 Sea
As	0.16	**0.69**	-0.30	0.52
Br	0.21	0.23	-0.06	**0.77**
Ca	-0.15	-0.22	0.49	0.33
Ce	0.13	**0.95**	0.11	0.04
Co	0.05	**0.96**	0.11	0.17
Cs	**0.89**	0.07	-0.11	-0.22
Fe	**0.90**	0.09	0.29	0.06
K	**0.85**	-0.08	-0.28	0.11
La	**0.93**	-0.11	0.09	-0.15
Na	-0.35	0.05	0.10	**0.80**
Rb	**0.89**	0.28	-0.13	0.13
Sb	-0.35	0.16	**0.89**	-0.02
Sc	**0.95**	0.09	-0.04	0.08
Se	0.25	-0.68	0.20	0.57
Zn	0.37	0.03	**0.87**	-0.08
% total variance explained	36.2	20.2	14.3	13.9

Figure 3.9– Spatial distribution of the elemental contents measured in the transplanted lichens for As, Br, Ca, Co, Cl, Cu, Fe, K, La, Na, Sb, Sc and Zn (values in ppm).

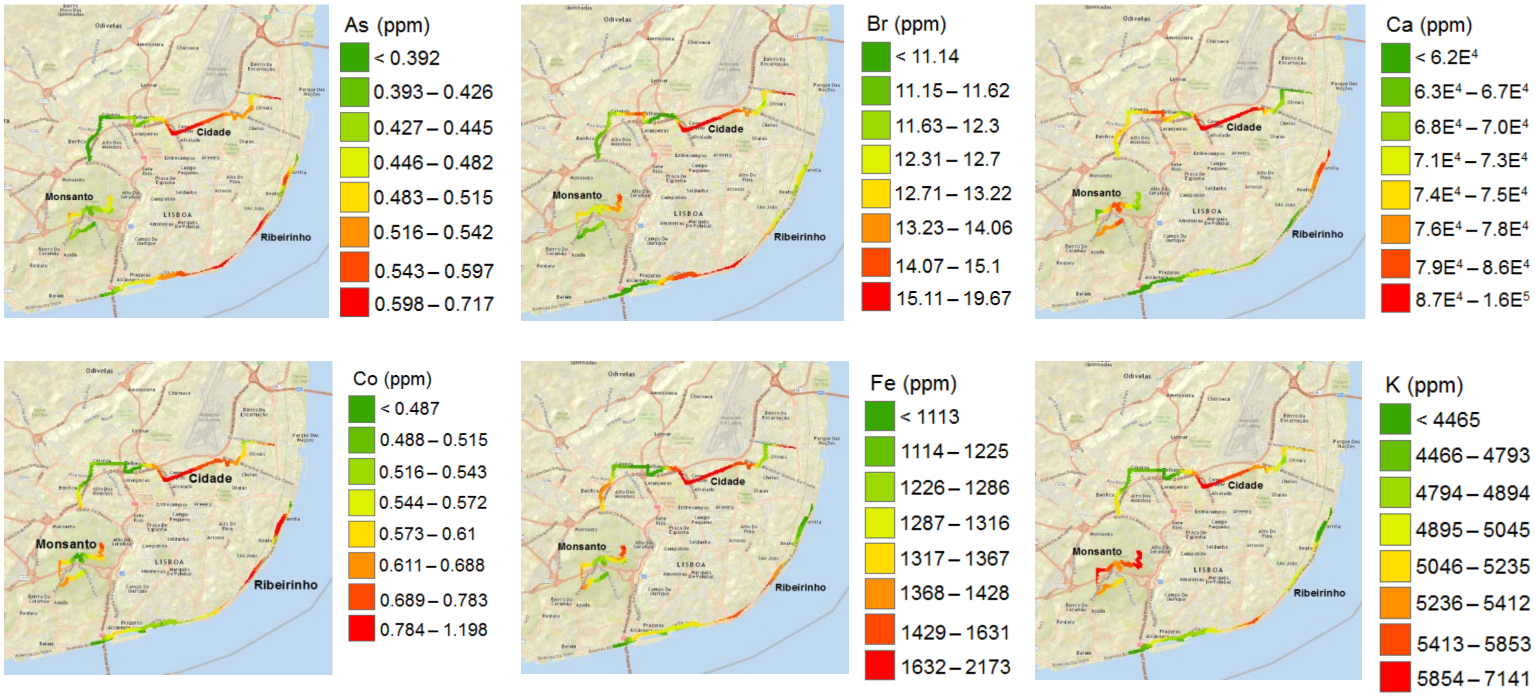
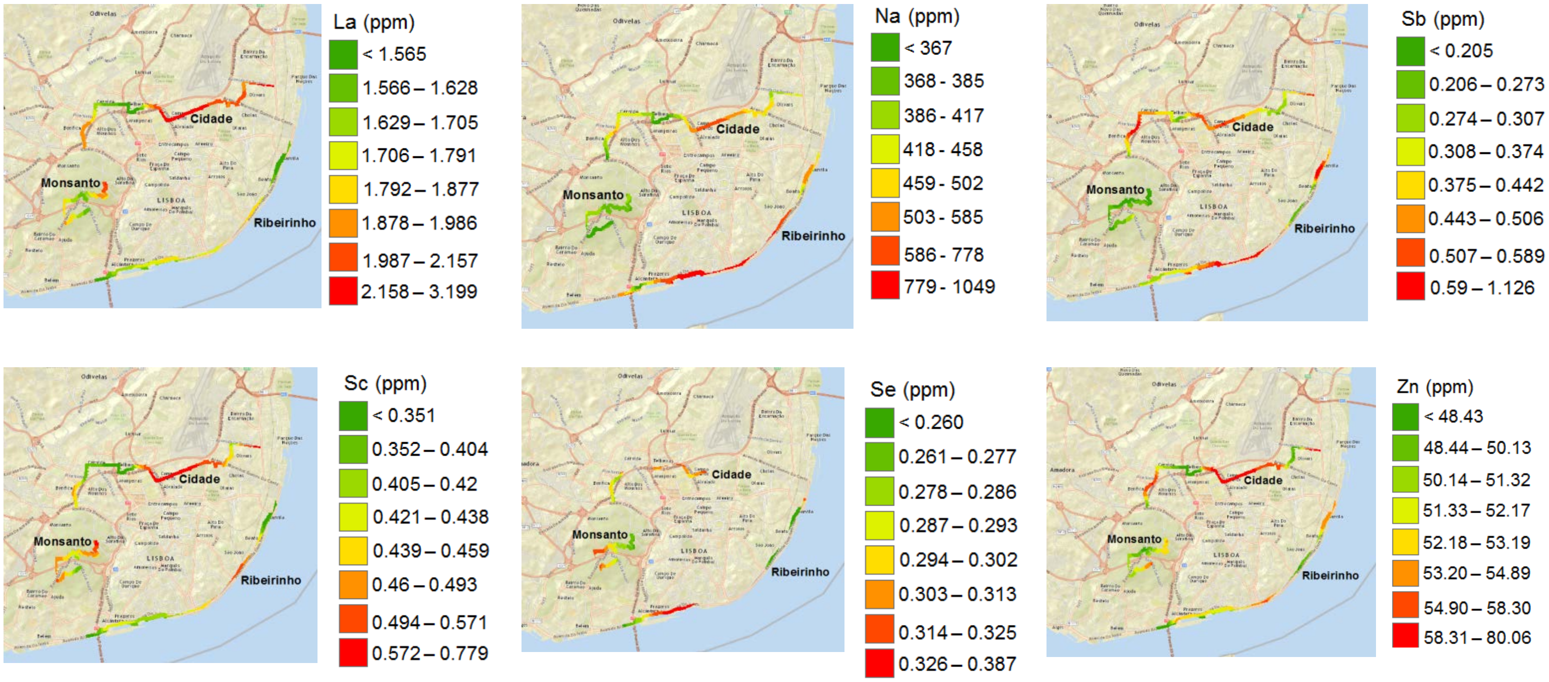


Figure 3.9 (cont.) – Spatial distribution of the elemental contents measured in the transplanted lichens for As, Br, Ca, Co, Cl, Cu, Fe, K, La, Na, Sb, Sc and Zn (values in ppm).



3.1.5. Conclusions

In this work a methodology was developed to assess, by the first time, the exposure to chemical element during cycling. For that besides the traditional use of instrumental approaches, biomonitoring techniques, which have already been successfully used in other contexts, were applied. The developed methodology not only allowed to map the exposure to PM and chemical elements, but also identified the areas with stronger influence of specific emission sources by using PCA, EF_{Sc} and GIS.

PM concentrations measured in different cycling routes and at different time periods and the spatial distribution of element contents indicated that using a bicycle commute route of lower traffic intensity compared to higher proximity of motorized traffic facilitates a significant reduction in exposure. The cycle path close to Tagus river presented the highest concentrations of particles than the other two cycle paths due to the influence of traffic, ship emissions and harbor operations. The evaluation of exposure during different weekdays and periods indicated the importance of the Lisbon low emission zone program in the reduction of pollutant exposure during the week.

The use of bicycles brings benefits not only to health but also for the environment. However, the effects of PM inhalation should not be dismissed when considering healthy populations performing exercise. Exercise should be avoided in areas where PM levels exceed the limit concentration defined by the EU directive e.g. along congested roadways. Considering the acute and chronic physiological responses to PM inhalation, individuals living and exercising in urban areas, in close proximity to major roadways and other emission sources, should consider ambient pollution levels before engaging in active modes of transportation, by selecting the cycling path at a greater distance from PM sources and by avoiding rush hours or other times with elevated vehicular congestion. Athletic filtration masks are also available to reduce PM exposure preventing the rise in systolic blood pressure and decreasing variability in heart rate that normally occurs following exposure to PM. Moreover, creating infrastructures that support alternatives to the use of private transport must be planned and structured. The location of the bike paths should be carefully evaluated and studied by authorities in order to enhance the benefits of this activity and to reduce the negative impacts on health. Cycle paths should be built far from high-traffic roadways and when not possible should have trees, preferably evergreens, placed between the road and the path.

4 Active Transportation

4.1 Air pollutants exposure and inhaled dose during urban commuting: a comparison between cyclists and motorized modes

Based on the article:
Air pollutant exposure and inhaled dose during urban commuting: a comparison between cycling and motorized modes
C.A. Ramos, H.T. Wolterbeek, S.M. Almeida
Air Quality, Atmosphere and Health [in press], DOI: 10.1007/s11869-015-0389-5

4.1.1 Abstract

Active commuting has great health, environment, economic and social benefits. However, cyclists are at risk for exposure to vehicle-related air pollutants due to their proximity to vehicle traffic and elevated respiratory rates. More information on differences in inhaled doses between different transport modes is needed. The aim of this study is to assess and map the exposure of air pollutants to travelers using different transportation modes, and to consider $\dot{V}E$ variability and travel duration for the calculation of inhaled dose. PM10, PM4, PM2.5, PM1, CO, VOC, CO_2, O_3 were measured between December 2013 to March 2014 in a total of 75 travels performed by bus, metro, car, bicycle and motorcycle at five periods of the day (8h, 11h, 14h, 17:30h, 21h). Results showed that car drivers and bus passengers in urban streets may be exposed to higher pollutant levels than cyclists traveling in the same streets. However, this enhanced air pollution exposure is compensated by the higher ventilation rates of cyclists, which presented the highest inhaled doses. To reduce exposure concentrations, spatial and temporal separation of cyclists from motorized vehicle traffic should be achieved with separated bicycle facilities, low volume routes and off-peak travel.

4.1.2 Introduction

A large number of studies have indicated strong associations between ambient air pollution levels and adverse health effects (Kampa and Castañas, 2008; Hall et al., 2010; Pascal et al., 2013). Studies of long-term exposure to air pollutants have showed an increased risk of chronic respiratory illness (Kariisa et al., 2015), cardiopulmonary mortality (Beelen et al., 2014) and development of several types of cancer (Barrett, 2014), whereas higher prevalence of bronchitis, acute cardiovascular disease, asthma and other symptoms have been associated with short-term exposure to air pollution during periods with enhanced concentration levels (Almeida et al., 2014; Cruz et al., 2015; Canha et al., 2011).

There is often substantial traffic density in urban areas and the dispersion of the emitted pollutants is strongly suppressed by the presence of buildings. Air pollution exposure is particularly high for travelers due to the proximity to mobile sources of pollution. Bigazzi and Figliozzi (2014) presented a conceptual diagram linking traffic-related pollution emissions and health effects. In resume, motorized vehicle emissions degrade urban air quality in accordance with atmospheric dispersive, chemical and physical processes. Travelers' exposure concentrations then depend on their travel trajectory and travel duration. The inhalation of traffic-related air pollution mainly depends on travelers' breathing volume while exposed to a pollutant concentration (Int Panis et al. 2010). Uptake of the inhaled pollutants into the body depends on processes in the respiratory tract and other body systems (Bigazzi and Figliozzi, 2014). Finally, the health effects of air pollution uptake doses are a function of the toxicity of the pollutants and physiology of the individual.

Active transportation and sustainable mobility are considered a priority in several European commitments and environmental declarations, such as Parma Declaration, Amsterdam Declaration and the Transport, Health and Environment Pan-European Program 2009-2014. These documents address the need of safe environments by decreasing the incidence of acute and chronic respiratory diseases by reducing exposure of people to air pollutants and improving conditions for physical activity. Moreover, these documents encourage policymakers to develop targeted strategies and concrete measures that support environment-friendly and health-promoting transport, including pedestrian and cycle-friendly cities (WHO, 2009c; WHO, 2010b). Several local policies attempt to reduce traffic emissions in cities by promoting active transportation, encouraging healthier behaviors, promoting physical activity, and improving public transportation (de Nazelle et al., 2012; CML, 2014; Rojas-Rueda et al., 2011; UNEP, 2009).

However, besides the unquestionable health, environmental, economic and social benefits of active transportation, bicycle commuters using on-road routes during peak traffic times are sharing a microenvironment with high levels of motorized traffic, a major emission source of air pollutants. Cyclists are exposed to higher peak concentrations since in-vehicle concentrations are buffered by limited air exchange. In addition, cyclists experience increased physical activity relative to less active commuters which travel via such methods as bus, metro and car. Increased physical effort leads to elevated inhalation rates, thus higher inhaled doses and subsequent higher lung deposition of air pollution per unit time spent in commute.

Moreover, travel duration also influence the potential exposure of bicycle commuters. Bicycling is, by far, the most studied mode of active transportation and has been compared with other transportation modes in several cities in the world (Kaur et al., 2007). However, few studies have taken into account that cyclists have variable and increased inhalation rates relative to other commuters, which influence their inhaled and lung deposition rates. The health risks of air pollution exposure during travel are not easily characterized because of numerous individual, environmental, and traffic factors involved. Health effect studies of cyclists' exposure to air pollution have focused on respiratory and cardiovascular biomarkers following acute (0.5–2h) exposures to traffic. However, these studies showed inconsistent results, some reporting insignificant acute effects and others reporting some cardiovascular or respiratory biomarker changes (Bigazzi and Figliozzi, 2014). Therefore, more research is needed to provide better quantification and understanding of the risks and benefits of changing to active transportation.

The objective of this chapter is to assess and map exposure to CO, CO_2, COV, O_3 and PM for travelers using different transportation modes: bicycle, car, motorcycle, metro and bus. This inter-modal pollution exposure comparison considers comprehensive and representative modal travel characteristics and the variables, $\dot{VE}$ and travel duration for the calculation of inhaled dose. This work contributes for the development of dose-response functions for health effects of chronic short-duration high-intensity air pollution exposure episodes, for the identification of measures to reduce air pollution exposure to the cyclists, and to understand whether the benefits of physical activity from cycling outweigh the risks from air pollution.

4.1.3 Methodology

4.1.3.1 Area of Study

The nature of modern society in many countries both affords and expects a high degree of personal mobility. The Portuguese population spends 80 minutes per day commuting (Renascença, 2015) with 70% of movement in Lisbon 70% are between home and work. Lisbon gathers special conditions affecting the use of bicycles, which motivate the authors to develop this inter-modal study in the capital city of Portugal.

Firstly, Lisbon has peculiar topographic characteristics recognized by its hills (with altitude up to 110m) and planaltic zones (with an average altitude of 80m) (Santos, 2009) so air pollution levels vary significantly between streets and even street sections.

Secondly, air quality in Lisbon frequently exceeds the limit values defined by the European Air Quality Directive (2008/50/EC), with Av. da Liberdade considered one of the most polluted avenues in Europe. In Lisbon, traffic is the main source of atmospheric pollution (Almeida et al., 2009a,b). Due to the geographic position of Lisbon – on the extreme southwest of Europe – and to the dominant western wind regime, influenced by the presence of the semi-permanent Azores high-pressure and the Icelandic low-pressure systems over the North Atlantic Ocean, high levels of pollutants should be expected. The transport of maritime air mass is usually associated with cleaner air masses from the Atlantic Ocean and with better dispersion conditions of pollutants coming from industrial areas (Almeida et al., 2013b). Nevertheless, high concentrations of air pollutants are registered under adverse meteorological conditions, low dispersion conditions and thermal inversions. Moreover, in Lisbon, natural PM sources cause a number of PM exceedances. Prior studies in Lisbon have shown that natural mineral particulate sources such as high-dust Saharan air mass intrusions interfere with the monitoring of the incidence of anthropogenic emissions on ambient air PM levels (Almeida et al., 2008; Almeida et al., 2013a; Almeida-Silva et al. 2013; APA, 2013).

Thirdly, in the last years Lisbon increased the incentive for the use of bicycles as an active mode of transportation, or as a complement of public transportation by including changes in the Portuguese Road Code and changes in the municipality regulations for cyclists (Barreto, 2013).

In the present study, exposure was determined for working days along a popular Lisbon route (figure 4.1) going from a residential zone (Telheiras neighborhood) to the working place located in the city center (Restauradores square). The route passes through the main squares of the city (Campo Grande, Entrecampos, Saldanha, Marquês de Pombal and Restauradores), where several areas of trade, services and employment are located. The 7km route was the same for bicycle, car, motorcycle, buses and metro. This route has a metro line (green line of the Lisbon Metro), a bus line and part of it has cycle paths (designated for cyclist use within the right of way of a public road but outside of the roadway) and a cycle lane (part of the road on both sides, separated by a solid white line).

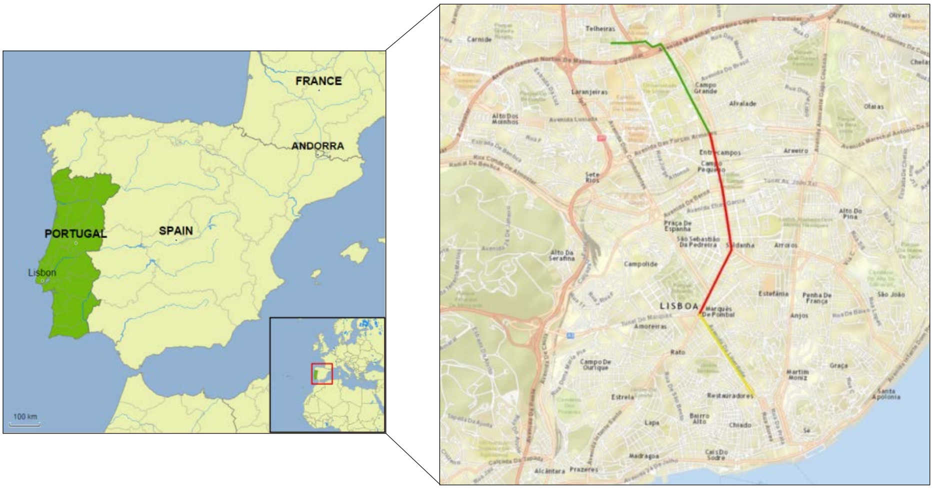


Figure 4.1 – Localization of Lisbon in Europe and identification of the selected route. Red line – road; Green line – cycle path; Yellow line – cycle lane.

In the present study, exposure was determined for working days along a popular Lisbon route (figure 4.1) going from a residential zone (Telheiras neighborhood) to the working place located in the city center (Restauradores square). The route passes through the main squares of the city (Campo Grande, Entrecampos, Saldanha, Marquês de Pombal and Restauradores), where several areas of trade, services and employment are located. The 7 km route was the same for bicycle, car, motorcycle, buses and metro. This route has a metro line (green line of the Lisbon Metro), a bus line and part of it has cycle paths (designated for cyclist use within the right of way of a public road but outside of the roadway) and a cycle lane (part of the road on both sides, separated by a solid white line).

Taking into account the day-to-day variations during sampling, the air quality data from two monitoring stations (Avenida and Entrecampos) of the national air quality network was compared. Mann-Whitney test results indicated no differences among the sampling days of aboveground transportation modes.

4.1.3.2 Equipment and Measuring Procedure

Measurements were performed over 15 days, evaluating the five transportation modes already indicated. In each sampling day, five periods were monitored in order to recognize daily patterns (8h, 11h, 14h, 17:30h and 21h). Monitoring occurred on three different occasions resulting in a total of 75 travels. Measurements were conducted from December 2013 to March 2014 on non-rainy days. Due to the lack of instruments sampling was not performed simultaneously for the different modes. Sampling days are presented in Table 4.1.

PM10, PM4, PM2.5 and PM1 were measured with a DustTrak monitor (8530 model, TSI, USA). For quality control, the DustTrak monitor and a Gent sampler worked in parallel during 8h over 1 week. Figure 4.2 shows that PM2.5 concentrations registered by the DustTrak were higher than the PM2.5 levels measured by gravimetry (considered as the reference method), while PM10 concentrations measured by both equipment were equivalent. A calibration factor (β) was calculated and applied to the PM concentrations obtained by the DustTrak. The calibration factor was obtained by calculating the ratio between the concentrations obtained by the gravimetric method and the concentrations measured by the DustTrak monitor (McNamara et al. 2011; Diapouli et al. 2008). Prior to each monitoring run, the unit was manually zeroed using a zero-air attachment connected to the equipment.

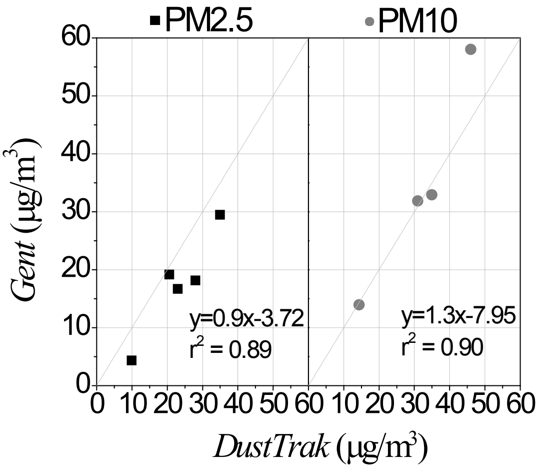


Figure 4.2 – Control chart showing the comparison between PM2.5 and PM10 measured by DustTrak and gravimetry (values in µg/m³).

CO_2, VOC, CO, O_3, temperature and relative humidity were measured with a Graywolf (610 IAQ probe, Wolfsense Solutions, USA). Measurements were recorded every second. Before measuring, sensors were calibrated in laboratory with standard gases according to the manufacturer specifications. DustTrak and Graywolf were calibrated annually by the manufacturer. A GPS unit (eTrex20, Garmin, USA) was utilized.

The bus transportation system in Lisbon is composed of recent buses (2007) equipped with air conditioning. The metro line is 52 years old (the first station of green line started at 1963 and the line was completed at 2002) but the carriages are 16 years old and are equipped with air conditioning system and the windows cannot be opened (Transportes de Lisboa, 2012). The motorcycle was a Honda Pan European 1.3 diesel, with measurements performed by two persons. Bicycle sampling was performed with two persons cycling side by side: one carried the Graywolf equipment and the other carried the DustTrak. In these samplings, the equipment was positioned inside a backpack and inlets were placed near the breathing zone. Car monitoring was carried out in a 2002 Citroën Saxo 1.1 gasoline and only the driver was inside the car during the sampling. The equipment was placed in the passenger seat and the driving conditions were standardized as much as possible: car was driven with the windows closed and air condition and the fan ventilation system off.

4.1.3.3 Statistical and Data Analysis

A geodatabase, collating repeated single trip data sets was used with geographic information system (GIS) software (ArcGIS10®, ESRI, USA) to graphically represent the exposure on the monitored commute routes. ANOVA and Wilcoxon matched pairs tests (in Statistica12® software) were used to assess the differences between transport modes and measuring periods. A level of significance as defined as $p<0.05$.

4.1.4 Results and Discussion

4.1.4.1 Meteorological Data

Table 4.1 presents the meteorological data, for Lisbon during the measuring campaigns obtained from a weather station located in the center of Lisbon (38°46'N, 9°08'W). Results show that measurements performed in March presented the highest temperature, with relative humidity always above 60%. Precipitation was 0mm for all the measured days. No statistical differences between the five modes were found for temperature (One Way ANOVA, $p=0.66$, IC 95%) or relative humidity (One Way ANOVA, $p=0.67$, IC 95%).

4.1.4.2 Differences in Exposure Between Modes of Transport

Taking into account the day-to-day variations during sampling, the air quality data from two monitoring stations of the national air quality network (Avenida and Entrecampos) was compared. Mann-Whitney test indicated no differences among the sampling days of aboveground transportation modes.

Table 4.2 presents the mean pollutant concentration measured during commuting. CO, a toxic by-product of incomplete combustion, is one of the main traffic-related air pollutants linked to health risks for road travelers. Highest concentration levels of CO occurred during rush hours (principally at

8h) and in cars and buses. The concentrations of this primary pollutant are particularly high near roadways and during rush hours. In cars and buses, CO concentrations increase not only due to the emissions from neighbouring vehicles but also from cars and buses own exhaust can infiltrate into the cabin (Wong et al., 2011).

Table 4.1 – Meteorological data for Lisbon during the campaigns

		Temperature (°C)			Relative Humidity (%)			Precipitation (mm)
Transport	Day	min	mean	Max	min	mean	Max	
Bicycle	03/12/13	5	9	14	48	60	76	0.00
	06/03/14	7	13	20	46	87	100	0.00
	13/03/14	7	13	20	43	75	94	0.00
Metro	05/12/13	7	12	17	52	70	93	0.00
	22/01/14	10	12	14	51	82	100	0.00
	21/02/14	7	10	13	51	75	93	0.00
Car	06/12/13	7	11	16	52	67	82	0.00
	04/02/14	9	11	14	77	89	100	0.00
	17/03/14	9	14	21	42	68	88	0.00
Bus	09/12/13	2	8	14	59	83	100	0.00
	20/01/14	7	10	14	63	81	93	0.00
	18/03/14	10	13	17	59	76	88	0.00
Moto	12/12/13	11	13	15	72	81	88	0.00
	24/01/14	8	11	14	72	82	93	0.00
	12/03/14	8	14	20	40	60	87	0.00

Limasset et al. (1993) observed that in buses, air intake from the roof rather than from the front of the bus resulted in significantly lower concentrations of CO. Previous studies recorded CO concentrations in a range of 2700 – 48000 µg/m^3 in buses (Wong et al. 2011) and 1300 – 4833 µg/m^3 in cars (de Hartog et al. 2010). This study presented lower values with an average CO concentration of 558 µg/m^3 for buses and 786 µg/m^3 for cars. The lowest CO concentrations were measured for bicycle commuters. Bigazzi and Figliozzi (2014) measured an average cyclists' exposure to CO between 600 and 15000 µg/m^3 which is higher than the values registered in the present study (average 140 µg/m^3). Figure 4.3 presents the ratios of exposure between bicycles and all the other transport modes and shows averaged CO ratios of 0.25, 0.67, 0.18 and 0.32 for bus, metro, car and motorcycle, respectively.

CO_2 levels were found to be significantly higher in-cabin. Car presented the highest average concentration (1960 µg/m^3), followed by bus (1820 µg/m^3) and metro (1270 µg/m^3). CO_2 is a pollutant emitted by the human metabolism (Ramos et al., 2014) and it is commonly used as an indicator of occupancy and poor ventilation (Canha et al., 2013). This fact explains why the highest levels were measured in public transports during rush hours when the occupancy is higher. The greatest concentrations of CO_2 in-car were measured during travels which took more time. Figure 4.3 shows CO_2 average ratios for bicycle/bus, bicycle/metro, bicycle/car and bicycle/motorcycle of 0.45, 0.67, 0.43 and 1.1, respectively.

PM concentrations were higher during travels which began at 8h, except for metro that presented the highest levels at 17h30. Greater number of vehicles are present during rush hour periods which produce particles, not only from the vehicles' exhaust but also from tires and brake wear and soil resuspension (Canha et al, 2014a; Almeida et al., 2015; Almeida-Silva et al., 2015). The proximity to vehicle exhaust plays a major role in infiltration, especially when the vehicles are queuing. In-cabin, PM levels are affected by air infiltration and indoor resuspension of floor dust due to various passenger-related activities, e.g. alighting, boarding and taking a seat (Canha et al., 2014b). The PM10 and PM2.5 average ratios for bicycle/bus (1.3, 1.4), bicycle/metro (1.2, 1.3), bicycle/car (1.3, 1.3) and bicycle/motorcycle (1.2, 1.1) show that the measured PM concentrations were higher during bicycle commutes. Studies have showed that PM2.5 exposure experienced by car drivers were modestly higher than those experienced by cyclists, with mean bicycle/car ratios of 0.9 (Hartog et al., 2010).

Cyclists' average PM10 exposure levels have been measured in the range of 50.2 ± 12.0 µg/m^3 (Bigazzi and Figliozzi, 2014) which is lower than the values registered in the present study (average of 76 µg/m^3). Cyclist PM2.5 exposure levels were extensively examined by Adams et al. (2001) in London. Cyclists recorded an average PM2.5 personal exposure concentration of 23.5 µg/m^3, for the same season as in this study (Adams et al., 2001). Kaur et al. (2005) observed similar exposure concentrations for cyclists (33.5 µg/m^3, 9.7–77.5 µg/m^3) in the same city. Average PM2.5 concentrations measured in Lisbon presented a higher value (66 µg/m^3) when comparing with these studies.

O_3 presented the highest average concentrations in bicycle (350 µg/m^3) and motorcycle (200 µg/m^3) commuting. O_3 occurs as a secondary pollutant, principally as a result from traffic.

Table 4.2 – Mean and standard deviations of pollutant concentrations for the five types of transportation, during the studied periods.

		8h (mean ± stdev)	11h (mean ± stdev)	14h (mean ± stdev)	17.30h (mean ± stdev)	21h (mean ± stdev)
VOC (µg/m^3)	Bus	224 ± 180	239 ± 52	287 ± 69	203 ± 34	198 ± 30
	Metro	198 ± 33	124 ± 25	164 ± 22	176 ± 26	125 ± 21
	Car	370 ± 38	447 ± 37	471 ± 29	326 ± 11	206 ± 12
	Moto	187 ± 224	120 ± 41	336 ± 762	675 ± 1447	174 ± 55
	Bicycle	149 ± 48	175 ± 207	212 ± 47	266 ± 92	287 ± 366
CO$_2$ (mg/m^3)	Bus	2304 ± 608	1804 ± 437	1990 ± 728	1681 ± 660	1343 ± 487
	Metro	1189 ± 293	1181 ± 243	1377 ± 332	1438 ± 381	1150 ± 52
	Car	1669 ± 344	1440 ± 245	1832 ± 318	2870 ± 707	1982 ± 492
	Moto	823 ± 86	785 ± 77	763 ± 73	812 ± 69	786 ± 87
	Bicycle	845 ± 91	823 ± 71	792 ± 44	880 ± 70	924 ± 100
O$_3$ (µg/m^3)	Bus	85 ± 43	56 ± 33	65 ± 36	95 ± 34	97 ± 32
	Metro	91 ± 30	46 ± 17	35 ± 23	59 ± 20	87 ± 21
	Car	496 ± 207	64 ± 25	36 ± 18	72 ± 13	69 ± 22
	Moto	313 ± 196	413 ± 316	340 ± 183	330 ± 239	355 ± 178
	Bicycle	259 ± 98	197 ± 72	169 ± 76	154 ± 79	222 ± 82
CO (µg/m^3)	Bus	961 ± 763	510 ± 445	534 ± 425	676 ± 363	112 ± 152
	Metro	772 ± 994	81 ± 130	64 ± 76	234 ± 244	101 ± 108
	Car	1235 ± 676	875 ± 437	823 ± 432	759 ± 391	239 ± 215
	Moto	689 ± 868	345 ± 480	466 ± 839	527 ± 1306	403 ± 816
	Bicycle	193 ± 530	188 ± 529	109 ± 232	208 ± 481	na
PM$_1$ (µg/m^3)	Bus	53 ± 51	29 ± 18	51 ± 26	49 ± 23	39 ± 16
	Metro	45 ± 16	32 ± 12	45 ± 19	63 ± 48	39 ± 9.4
	Car	59 ± 11	54 ± 13	51 ± 10	40 ± 6.9	34 ± 6.6
	Moto	89 ± 298	55 ± 47	38 ± 25	57 ± 107	53 ± 21
	Bicycle	83 ± 64	69 ± 47	43 ± 38	66 ± 43	78 ± 26
PM$_{2.5}$ (µg/m^3)	Bus	56 ± 55	30 ± 18	52 ± 27	50 ± 23	40 ± 16
	Metro	51 ± 18	35 ± 13	48 ± 20	67 ± 49	42 ± 9.9
	Car	61 ± 12	55 ± 13	52 ± 10	41 ± 7.0	35 ± 6.7
	Moto	91 ± 298	57 ± 47	39 ± 25	59 ± 107	54 ± 21
	Bicycle	85 ± 66	70 ± 47	44 ± 38	67 ± 43	65 ± 26
PM$_4$ (µg/m^3)	Bus	64 ± 69	32 ± 20	56 ± 30	53 ± 25	40 ± 17
	Metro	58 ± 21	39 ± 15	53 ± 20	71 ± 50	45 ± 11
	Car	64 ± 13	58 ± 15	54 ± 11	42 ± 7.6	37 ± 6.9
	Moto	93 ± 298	59 ± 48	41 ± 25	61 ± 108	56 ± 21
	Bicycle	89 ± 72	71 ± 48	45 ± 38	69 ± 44	81 ± 26
PM$_{10}$ (µg/m^3)	Bus	70 ± 82	39 ± 29	67 ± 44	60 ± 33	45 ± 25
	Metro	68 ± 24	43 ± 17	60 ± 22	80 ± 58	51 ± 13
	Car	73 ± 20	66 ± 21	61 ± 13	46 ± 9.5	39 ± 7.9
	Moto	98 ± 300	62 ± 48	43 ± 26	66 ± 110	58 ± 22
	Bicycle	101 ± 100	74 ± 51	48 ± 39	74 ± 46	84 ± 28

Outdoor concentrations of this pollutant are found to be higher than the levels measured indoors (Almeida-Silva et al., 2014a). Average O_3 ratios between bicycle/bus (1.3), bicycle/metro (1.2), bicycle/car (1.3) and bicycle/motorcycle (1.1) show that measured O_3 concentrations were higher during bicycle commuting.

Highest VOC average concentrations were recorded in car (364 $\mu g/m^3$). Car commuters have generally been shown to be exposed to highest concentrations of VOC air pollution. This may be due to their position on the road, close to where the majority of VOC emissions originate (in main traffic lanes close to car exhausts). McNabola (2008) showed that transportation by car has greater concentrations of VOC species (benzene, butadiene, ethane, ethylene and acetylene) when compared with public bus, bicycle and pedestrian. The ratios between bicycle and all the other transport modes varied not only with the transport mode but also with the hour of the day. This fact can be due to the multi-origin of VOC. Outdoors, VOCs are emitted principally by vehicles; indoors, these pollutants can be released from indoor materials and occupants (Ramos et al., 2014; Canha et al., 2012a) in addition to outdoor infiltration.

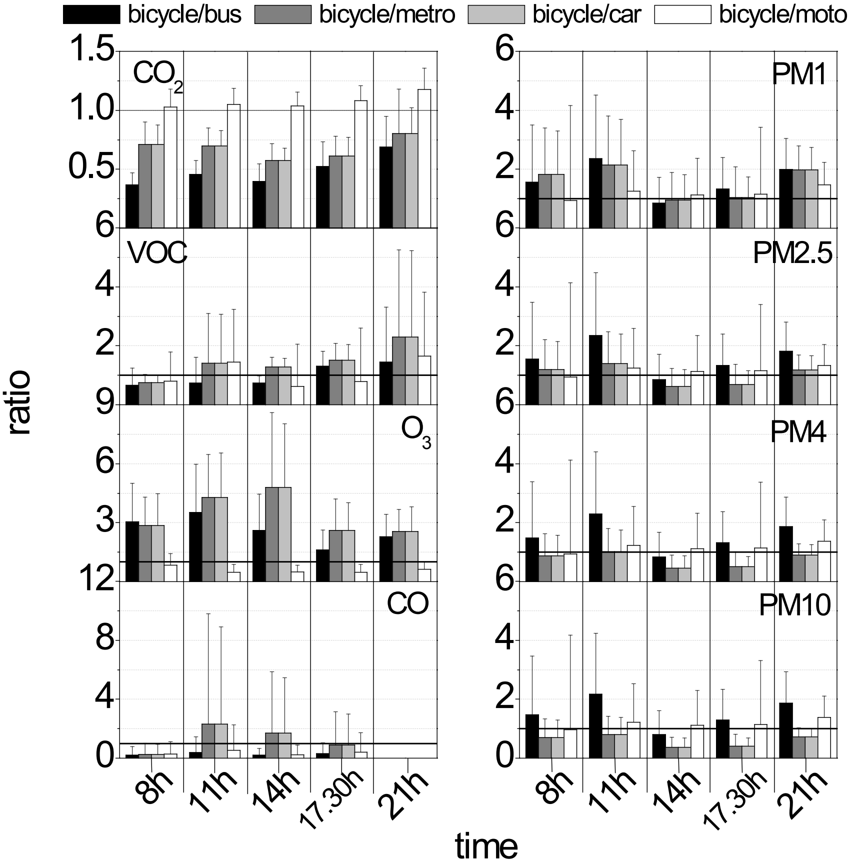


Figure 4.3 – Ratios of concentrations between bicycle and all the other studied transportation modes.

4.1.4.2.1 Public transports

With the exception for PM, which did not present significant differences between transport modes, metro showed lower concentration of pollutants compared with buses. A number of studies have revealed poor air quality in metro systems, especially concerning the levels of PM (Salma et al., 2007; Raut et al., 2009; Ye et al., 2010). Other studies performed in metro systems report, contrastingly, relatively low PM levels (Kam et al., 2011; Múgica-Álvarez et al., 2012). Salma et al. (2007) interpreted such differences in PM levels among the metro systems to be due to the age of the metro, abrasion of railways and catenary metal, braking systems and ventilation. According to Querol et al. (2012) the main PM emission sources in metro are the mechanical abrasion of rail/wheel, the resuspension of material caused by air turbulence, PM emitted during night-time maintenance works, cleaning activities, surface air uptake from the surfaces and wind erosion by intense air flow within the tunnels and platforms. PM results measured in Lisbon were in the same range of values as measured by Querol et al. (2012) in Barcelona's metro for PM10 (43-79 µg/m^3 in Lisbon and 36-100 µg/m^3 in Barcelona) but higher for PM2.5 (35-66 µg/m^3 in Lisbon and 11-32 µg/m^3 in Barcelona). Adams et al. (2001) measured higher PM2.5 concentrations in London (105-371 µg/m^3). Higher concentrations of CO_2 were measured in buses than in metro across the five periods of time measured. This fact may be related with the size of the carriage (which are larger in metro, thus promoting the dilution of the pollutants), the number of occupants and the efficiency of the ventilation system. CO and VOC concentrations in buses were also higher in buses than in metro due to their proximity to traffic, their principal emission sources. The majority of tropospheric O_3 formation occurs when NOx, CO and VOC react in the atmosphere in the presence of sunlight, so it was expected that the concentration of this pollutant would be higher in buses because they circulate at surface, where all the conditions necessary for O_3 formation are gathered.

Taking the metro or the bus involves waiting periods at the respective stations which also accounts for the daily human exposure. Figure 4.4 compares the average concentrations measured during the commute and during the periods spent waiting at platforms, for bus and metro (in the five periods of sampling and in three sampling days). Results show that inside the bus and metro, CO_2 concentrations were greater than at the platform, due to the higher human density and lower dilution that occurs in-cabin. O_3 concentrations were higher at bus waiting platforms than in-cabin. For the metro, no significant differences were observed except for two measuring periods that presented higher O_3 levels in-cabin. CO and VOC concentrations were higher inside buses than on waiting platforms, inpart due to the infiltration of atmospheric pollutants, combustion gases and gasoline vapor from the vehicle itself or surrounding vehicles. Moreover, some VOC species, such as toluene, are major constituents used as solvents in painting and surface coating in vehicles (Chen et al., 2011; Su et al., 2013). In metro, VOC concentrations on the platform and in-cabin did not present significant differences, except during the 8h rush hour that was characterized by higher in-cabin VOC levels. This fact can be explained by a higher number of passengers inside the train that can be considered VOC sources due to the use of hygiene products (perfume, deodorants, hair spray) (Wang C. et al., 2014; Steinemann, 2015) .

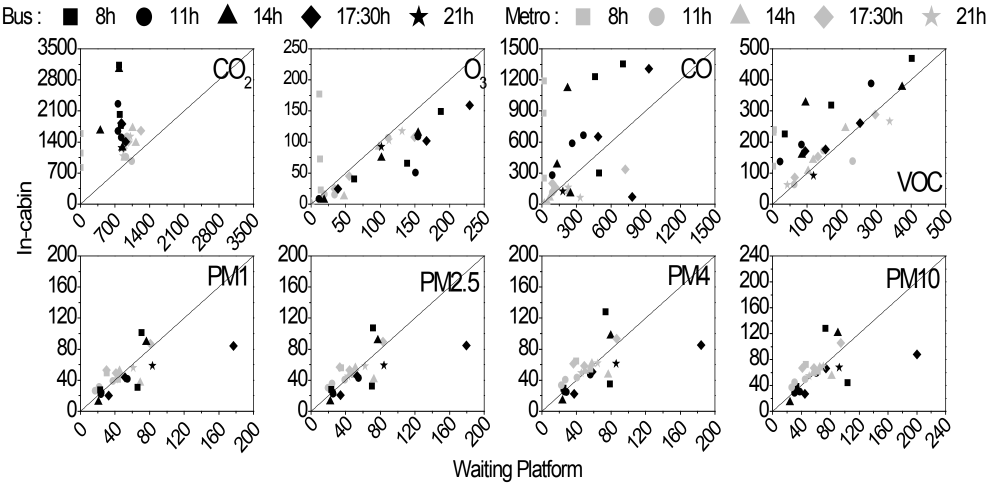


Figure 4.4 – Concentrations in the waiting platform and of indoor environment of the transport. Values in mg/m^3 for CO_2 and in µg/m^3 for the remaining pollutants.

PM levels on bus and metro station platforms did not significantly differ from the concentrations measured in-cabin. In Barcelona, Querol et al. (2012) measured greatly elevated PM levels on the station platforms, being around 3-4 times higher than in trains. In Lisbon, PM10 concentrations on platforms and cabins were 56 and 60 µg/m^3, respectively. In previous works, the mean PM10 levels on the platform ranged from 51 to 407 µg/m^3 (Querol et al., 2012). PM2.5 concentrations on platform and in-cabin measured in our study were 46 and 49 µg/m^3, respectively. Mean PM2.5 concentrations measured on the platforms of the priorly referenced studies ranged from 33 to 129 µg/m^3.

4.1.4.2.2 Private transports

Car, motorcycle and bicycle are the three types of private transports that were studied. The exposure during commuting by these modes of transportation were completely different, dependent on a large number of factors such as the selected route, car speed, trip duration, car type, ventilation status, driving behavior, street configuration and weather conditions (Kaur et al., 2007). However, in general, indoor air quality is affected by limited pollutant dilution and indoor emissions. On motorcycles and bicycles, individuals do not have any barrier against the pollutants generated outdoor. Consequently, the highest O_3 and PM levels were measured during the use of motorcycle and bicycle whereas cars presented the highest concentrations of CO_2, VOC and CO. Differences of exposure to air pollutants between traffic modes is likely due to varying proximity to emission sources. In general, cyclists receive lower levels of exposure to primary pollutants, originating from traffic, as they are able to avoid pollutant sources by dodging between vehicles and by consistently being next to the curb, whereas vehicle drivers are have greater exposure by being in the direct proximity of emission sources – although inside the vehicle.

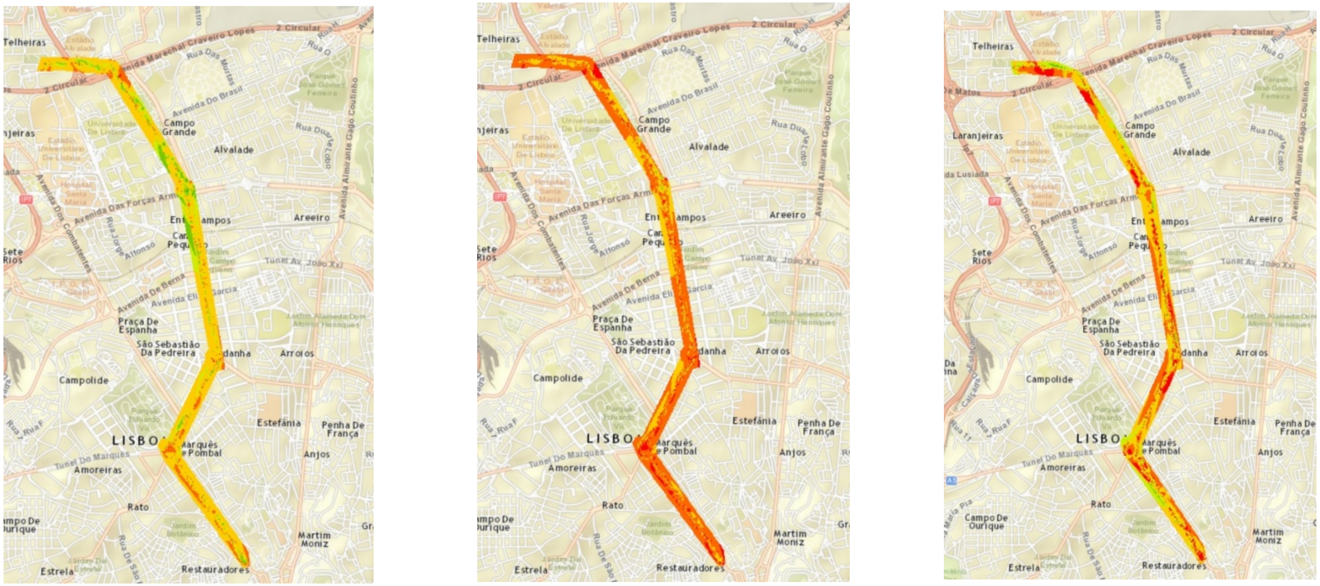
Differences in peak concentrations between car driving and cycling were observed. Higher short-term peaks occurred while cycling but they were of shorter duration than the peaks measured in the car. This is probably explained by the mixing that take place within the air volume of the car and possibly by the shorter contact times with the emissions of the vehicles while cycling. Relatively long peaks measured in cars may be caused by the high-emission from closer vehicles as well as from self-contamination by own emissions.

4.1.4.3 Pollutant Maps

A pollutant map is a graphic representation of pollutants' concentrations projected on a street plan or aerial photograph. Pollutant maps depict measured concentrations coupled with the GPS position and then projected in the entire route. These maps are very suitable to detect hotspots of high pollutant concentrations (Berghmans et al., 2009). The concentrations were mapped using ArcGIS 10 (ESRI®) and interpolated with the Natural Neighbor technique to obtain a continuous representation of the data. The PM10, PM2.5 and O_3 classes were created according with the European directive for ambient air quality (2008/50/CE). In all maps (figure 4.5), it is possible to observe hot spots along the route. PM10 and PM2.5 demonstrated lower concentrations, especially in the cycle paths from Telheiras and Campo Grande (69 µg/m^3 and 59 µg/m^3, respectively). PM10 hotspots coincide with stop lights placed in zones with high traffic (closer to 2^a circular road and Marquês de Pombal). PM2.5 concentrations were very high along most parts of the route, in particular in the segment Marquês de Pombal – Restauradores (68 µg/m^3), depicting increased concentrations of red regions (representative of high concentrations of pollution). O_3 presents disperse concentrations of hotspots along the route. PM2.5 and O_3 are known as a precursor for cardiovascular and respiratory illnesses (Srebot et al. 2009; Kim et al. 2011; Cruz et al. 2015), thus representing a cause for alarm regarding possible health effects on Lisbon cycle users (Garret and Casimiro 2011).

Giles and Koehle (2013) reported in their review the negative impact of O_3 on lung function during exercise, not only in children but also in healthy male and female and athletes. VOC concentrations were higher in the road zone (219 µg/m^3) comparing with the cycle path (181 µg/m^3). This visualization reveals that distance as a function to source emissions have a beneficial effect in reducing the levels of exposure to air pollutants. The lowest concentrations of PM10, PM2.5, O_3 and VOC were obtained for the cycle paths which were placed further away from the traffic.

Figure 4.5 – Map of pollutant average concentrations of a) PM10, b) PM2.5, c) O_3, d) VOC and e) CO for the bicycle route.



a) [PM10] $\mu g/m^3$
Mean = 68
St.dev = 27

b) [PM2.5] $\mu g/m^3$
Mean = 63
St.dev = 23

c) [O_3] $\mu g/m^3$
Mean = 227
St.dev = 56

Figure 4.5 (cont) – Map of pollutant average concentrations of a) PM10, b) PM2.5, c) O_3, d) VOC and e) CO for the bicycle route.

d) [VOC] $\mu g/m^3$

Mean = 161

St.dev = 159

e) [CO] $\mu g/m^3$

Mean = 129

St.dev = 280

4.1.4.4 Inhaled Dose During Commuting

Minute ventilation rates of cyclists are higher than that of motorcycle, car, bus or metro passengers due to their increased physical activity. This fact and the duration of the commute have a great impact on the inhaled doses associated with each transport mode.

The inhaled dose was estimated for each transport mode and air pollutant according to equation 2 (chapter 3.1.4.2.2). Table 4.3, which presents the mean time spent on the round commute from Telheiras to Restauradores, shows that commuting by bus and bicycle results in the highest travel time. The $\dot{VE}$ used was already presented on table 3.1 (chapter 3.1.4.2.2).

Table 4.4 shows the estimated inhaled doses of pollutants for the different sampling periods. The comparison between table 4.2 and table 4.4 shows that comparisons of exposure concentrations by travel modes are not entirely relevant because of the dominating effect of breathing differences among modes. Comparisons of pollution doses between travel modes go beyond examining only exposure concentrations by including the $\dot{VE}$ and travel duration to compare intake dose per trip. Results show slower travel speeds and a fourfold increase in $\dot{VE}$ for cyclists compared to motorcyclists, which resultantly outweighs any beneficial exposure concentration differences. The only exception was observed for CO at 8h (17.13 µg/km for bus and 12.89 µg/km for bicycle) and 14h (9.18 µg/km for bus and 7.74 µg/km for bicycle).

Table 4.3 – Mean time (*t*) spent in the round commuting trips from Telheiras to Restauradores (values in minutes)

	Bus (mean ± stdev)	Metro (mean ± stdev)	Car (mean ± stdev)	Bicycle (mean ± stdev)	Moto (mean ± stdev)
8h	91 ± 7.4	52 ± 2.8	37 ± 2.4	82 ± 33	44 ± 8.5
11h	81 ± 5.9	55 ± 8.8	34 ± 2.7	86 ± 10	36 ± 5.2
14h	87 ± 19	52 ± 12	39 ± 3.9	88 ± 1.9	32 ± 10
17:30h	64 ± 21	46 ± 4.8	46 ± 2.5	94 ± 17	36 ± 3.5
21h	63 ± 26	38 ± 15	32 ± 10	51 ± 6.0	27 ± 2.7

Comparing the results of table 4.4 obtained in the present work with those in the study developed by Nyhan et al. (2014), it is possible to verify that the PM10 mean ratio bicycle/bus inhaled dose is higher in Lisbon (6.4 compared with 4.4). Nazelle et al. (2012) also presented lower ratios of bicycle/bus and bicycle/car for PM2.5 (respectively, 1.82 versus 6.6 and 1.66 versus 12.2) and for CO, with the exception of bicycle/bus (respectively, 1.06 versus 1.01 and 0.35 versus 1.44).

Table 4.4 – Estimated doses for each transport mode (bus, metro, car, bicycle and motorcycle), during the studied periods.

Transport	Pollutant	8h	11h	14h	17h	21h
VOC (µg/km)	Bus	10.02	6.63	8.29	7.92	4.65
	Metro	19.70	18.86	24.38	12.69	12.09
	Car	13.16	14.71	17.95	14.55	6.33
	Moto	7.97	4.25	10.34	11.88	4.60
	Bicycle	49.06	59.74	74.10	99.69	57.92
CO_2 (mg/km)	Bus	60.31	63.05	69.56	64.90	42.88
	Metro	202.59	142.46	168.84	105.25	82.07
	Car	59.39	47.38	69.72	128.20	60.95
	Moto	35.06	27.69	23.46	28.71	20.81
	Bicycle	278.03	281.51	278.16	330.09	186.72
O_3 (µg/km)	Bus	4.60	2.46	1.79	2.67	3.25
	Metro	7.47	4.40	5.52	5.95	5.92
	Car	17.66	2.11	1.37	3.19	2.12
	Moto	13.32	14.56	10.47	11.67	9.41
	Bicycle	85.35	67.37	59.48	57.81	44.87
CO (µg/km)	Bus	39.17	4.34	3.22	10.55	3.77
	Metro	84.45	40.25	45.26	42.33	6.84
	Car	43.95	28.79	31.31	33.92	7.35
	Moto	29.33	12.19	14.34	18.64	10.67
	Bicycle	63.52	64.18	38.13	78.22	0.02
PM_1 (µg/km)	Bus	2.29	1.71	2.28	2.85	1.47
	Metro	4.65	2.30	4.29	3.08	2.39
	Car	2.09	1.78	1.95	1.79	1.06
	Moto	3.77	1.94	1.17	2.02	1.40
	Bicycle	27.19	23.48	15.10	24.69	15.72
$PM_{2.5}$ (µg/km)	Bus	2.57	1.88	2.45	3.00	1.56
	Metro	4.89	2.36	4.40	3.15	2.42
	Car	2.16	1.82	1.99	1.83	1.08
	Moto	3.86	2.00	1.21	2.07	1.43
	Bicycle	28.01	23.81	15.38	25.23	13.04
PM_4 (µg/km)	Bus	2.94	2.08	2.69	3.21	1.69
	Metro	5.64	2.53	4.71	3.31	2.50
	Car	2.28	1.90	2.07	1.89	1.12
	Moto	3.97	2.07	1.25	2.14	1.48
	Bicycle	29.13	24.25	15.77	25.97	16.31
PM_{10} (µg/km)	Bus	3.43	2.32	3.04	3.60	1.88
	Metro	6.15	3.09	5.69	3.77	2.74
	Car	2.60	2.17	2.31	2.06	1.21
	Moto	4.18	2.18	1.33	2.33	1.54
	Bicycle	33.09	25.35	16.89	27.83	16.88

Due to the lack of instruments, the major weakness of the study is the fact that the sampling was not performed simultaneously for the different modes. It would be important to normalize our measurements with background measurements performed by a static monitoring station according to methodologies described in the literature (Dons et al. 2012). However, data from a fix monitoring station are not available for all studied pollutants.

The calculation of the inhaled doses using VĖ values defined in literature can also be considered a limitation of this study. VĖ vary with the hearth rate of the subject but there are other variables that pose some influence such as gender, travel speed, terrain, bicycle weight, weather and subject fitness status (Bigazzi and Figliozzi 2014; Ramos et al. 2015b).

4.1.5 Conclusions

Promoting cycling instead of car driving for short trips is a policy option that meets with increased interest within Lisbon and European cities facing persistent ambient air pollution problems. A modal shift to active transportation has a great societal impact, including reduction of greenhouse gas emissions, reduction of ambient concentrations of health relevant air pollutants and increased physical activity. However, cyclists are more exposed to air contaminants from surrounding traffic, with increased VĖ resulting in greater inhaled doses of air pollutants. This study showed that car drivers and bus passengers on urban streets may be exposed to higher pollutant levels than cyclists traveling on the same streets. This is due to exhaust gases from vehicles in front that may enter into the vehicle directly through the ventilation system or from emission sources inside the vehicle. This reduced exposure of cyclists to air pollution is, however, compensated by their higher inhalation rate. Health implications from exposure to short and high peak concentrations of pollutants during cycling instead of the lower, longer peak pollution concentrations in cars and buses could be important. Considering the health, environmental and social positive impacts of active commuting, cycling should not be disregarded. This study and a broader field of research have the potential to encourage policy-makers and city planners to expand infrastructure to promote safe and healthy bicycle commuting. City planners should create bicycle lanes with less contact with motorized traffic and cyclists should be encouraged to select routes between home and work with lower traffic and to travel outside of rush hour time periods. To help commuters choose routes with low traffic exposure though the city, novel route planners should be developed or currently available route planners modified for choosing routes with the shortest distance or the shortest travel times.

5 General Discussion

This thesis addressed the exposure of persons undertaking physical exercise to air pollution and contributed to the state of the art on the scientific field of indoor and outdoor air. The main focus of this thesis was to characterize the exposure and the dose of individuals in three different situations: in fitness centers, cycling on cycle paths and during active transportation. The main factors that influence people's exposure and dose were discussed along the chapters of the thesis. This thesis starts with the characterization of indoor air in fitness centers, assessed physiological parameters of exercise practitioners' to calculate the inhaled dose during fitness classes, evaluated the use of bicycles on cycle paths for recreational activities and compared exposure to air pollutants between active transportation and other modes of transport.

5.1 Overview

Chapter 2 presented the results of an IAQ monitoring program in eleven fitness centers and the calculation of inhaled doses in this specific physical activity. In recent years, studies on IAQ were focused in homes and schools, although the work herein has, for the first time, developed information about fitness centers, contributing to the state of the art on this field. Additionally, this work presented the first approach on the estimation of inhaled doses of pollutants during indoor physical activities by calculating the $\dot{V}E$ for two fitness classes and using the data obtained during the IAQ monitoring program. The results obtained in this chapter are important for future studies in this setting.

The survey allowed for the observation of a picture about the IAQ situation in these buildings, through assessment of seven main pollutants (CO_2, CO, O_3, CH_2O, PM1, PM2.5, PM10), comfort parameters and ventilation rates. In order to obtain the daily pattern of pollutants and identify pollution sources, three of the eleven fitness centers were selected to perform a deeper assessment with longer measurement periods and more parameters, such as element identification, nanoparticle deposition in lungs and microbial characterization.

The ventilation rates were calculated based on a novel and easy method, applied for the first time in fitness centers. Most of the fitness centers assessed in this study used mechanical ventilation which should provide the minimum ventilation required by guidelines, although the results showed poor ventilation rates. This has consequences on pollutant accumulation indoors and, therefore, on practitioner exposure. With the long assessments, it was possible to identify the events/activities occurring and connect them with the pollutants. With this analysis, it was possible to observe the influence of cleaning procedures on VOC concentrations and confirm the human presence as a significant source of pollution, particularly on particle resuspension and CO_2 production. The ventilation type also showed to play an important role on IAQ due to filtration of outdoor air, especially on reducing PM10 and fungal concentrations.

After having studied the exposure indoors, it was imperative to study the exposure to air pollutants during physical activity outdoors. In this context, cycling was assessed due to its recognized benefits on transport efficiency, environment, health and fitness advantages (European Commission, 2010). Due to the harmful impact that particles have on human health, chapter 3 described the characterization of the exposure and the dose during cycling on three different cycle paths, for PM10 and PM2.5. The exposure in cycle paths has a strong influence from traffic patterns. The distance between road traffic and the cycle path is crucial to reduce personal exposure. The combination of instrumental and biomonitoring techniques in this work allowed for a better comprehension of the contributing sources. The development of an exposure visualization system using geographical information was very useful to identify the location of hotspots of pollutants' concentrations along the routes.

Chapter 4 addressed the use of bicycles instead of motorized transport modes. The exposure was higher for car and bus users than for other modes, but the dose was always higher for cyclists due to their increased $\dot{V}E$. This chapter shows that encouraging the use of bicycles in the city should be done in combination with the decrease of car use, implement policies and incentives for the renewal of the car fleet to less polluting vehicles, and the use of more environmentally efficient public transport. The creation of pollutant maps from the exposure visualization system was determinant to recognize differences on the exposure along the route for the cycling mode. The route selection for cycling should be done in order to reduce the exposure to air pollutants.

5.2 Final Remarks

This thesis approached, in an integrated way, the issues of pollutant exposure during exercise based upon three distinct contexts. Each one of the contexts revealed aspects that can be improved to enhance the benefits of physical activity. Fitness centers should be dimensioned to a specific number of persons; with these criteria, the ventilation systems can be designed according with the population inside. Overcrowded rooms increase CO_2 concentrations and particle resuspension; therefore ventilation rates are insufficient to renewal the air. The managers of fitness centers should be aware of IAQ symptoms in a building to promote a safer environment. The practitioners also have an important role in this setting. The use of exclusive indoor shoes can reduce particles and dust that are derived from outdoors. Practitioners should have an active position on the building environment and inquire the managers about the IAQ of the gymnasium, cleaning products and procedures, as well as the HVAC maintenance plan.

The implementation of cycle lanes and cycle paths in cities must be weighted with natural or artificial barriers between the path and the road to minimize the exposure of practitioners to road traffic pollutants. Urban planners have a leadership role to play in designing and maintaining the built environment in ways that promote active living (WHO, 2008). Before creating a new cycle path or a sport facility, a SWOT analysis, within the scope of exposure to pollutants and air quality, could be useful to identify the benefits and the risks of a new infrastructure.

Individuals should care about air quality before engaging in vigorous exercise, especially people with asthma or with a weakened respiratory system. PM and O_3 are concerning pollutants in outdoor air that are capable of causing airway damage. In all the situations covered in this thesis the precautionary principle should be considered.

Sport has an important role in preventing diseases, and guidelines from WHO recommend an increase in exercise and physical activity rates in Europe. One of the conclusions from this thesis is that people have to exercise in safer and cleaner spaces to potentiate the benefits of physical activity.

5.3 Future Research

Ventilation rates are clearly linked with energy consumption in buildings and air quality. The development of an integrated strategy to balance ventilation and energy use without compromise to IAQ could be very useful to fitness centers managers. The emerging pollutants such as phthalates, commonly found in vinyl floor tiles, carpet backing, paints and wall materials in gymnasiums should be considered, likewise the VOC speciation. Research on chemical emissions associated with human occupants of indoor spaces is limited. Ozone reacts rapidly with constituents of skin surface lipids on exposed skin, hair, and clothing, substantially reducing indoor ozone concentrations but increasing airborne levels of mono- and bifunctional compounds that contain carbonyl, carboxyl, or a-hydroxy ketone groups (Weschler, 2015) therefore influencing the composition of the air (Nazaroff, 2015).

Other sporting environments might be subject of study. In covered swimming pools, chlorine, widely used as water disinfectant, reacts with natural organic matter in the water (that result from perspiration, urine, hair and saliva) forming a wide range of compounds known as disinfection by-products, the most common being trihalomethanes (THMs). Taking into consideration that it may be necessary at least a period of one night for an adult to excrete the mean quantities of THMs absorbed from swimming pools (Sá et al., 2011), insufficient time between training sessions can possibly lead to a toxic build-up of THM, warranting it importance to study swimming athletes.

Outdoors, investigation in modelling air quality can be advantageous for local authorities to be able to comprehend which are the best locations to establish outdoor sport facilities and cycle paths.

List of Abbreviations

Bb – Bodybuilding room

BTEX – Benzene, Toluene, Ethylbenzene, Xylene

CFU – Colony Forming Unit

EF – Enrichment Factor

AER – Air Exchange Rates (h$^{-1)}$

GIS – Geographic Information System

GPS – Global Position System

HR – Hearth Rate (min^{-1})

HVAC – Heating, Ventilation and Air Conditioning

IAEA – International Atomic Energy Agency

IAQ – Indoor Air Quality

IARC – International Agency for Research on Cancer

INAA – Instrumental Neutron Activation Analysis

LV – Limit Value

MEA – Malt Extract Agar

NCD – Non Communicable Diseases

NIST – National Institute of Standards and Technology

OECD – Organization for Economic Co-operation and Development

PCA – Principal Component Analysis

PM – Particulate Matter

PM1 – Particulate Matter with an aerodynamic diameter of 1

PM10 – Particulate Matter with an aerodynamic diameter of 10

PM2.5 – Particulate Matter with an aerodynamic diameter of 2.5

PM2.5-10 – Particulate Matter with an aerodynamic between 2.5 and 10

PM4 – Particulate Matter with an aerodynamic diameter of 4

PNC – Particle Number Count

RPE – Perceived Exertion Rate

S1 – Studio 1

S2 – Studio 2

SBS – Sick Building Syndrome

THM – Trihalomethanes

TSA – Tryptic Soy Agar

UFP – Ultrafine Particle

VĖ – Minute Ventilation (L/min)

VOC – Volatile Organic Compounds

VR – Ventilation Rate (lps/person)

WHO – World Health Organization

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Summary

The context for this thesis is the concern that people who practice physical activity are more susceptible to air pollution. For the studies presented here, three perspectives of physical activity were considered: in indoor, i) physical activity in fitness centers; in outdoor ii) the use of bicycle in cycle paths; and iii) active transportation. Knowing the effects that air pollution has in the respiratory function, the increased VĖ (Minute Ventilation) that practitioners experience during exercise, lead to higher inhaled doses of air pollutants. The primary aim of this thesis is to provide information that is needed to deal with air pollutants and to come to better and healthier choices while practice physical activity in indoor and outdoor environments. It is clear that the sport facilities have to be correctly designed, with the aim of reduction of risk exposure and enhancing the benefits of exercise.

Chapter 1 summarizes the context for air quality and physical activity, clarifies their relations, and addresses the most relevant topics interpreted in this thesis.

Chapter 2 evaluates the exposure of sport practitioners to air pollutants and their inhaled doses. To achieve this goal, IAQ (Indoor Air Quality) parameters and VĖ were assessed in eleven fitness centers in Lisbon. After that, the IAQ in three fitness centers was monitored during one week to recognize temporal variations of pollutants and identify pollution sources. The novelty introduced in this chapter was the estimation of the inhaled doses for the typical indoor air pollutants during an aerobic and a holistic fitness class.

Chapter 3 assesses the exposure and the dose to air pollutants while using cycle paths. For this, three different cycle paths in Lisbon were chosen and two complementary techniques were used: the instrumental sampling to evaluate particles (PM: particulate matter) and biomonitoring with lichens to obtain the elemental concentrations along the cycle paths. Pollutant maps were created for particles and for the elemental concentrations in order to visualize spatial distribution of pollutants.

Chapter 4 focuses on the alternative mode of active transportation in cities. A typical route of Lisbon was selected to assess five air pollutants in five periods of the day, to observe the variation of those pollutants during the day. The exposure and the inhaled doses during the use of bicycle were compared with the use of motorized modes of transport (car, moto, bus and metro). The pollutant maps were performed for bicycle concentrations to identify the hotspots along the route.

Chapter 5 provides an overall discussion and main outcomes of the thesis.

With the integrated approach given on this thesis, new information on IAQ constrains in fitness centers, and data on $\dot{V}E$ during fitness classes was presented. New information on what contributes to personal exposure during cycling and the contrasts between travel modes in relation to active transportation during commuting are brought in this work.

Samenvatting

De context van dit proefschrift is de bezorgdheid dat mensen die aan fysieke inspanning doen gevoeliger zijn voor luchtverontreiniging. Voor de studies die hier gepresenteerd worden, zijn drie invalshoeken voor fysieke inspanning beschouwd: binnen: i) fysieke activiteit in fitness centra, buiten: ii) fietsen op fietspaden, en iii) gemotoriseerde vormen van transport. Wetende wat de effecten zijn van luchtverontreiniging t.a.v. het funktioneren van het ademhalingssysteem, zal de verhoogde ademhalingssnelheid (VE) van mensen die aan oefeningen doen leiden tot een hogere geïnhaleerde dosis aan luchtverontreinigingen.

Het hoofddoel van dit proefschrift is om informatie te verschaffen die nodig is om om te gaan met luchtverontreiniging en om tot betere en meer gezonde keuzes te komen ten aanzien van fysieke activiteit in de indoor en outdoor omgeving. Het mag duidelijk zijn dat sportfaciliteiten op de juiste wijze ontworpen moeten worden met als doel het blootstellingsrisico te verkleinen en het positief effect van het doen van oefeningen te vergroten.

Hoofdstuk 1 geeft een samenvatting van de context t.a.v. luchtkwaliteit en fysieke inspanning, verklaard hun relaties, en beschouwd de meest relevante onderwerpen die in het proefschrift verder geïnterpreteerd worden.

In Hoofdstuk 2 worden de blootstelling van sportbeoefenaars en hun geïnhaleerde doses geëvalueerd . Om hiertoe te kunnen komen werden IAQ (indoor Air Quality) en VE parameters vastgesteld in elf fitness centra in Lissabon. Daarna werden de IAQ's in drie fitness centra gemonitord gedurende een week, om tijdsafhankelijke variaties te herkennen, en om tot identificatie van verontreinigingsbronnen te kunnen komen. Een nieuwe aanpak geïntroduceerd in dit hoofdstuk was de schatting van de geïnhaleerde dosis voor de typische indoor verontreinigingen gedurende een aerobic- en een volledige fitness oefening.

In Hoofdstuk 3 worden de blootstelling en de dosis aan luchtverontreinigingen gemeten van fietsers die fietspaden gebruiken. Hiertoe werden drie verschillende fietspaden in Lissabon benut, en werden twee complementaire technieken gebruikt: instrumentele bemonstering, en biomonitoring met behulp van korstmossen, om tot elementconcentraties langs de fietspaden te komen. Er werden kaarten van verontreinigingen gemaakt voor deeltjes (PM: particulate matter) en voor elementen (hun concentraties) om de ruimtelijke verdeling van verontreinigingen te visualiseren.

Hoofdstuk 4 richt zich op alternatieve vormen van transport in de stad. Er werd een typische route in Lissabon geselecteerd waarbij vijf luchtverontreinigingen werden gemeten, over vijf dagelijkse perioden, dit om de dagelijkse variaties te kunnen herkennen. De blootstelling en de geïnhaleerde dosis van fietsers werden vergeleken met die via het gebruik van gemotoriseerde vormen van transport (auto, motorfiets, bus en metro). Kaarten van verontreinigingen werden opgesteld voor fietsers om hotspots langs de route te identificeren.

Hoofdstuk 5 geeft een algemene discussie van de resultaten en van de voornaamste bevindingen van het proefschrift.

Op basis van de geïntegreerde benadering die gevolgd is in het proefschrift, kon nieuwe informatie t.a.v. IAQ restricties in fitness centra, en gegevens t.a.v. VE gedurende fitness oefeningen worden gepresenteerd. Nieuwe gegevens konden worden toegevoegd aan wat bekend is t.a.v. individuele blootstelling tijdens het fietsen en de verschillen tussen de diverse vormen van transport in woon-werk verkeer.

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Curriculum Vitae

Carla Alexandra Almeida Martins Cortês Ramos was born on the 9th January 1988 in Lisbon, Portugal. She holds a degree on Environmental Health from Escola Superior de Tecnologia da Saúde de Lisboa (2010). During her academic course, she held two academic internships: one on the Public Health area and the other at the Technological and Nuclear Institute (now IST/CTN) where she developed scientific work (PM*fugitive* project), under the supervision of Dr Marta Almeida.

After that, she worked as Consultant of Health and Safety at Work (2010-2011) in a private company. In 2011 she won a PhD fellowship funded by FCT. In 2013 Carla finished the MSc in Integrated Management on Quality, Environment and Safety from Instituto Superior de Educação e Ciências.

Since 2011, she develops her PhD work in Instituto Superior Técnico by TU Delft. Her main research interests are environmental health, personal exposure to air pollution and indoor air pollution. During the last years she collaborate in different research projects such as EFICARE project (Monitoring Model of Maintenance Management, Energy Efficiency and Indoor Air Quality), an IAEA project (Air Quality Management RER/1/013), and laboratorial intercomparisons for IAEA.

C.A. Ramos is author of 9 articles in international peer-reviewed journals (4 as first author). She participated in 10 national and international conferences where she presented 6 oral presentations, 7 posters (6 as first author) and four abstracts in conference proceedings (as first author). She has a total of 34 citations and an H-index of 4.

Her main research area was the exposure to air pollutants during exercise. The results of her research are presented in this book.

List of publications

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