MULTIPLE ECOSYSTEM SERVICES PROVIDED BY URBAN FORESTS IN THE URBAN AREA OF MILAN

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The future is not somewhere we are going, it is something we are making.

Richard Hobbs, 1997
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**Abstract**

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### GENERAL CONCLUSIONS

**REFERENCES**

**SUPPLEMENTARY MATERIALS**

- Urban forests and biodiversity
- The soundscape methodology for bird monitoring in urban parks

**PAPERS AND CONFERENCE PROCEEDINGS**

**AKNOWLEDGMENTS**
Cities, just as any other social-ecological system, depend on ecosystems and their components to sustain long-term conditions for life. The provision of ecosystem services (ES) varies spatially across landscapes, determined by diverse human social, political and ecological interactions and it involves the analysis of the environment in an interdisciplinary perspective based on the integration of ecological and socio-economic dimensions. A growing empirical base confirms that urbanization profoundly affects how we connect with and use natural resources but how these impacts play out, in particular with reference to ecosystem functioning and biodiversity, is not yet well understood. Quantification of ES can be difficult in areas containing complex land-cover mosaics, and that often represent novel ecosystems in terms of their composition. However, it has been extensively demonstrated that urban areas may be important source of ES. Because of their importance, a deeper knowledge about ES provisioning in urban areas is strongly advocated. In this context, particular importance is emphasized for urban parks and new forest plantings in urban, peri-urban or urbanized contexts. This study represented a multi-disciplinary research that focused on the analysis of multiple ecosystem services in the urban area of Milan (Italy) with particular regard to those provided in urban parks and forests. Five specific studies had been developed in order to analyse different ecosystem services related to biodiversity, pollution mitigation, organic carbon storage by soils, and cultural services.

The first study proposed a methodology to spatially define the extent of the urban region (UR) of Milan. Milan UR area was morphologically characterized by a northern band of natural and semi-natural lands which was separated from the southern agricultural lands by a semi-continuous urban ‘belt’. In this region, the expansion of the urban tissue was analysed by quantifying the current degree of landscape fragmentation and characterizing historical trends (in the past 50-years) of urban sprawl in the UR. The fragmentation was computed with a metric that produced a map of the spatial distribution of fragmentation, as well as quantitative data on the level of fragmentation present in different planning areas relevant to planners. Results revealed that the UR of Milan suffers of high degree of landscape fragmentation throughout the area, with the major fragmented territories in the north of the city Milan (Monza and Varese). Urban sprawl showed an increasing trend in the last 50 years, reaching alarming values to date. Overall, Milan UR showed a chaotic proliferation of dispersed buildings and under-utilized infrastructure that lead to increase in energy requirements and resources depletion. A balance in urban compaction and maintenance of green spaces is a major challenge for urban planners.

In the second study the role of urban forests in providing suitable habitats for biodiversity was analysed. In order to detect how environmental features of the urban area of Milan affected biodiversity, landscape features, site features and vegetation structure were analysed with a multi-scale analysis. For twenty-eight birds species
was described the different response (presence) to specific environmental features. The study showed how the biodiversity patterns found can be explained by considering the effects of environmental features acting at different scales and that urban green areas can therefore be actively managed by foresters and city planners to preserve the biological diversity that they harbour.

The third study dealt with the regulative role of vegetation in urban areas in contributing to reduce air pollution levels and offset greenhouse gas emissions. This study represents a preliminary work that aimed at analyse the hydrocarbon-degrading potential of epiphytic microbial communities on *Platanus x acerifolia* trees commonly used in an urban area. Further investigations are needed in order to quantify the actual contribution of bacteria in air pollutant removal per unit of leaf weight or leaf area under different environmental conditions, and at the evaluation of the efficiency of different plant-bacteria systems in air quality improvement.

The fourth study dealt with the estimation of organic carbon (OC) storage by urban soils of parks, forests and other typology of urban land covers. As urban soils are normally excluded from traditional OC stock estimation because of the misleading assumption that they do not store C, this study aims to investigate urban soils characteristics and compare C stock of different type of urban land cover and use. Mean OC stock found for the urban soils of Milan was comparable to values found in the Lombardy region by previous studies. OC stock was found higher for parks compared to other areas, but no significant differentiation was found in order to define a typical C stock for each land cover or use type. Results highlight how the complex heterogeneity of urban soils together with the uncertainty of their origins, their history and management may summarize the reasons of the limitation in our capacity to model their characteristics.

The fifth study analysed cultural ecosystem services (CES) perceived by citizen in one of the larger park of the study area (Parco Nord). The study aimed to present a methodology to integrate CES mapping with decision support in land use planning. Public participatory GIS techniques (PPGIS) were used to map people perception of cultural benefits. A comparison analysis with cultural benefits perceived by people and those intended to be provided by park management was performed. Results showed places in the park that had bundles of different values, and other places with clusters of the same value type. Comparison analysis revealed hotspot and coldspot of CES in terms of richness. Methodological opportunity and needs of the methodology used were discussed in the light of better integration between CES mapping and decision support in land use planning.

The research investigated multiple ecosystem services provided by urban parks and forests and it addressed some limitations in the current ES assessment and ES implementation into urban planning. To ensure the delivery of urban ES we need heterogeneous, multifunctional and accessible green spaces throughout our cities. As are both cultural and biological diversities that underpin resilience and sustainability, the integration of multiple ecosystem services assessment and their management would help in reduce the ecological footprint of cities while enhancing resilience and quality of life of their inhabitants.
GENERAL INTRODUCTION

THE MILLENNIUM ECOSYSTEM ASSESSMENT
(from MA, 2005)

The Millennium Ecosystem Assessment (MA) is a comprehensive scientific study launched by the United Nation Secretary General Kofi Annan in 2000 in order to assess the human impact on the environment, to better understand the consequences of current changes to ecosystems and to evaluate scenarios for the future. The work emphasize the linkages between ecosystems and human well-being and, in particular, on ecosystem services. The human species, while buffered against environmental changes by culture and technology, is fundamentally dependent on the flow of ecosystem services. MA identify four main findings to be addressed and the actions needed to enhance the conservation and sustainable use of ecosystems.

The first finding claims that over the past 50 years, humans have changed the structure and functioning of ecosystems more rapidly and extensively than in any comparable period of time in human history. Most of these changes to ecosystems have been made to meet a dramatic growth in the demand for food, water, timber, fiber, and fuel. Between 1960 and 2000, the demand for ecosystem services grew significantly as world population doubled to 6 billion people and the global economy increased more than sixfold. To meet this demand, food production increased by roughly two-and-a-half times, water use doubled, wood harvests for pulp and paper production tripled, installed hydropower capacity doubled, and timber production increased by more than half. The growing demand for these ecosystem services was met both by consuming an increasing fraction of the available supply (for example, diverting more water for irrigation or capturing more fish from the sea) and by raising the production of some services, such as crops and livestock. The latter has been
accomplished with the use of new technologies (such as new crop varieties, fertilization, and irrigation) as well as through increasing the area managed for the services in the case of crop and livestock production and aquaculture. Moreover, these changes resulted in a substantial and largely irreversible loss in the diversity of life on Earth: the number of species and the genetic diversity are declining globally and the distribution of species is becoming more homogenous across the globe.

The second finding highlights that the changes that have been made to ecosystems have contributed (in the aggregate, and for most countries) to substantial net gains in human well-being and economic development. Agriculture, including fisheries and forestry, has been the mainstay of strategies for the development of countries for centuries, providing revenues that have enabled investments in industrialization and poverty alleviation. However, these gains have been achieved at growing costs in the form of the degradation of many ecosystem services, increased risks of non-linear changes in ecosystems (i.e. disease emergence, abrupt alterations in water quality or the collapse of fisheries).

Approximately 60% (15 out of 24) of the ecosystem services evaluated in the MA assessment (including 70% of regulating and cultural services) are being degraded or used unsustainably. Moreover, despite the progress achieved in increasing the production and use of some ecosystem services, we have witnessed at the exacerbation of poverty for some people and to growing inequities and disparities among groups of people. The degradation of ecosystem services is harming many of the world’s poorest people and is sometimes the principal factor causing poverty.

The third finding deals with the development of four scenarios to explore plausible futures for ecosystems and human well-being. Under all four MA scenarios, the projected changes in drivers result in significant growth in consumption of ecosystem services, continued loss of biodiversity, and further degradation of some ecosystem services. The eight Millennium Development Goals (MDGs) adopted by the United Nations in 2000 aim to improve human well-being by reducing poverty, hunger, child and maternal mortality, by ensuring education for all, by controlling and managing diseases, by tackling gender disparity, by ensuring environmental sustainability, and by pursuing global partnerships. Although socio-economic policy changes will play a primary role in achieving
most of the MDGs, degradation of ecosystem services poses a significant barrier, and many of the targets are unlikely to be achieved without significant improvement in the management of ecosystems.

The **four finding** says that the challenge of reversing the degradation of ecosystems while meeting increasing demands for their services can be partially met under some scenarios that the MA has considered (three of the four). These involve significant changes in policies, institutions, and practices that can mitigate many of the negative consequences of growing pressures on ecosystems, although the changes required are large and not currently under way. Many options exist to conserve or enhance specific ecosystem services in ways that reduce negative trade-offs or that provide positive synergies with other ecosystem services. Ecosystem degradation can rarely be reversed without actions that address the negative effects or enhance the positive effects of one or more of the five indirect drivers of change: population change (including growth and migration), change in economic activity (including economic growth, disparities in wealth, and trade patterns), socio-political factors (including factors ranging from the presence of conflict to public participation in decision-making), cultural factors, and technological change.

To summarize, the MA provide a state-of-the-art scientific appraisal of the condition and trends in the world’s ecosystems and the services they provide and the options to restore, conserve or enhance the sustainable use of ecosystems. By assessing current knowledge, scientific literature and data, the MA at the most basic level, synthesize information that has previously been available, and does not conduct new research. However, the relevant findings reported the assessment can only be reached when a large body of existing information is examined together. In addition, several aspects of the MA do represent important new contributions. One of the unique contributions is the focus on ecosystem services, the development needs and their link to human well-being. In fact, by examining the environment through the framework of ecosystem services, it becomes much easier to identify how changes in ecosystems influence human well-being and to provide information in a form that
decision-makers can weigh alongside other social and economic information. Importantly, MA also identified major gaps in our current knowledge. One of this gap is inherent with the relatively limited information that exists about the status of many ecosystem services at a local and national scale. At the same way, basic global data on the extent and trends in different types of ecosystems and land use are surprisingly scarce. The MA, similarly to the IPCC, primarily synthesized the findings of existing research, and make them available in a form that is relevant to current policy questions. The overall aims of the MA were to contribute to improved decision-making concerning ecosystem management and human well-being, and to build capacity for scientific assessments of this kind.

ECOSYSTEM SERVICES

The ecosystem services (ES) framework highlights the long-term role that healthy ecosystems play in the sustainable provision of human well-being and economic development across the globe (Costanza and Dely, 1992). There has been growing international recognition that the contribution that nature makes to human well-being is often not adequately valued or integrated in decision-making, and that ecosystem services are being eroded as a result (MA, 2005), with considerable cost to societies (Kumar, 2010). The ecosystem services concept is an increasingly useful way to highlight, measure, and value the degree of interdependence between humans and the rest of nature. Although the MA emphasizes the linkages between ecosystems and human well-being, it recognizes that the actions people take that influence ecosystems result not just from concern about human well-being but also from considerations of the intrinsic value of species and ecosystems. The ES approach is complementary with other approaches to nature conservation, but provides conceptual and empirical tools that the others lack and it communicates with different audiences for different purposes. Ecosystem services can be grouped in four major categories according to the Millennium Ecosystem Assessment (MA, 2005) and The Economics of Ecosystem Services and Biodiversity (TEEB, 2010): provisioning, regulating, supporting or habitat, and cultural services.
Provisioning services are ecosystem services that describe the material or energy outputs from ecosystems. They include food, water, raw materials, medicinal and other resources. Regulating services are the services that ecosystems provide by acting as regulators. It includes for example the regulation of local climate and air quality, carbon sequestration and storage, moderation of extreme events, wastewater treatment, erosion prevention and maintenance of soil fertility, pollination and biological control. Supporting or habitat services are those services that are necessary for the production of all other ecosystem services including soil formation, photosynthesis, primary production, nutrient cycling and water cycling and the habitats provided for species as well as the maintenance of genetic diversity. Cultural services include the non-material benefits that people obtain from ecosystems such as spiritual enrichment, intellectual development, recreation and aesthetic values.

Figure 1. Strength of linkages between categories of ecosystem services and components of human well-being and indications of the extent to which it is possible for socio-economic factors to mediate the linkage (from MA, 2005).
ECOSYSTEM SERVICES AND URBAN AREAS

Ecosystem service provision varies spatially across landscapes, determined by diverse human social, political and ecological interactions (Peh et al., 2013). Currently, we are entering an urban era (Seto and Reenberg 2014), and 75% of the world population is projected to live in cities and their peri-urban surroundings in 2050 (UN, 2013). In Europe, almost 80% of the people already live in cities and urban areas, and there is no sign that this urban trend will abate (Haase et al., 2014). The continuous growth in the number and size of urban areas along with an increasing demand on resources and energy, poses great challenges for ensuring human welfare in cities while preventing an increasing loss of soil, habitats, resources, and biodiversity (Haase et al., 2013). This massive urbanization will likely have significant effects on the natural environment and the services it can supply to humanity (Bolund and Hunhammar, 1999; Forman, 2008; McDonald 2008), both directly through the expansion of urban area and indirectly through changes in consumption and pollution as people migrate into cities (McKinney, 2002; Liu et al., 2003; McGranahan and Satterthwaite, 2002).

One of the challenges to ecosystem functionality and the provision of ecosystem services is the increasing fragmentation of landscapes and ecosystems (Lafortezza et al., 2010). Mounting levels of urbanization and transport infrastructure have created a network of barriers that results in a patchwork of land uses and isolate open space areas with the consequence that natural ecosystems have become scattered across the landscape and displaced by new land-use developments (Scolozzi et al., 2012; Geneletti, 2004; Lafortezza and Brown, 2004). In this context, new forest resources arising from an artificial implant or secondary succession due to cultural abandonment have grown in importance in urban and suburban areas (Hladnik and Pirnat, 2011).

Urbanization is a complex social, economical, political, and technological process, and there are no uniform patterns of urbanization. Urbanization manifests itself primarily in creating urban landscapes with densification, expansion/sprawling, and shrinkage patterns. The way these patterns emerge and their impact on land and the environment require new methods and new approaches to be investigated.
Gene
tral Introduction

(Haase et al., 2013). In this social-ecological systems there is a need to understand the synergies, interdependencies and trade-offs between society and ecosystems (Haase et al., 2014). In fact, it is in cities where a social-ecological co-production of ecosystem services (ES) and society might open new ways for ensuring resilience and liveability (Gomez-Baggethun et al. 2013). A growing empirical base confirms that urbanization profoundly affects how we connect with and use natural resources (Haase et al., 2014). How these impacts play out, in particular with reference to ecosystem functioning and biodiversity, is not yet well understood (Elmqvist et al., 2013; Haase et al., 2012). Mismatches between spatial and temporal scales of ecological processes and patterns on the one hand, and social scales of use, monitoring and decision-making on the other, have in the past not only limited our understanding of ecological processes in urban landscapes, but have also limited the integration of urban ecological knowledge into urban planning (Kabisch and Haase 2014).

Existing at the nexus of high population density and high consumption, urban areas are typically thought of in the context of reducing resource demand rather than producing ecosystem services (ES) (Ziter, 2015). Quantification of ES can be difficult in areas containing complex land-cover mosaics, and that often represent “novel ecosystems” in terms of their composition (Wu, 2014). However, it has been extensively demonstrated that urban areas may be source of ES (reference). For example, some cities have been estimated to store the equivalent carbon per unit area as tropical forests (Churkina et al., 2010). Urban environments have many distinctive features, the most prominent of which is their extreme heterogeneity: there are patches where both biodiversity and ecosystem service delivery is minimal, for example, where land surfaces are covered with concrete or tarmac, and others where biodiversity and other ecosystem services may be very high, as in some gardens and parks (Hester and Harrison, 2010). Urban green spaces (urban parks, forests, gardens, playing fields,…) provide a wide range of ecosystem services and are of strategic importance in order to deal with problems arising at city scale and to mitigate the impact of human activities. A large number of ecosystem services provided by urban areas result from the presence of green infrastructures (McPhearson, 2011): residential gardens, urban farms, and green-roofs support provisioning services;
climate and air quality regulation in urban areas rely to a certain extent on the presence of vegetation (i.e., moderation of the urban heat island effect, reduction of atmospheric greenhouse gas, and particulate matter concentrations); pervious surfaces and the presence of a vegetation cover allow infiltration and decreases the quantity of available water reducing risks of runoff and flooding; and green infrastructure may attenuate noise as a major environmental issue in urban areas (Morel et al., 2015). Green urban areas may also be reservoirs for native and rare species and thus contributing to biodiversity maintenance.

**VALUATION OF URBAN ECOSYSTEM SERVICES**
(from Gómez-Baggethun et al., 2013)

Valuation of ecosystem services involves dealing with multiple, and often conflicting value dimensions (Chan et al., 2012a; Martín-López et al., 2013). Different value dimensions (biophysical, economic and socio-cultural) are identified. Biophysical measures and indicators allow us to quantify some ecosystem service performance, but the difficulty of measuring increases as the focus shifts from provisioning, to regulating to habitat, to cultural services (Gómez-Baggethun et al., 2013b). These quantifications pass through direct measures of ES (i.e. tons of carbon sequestered per hectare per year, tons of food per hectare per year) or using proxies and indicators (i.e. Leaf Area Index, species diversity and abundance) (Gómez-Baggethun et al., 2013b). Conventional economic valuations are restricted to priced goods and services, which represent only a limited subset of ecosystem services (i.e., those which are exchanged in markets). Economic valuation of ecosystem services attempts to make visible the ‘hidden’ economic costs from the conversion of ecological infrastructure to built infrastructure (or from natural capital to human-made capital). Over the last few decades, a range of methods have been developed to calculate economic costs resulting from loss of ecological infrastructure. In particular, meta-analyses on economic valuations of ecosystem services show that hedonic pricing and stated preference methods have been the methods most
frequently used in urban contexts (Boyer and Polasky 2004; Tyrväinen et al., 2005; Costanza et al., 2006a; Kroll and Cray, 2010; Sander et al., 2010; Brander and Koetse, 2011). Social and cultural values broadly refer to various material, moral, spiritual, aesthetic, and other values people attach to the urban environment. These include emotional, affective and symbolic views attached to urban nature that in most cases cannot be adequately captured by commodity metaphors and monetary metrics (Norton and Hannon 1997; Martinez Alier et al. 1998; Gómez-Baggethun and Ruiz- Pérez 2011; Daniel et al., 2012). Social and cultural values are included in all prominent ecosystem service typologies (Daily et al. 1997; de Groot et al. 2002; MA, 2005). Yet, compared with economic and biophysical values, social, cultural, and other non-material values of ecosystems and biodiversity have generally been neglected in much of the ecosystem services literature. Despite some cases tools have been developed to measure these values using constructed scales, in other cases translating cultural values into quantitative metrics may be too difficult or produce results that are nonsensical or meaningless.

Economic and non-economic valuation (that refer to biophysical measures and social and cultural values) of ecosystem services is often demanded by policy makers and practitioners as supporting information to guide decisions in urban planning and governance. Despite biophysical measures of ecosystems services are often presented as a prerequisite for sound economic valuations, these measures themselves often provide powerful information to guide urban planning (Gómez-Baggethun et al., 2013b). Importantly, using combinations of valuation methods is often necessary to address multiple ecosystem services (Boyer and Polasky 2004; Costanza et al. 2006a; Escobedo et al., 2011).

The choice of valuation methods is determined by factors including the scale and resolution of the policy to be evaluated, the constituencies that can be contacted to obtain data, and supporting data constraints, all subject to a study budget.

City managers are concerned with the creation of suitable conditions for the cities of tomorrow, and the future of urbanization is often qualified as “sustainable,” “resilient,” “self-sufficient,” “biophillic,” and/or “adapted to global change” (Morel et al., 2015). Such ambitious goals rely on an
integrated management of the urban ecosystem, taking into account every available local resource, and the management of ecosystem services in the city is regarded as a response to global change and to other the main challenges (Reeve, 2012). However, a sustainable city is more than ecologically resilient (Buchel and Frantzeskaki, 2015). As Chiesura (2004) reviewed, green spaces in an urban setting increase the well-being and quality of life of its inhabitants in many ways, for example by reducing stress and providing a sense of tranquillity and health. Thus, the use of the concept of urban ecosystem services can play a critical role in reconnecting cities to the biosphere, and reducing the ecological footprint and ecological debt of cities while enhancing resilience, health, and quality of life of their inhabitants (Gómez-Baggethun et al., 2013).

PROJECT AIMS AND FRAMEWORK

As the forests in urban and peri-urban areas play a key role in providing ES and thus environmental mitigation, a better knowledge about ES provision is of fundamental importance to enhance management of ES and cities resilience. This research project focused on the analysis of multiple ecosystem services (ES) in the urban area of Milan (Italy) with particular regard to those provided in urban parks and forests. It aimed to quantify ES with reference to the current situation of urban land use and to provide information to support the development of management options for the storage of OC in soils, but also for support the protection of biodiversity and cultural services. Fifteen urban and peri-urban parks in Milan and surrounding areas were selected and biophysical (biodiversity and carbon storage) and socio-cultural (cultural benefits) valuation of ecosystem services were developed for each case-study.
In the first study (Chapter I), I focused on the definition of the urban region of Milan by proposing a standardized and repeatable methodology that utilize easy-retrievable data. In the area, landscape fragmentation and dispersion of urban tissue (sprawl) were analysed and mapped using quantitative metrics. In order to analyse different ecosystem services provided by urban parks and forests in the study area, four specific studies had been developed (Figure 2). Chapter II focused on habitat provisioning for biodiversity in urban parks and forests and analysed which environmental features of the urban environment affected biodiversity patterns at different scales and how vegetation structure influenced the presence of different birds species. Chapter III investigated the regulative role that vegetation play in urban areas by analysing the potential of tree leaf bacterial communities in air pollutant removal. This study was not included in a first version of the project, but was developed during the PhD as a results of a fruitful collaboration with the laboratory of Microbiology of our Department. My contribution in this study was to select the appropriate tree species for the urban area of Milan and to design and conduct the sampling on Platanus x acerifolia leaves. Chapter IV deal with the estimation of organic carbon (OC) storage by urban soils of parks, forests and other typology of urban land covers. As urban soils are normally excluded from traditional OC stock estimation because of the misleading assumption that they do not store C, this study aims to
investigate urban soils characteristics and compare OC stock of different type of urban land cover and use. Chapter V analysed cultural ecosystem services (CES) perceived by people in one of the larger park of the study area (Parco Nord). The study aimed to present a methodology to integrate CES mapping with decision support in land use planning with the use of public participatory GIS techniques (PPGIS) and comparison analysis.

This project represents a multi-disciplinary research that has the main aim to improve our knowledge about ecosystem services provided within urban areas and to address some limitations in the current ES assessment and ES implementation for a more sustainable urban planning.

Figure 3. Urban and peri-urban parks selected in the study area.
Chapter I

Landscape fragmentation and urban sprawl in the urban region of Milan

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Abstract

Spatial expansion of cities appears to be unavoidable. Despite concern around landscape alterations caused by urban expansion, the understanding of the amounts and patterns of this phenomenon is sorely lacking. In this study, we quantified the current pattern of landscape fragmentation in the urban region (UR) of Milan and we analysed the trend of urban sprawl over more than 50 years. The UR of Milan was spatially defined using a standardized and repeatable methodology that combine land-use and population density data. Fragmentation was assessed using the effective mesh size ($m_{eff}$) and results allowed us to obtain a map of the current degree of landscape fragmentation in the UR. The trend of sprawl was monitored between 1954 and 2012 and over different areas in order to detect the magnitude (amount and direction) and patterns of changes. Results revealed a positive trend and a high degree of sprawl over the whole study area.

1. Introduction

Urbanization is fast-growing worldwide. Because the population living in cities is expected to increase in the next decades (UN, 2014), urban expansion appears to be an unavoidable fact (Inostroza, Baur, & Csaplovics, 2013). Spatial expansion of cities is often accompanied by processes such as urban sprawl and fragmentation. The former is considered a serious urban development issue. It is characterized by low urban densities accompanied by expansion of urban space and inefficient resource utilization (Bhatta, Saraswati, & Bandyopadhyay, 2010; Travisi, Camagni, & Nijkamp, 2010). Sprawling cities (in opposition to compact cities) consume extended lands by dispersed expansion, leading to inefficient urban development (Inostroza et al., 2013; Jaeger & Madrinan, 2011). One of the direct implications of this phenomenon is the overconsumption of land, with loss of natural and agricultural areas and the increase in built-up and paved surfaces (Sudhira &
It is common assumption that dispersed patterns of urban expansion contribute to increasing the degree of fragmentation of extended territories (Selva et al., 2011). However, the relationship between urban sprawl and landscape fragmentation resulted to be non-stationary, to be influenced by the scale of analysis and to vary considerably between modelling techniques (Torres et al., 2016). Landscape fragmentation ‘is a process that results in the disruption of existing ecological connections between spatially separated elements of landscapes’ (Forman, 1995; Haber, 1993). This leads in turn to critical impoverishment of landscape ecological functionality. Fragmentation operated by urbanization leads to the formation of isolated habitat patches embedded in a matrix of urban tissue, with ecological interactions among them that are highly altered (Forman, 1995; Harris, 1984; Saunders, Hobbs, & Margules, 1991). The degree of fragmentation is a useful indicator for evaluating landscape alterations caused by urban expansion and thus it provides an indication of the sustainability of land use changes (Wade, Riitters, Wickham, & Jones, 2003). The concern around landscape alterations caused by urban processes is due to the implications they have in compromising sustainable urban development. Disturbances caused by human activities and its deleterious effects on biodiversity and on the provision of key ecosystem services may diminish the resilience of ecosystems as for example their resistance to disturbances or their recovery after perturbations. Despite recent studies have addressed the issue (Nazarnia, Schwick & Jaeger, 2016; Inostroza et al., 2013; Angel, Parent, Civco & Blei, 2010; Kasanko, Barredo, Lavalle, McCormick, Demicheli, Sagris, Brezger, 2006), a full understanding of the amounts and patterns of urban spatial expansion is still lacking (Schneider & Woodcock, 2008).

Recent patterns of urban development of many large metropolitan areas has posed the problem of redefining the scale at which urban systems are analysed and, in this respect, an ecological regional perspective is needed (Forman, 2008). To analyse problems arising from urban development and offer solutions that last, a city’s urban region is a useful scale on which to work (Forman, 2008). The aims of this work were i) to define the spatial extent to which to analyse urban sprawl and landscape fragmentation by mapping the urban region (UR) of Milan, ii) to assess the current degree of
landscape fragmentation in the UR and iii) to monitor trends of urban sprawl over time and over different areas within the study area in order to detect the magnitude (amount and direction) and patterns of changes. For these purposes, we first mapped the UR of Milan by implement a standardized methodology that can be easily repeatable. We then quantified landscape fragmentation using a spatially explicit metric and we produced a map of the current degree of fragmentation of the UR. Finally, we quantified urban sprawl over different areas (region, sub-areas and transects) in the study area and we described its trends over a period of more than 50 years (from 1954 to 2012). The results of this work allowed us to evaluate the degree of urban disturbance at landscape scale in the area of Milan (Figure 1), which is Italy’s most extended metropolitan area. The understanding of urban development patterns at the urban region scale is informative for urban planning in implementing adequate spatial strategies for a sustainable urban development (Inostroza et al., 2013).

Figure 1. Study area: Lombardy is the administrative region where the city of Milan and its district are located (urban region is not represented here).
1.1 Measuring landscape fragmentation and urban sprawl

Generally speaking, an urban region can be described as a widespread geographical area with a major city linked to surrounding lands and minor satellite centres (Forman, 2008). It is therefore a distinctive and increasingly important type of region and the importance of focusing attention on this scale rather than a city scale is continuously underscored by a series of leaders (Bullard, Johnson, & Torres, 2000; Dreier, Mollenkopf, & Swanstrom, 2004; Forman, 2008; Hall, 2014; Ravetz, 2000; Soja, 2000). Some urban areas have evolved from monocentric agglomerations to more complex systems composed of integrated urban centres (cores) and sub-centres (Brezzi, Piacentini, Rosina, & Sanchez-Serra, 2012). This changing spatial organisation of cities and surrounding territories alter landscapes and lead to profound land-use and land-cover transformations. Land use and land cover maps play an important role in monitoring and modelling land changes within and around urban areas, (Herold, Goldstein, & Clarke, 2003; Howarth & Boasson, 1983) and they can reveal explicit patterns of land cover and land use changes (Jensen & Cowen, 1999) as well as helping in understanding patterns of urban expansion. The computation of indices to measure fragmentation and sprawl help in understanding the process of urbanisation at a landscape level (H. S. Sudhira, Ramachandra, & Jagadish, 2004). The effective mesh size ($m_{eff}$) is a suitable and powerful indicator of landscape fragmentation introduced by Jaeger (2000) and later improved by Moser et al. (2007). It evaluates the possible connectivity between areas, which is the probability that two randomly chosen points in a region will be found in the same undissected area (J. A G Jaeger, 2000). In this study, we chose to use $m_{eff}$ among other methods because it has some advantages such as its mathematical simplicity and intuitive interpretation as well as being an area-proportionately additive (that means that it is independent of the size of the region investigated). Urban sprawl can be assessed using Shannon’s entropy (Yeh & Li, 2001). Shannon’s entropy is perhaps the most widely used technique to measure the extent of urban sprawl with the integration of GIS (Bhatta, 2009; H. S. Sudhira, Ramachandra, & Jagadish, 2004; Yeh & Li, 2001). Entropy can be used to measure the degree of spatial concentration or dispersion of a geographical variable ($x_i$) among $n$ zones. Furthermore, the changes in entropy can be used to identify whether land...
development follows a dispersed (sprawled) or compact pattern of urban growth. Bhatta (2010) presented an interesting review of the methods used to evaluate urban sprawl and highlighted the concern about the lack of commonly accepted and persuasive metrics to measure it. Here, we measured sprawl using Shannon’s entropy because it is a more robust spatial statistic than others (Yeh & Li, 2001) and its common use makes it a suitable metric for comparisons. Differently from traditional spatial dispersal statistics, entropy value is invariant with the value of n (number of regions), although it is still to some extent sensitive to variations in the shapes and sizes of the regions used for collecting the observed proportions of the phenomenon investigated (which in this study is the proportion of built-up surface).

2. Materials and methods

2.1 Defining the urban region of Milan

Administrative definitions are widely used in spatial analysis, but they introduce distortions and are not directly comparable for different cities and countries (Inostroza et al., 2013). We decided to use a spatial approach that goes beyond the administrative boundaries to define the geographical extension of Milan’s urban region. Because a standard methodology to define an urban region spatially does not exist or is expert-based (Forman, 2008) we decided to delimit the urban region of Milan (from here also referred to as UR) by combining data on built-up surface and population density. Our purpose was to identify the area to investigate urban sprawl and landscape fragmentation in more detail. The methodology we followed in defining the UR is schematically represented in Figure 2. The mapping was performed using ArcGis software (version 10.0, ESRI Inc.). As input information, we used a land-use map and data on resident population. Data on resident population were derived from the census made by the National Institute of Statistics (ISTAT, 2011) and were available at municipality level (number of municipalities = 1546). The cartographic database was based on DUSAF (Land Use of Agricultural and Forest Land), a project funded by the Lombardy
Region (the region where the city of Milan is located, Figure 1) and carried out by the Regional Authority for Services to Agriculture and Forests (ERSAF) with the cooperation of the Regional Agency for the Protection of the Environment. The land use and cover data from the DUSAF project are shared in the context of the Lombardy Infrastructure for Territorial Information (IIT) through the Regional Geoportal (www.cartografia.regione.lombardia.it). The cartography is based on the photointerpretation of aerial photos taken in 1954-1955 (flight GAI made by Italian Aerial Group), in 1980-1982 (flight Tem1 made by Lombardy Region) and 1998-1999 (Flight IT2000 made by Blom CGR). In a subsequent version, the IT2007 orthophotos (taken by Blom CGR) and the AGEA 2012 orthophotos (taken by AGEA) were used and all the regional territory was updated as a result. In the updating process, the original geometrical structures were maintained if no changes in land use were mapped and the polygons and polylines were modified directly on the land use thematic layer of the previous version (Pileri, 2012). Furthermore, the database has evolved from the level obtained only by photointerpretation to a true “Information System” (with ancillary data) built by supplementing photointerpretation with data derived from the numerous databases and territorial projects developed regionally (Pileri, 2012). The database is periodically updated and it is currently available for five temporal periods: 1954, 1980, 2000, 2005-2007 and 2012. Each version of the databases is made up of a polygonal-type information layer representing the land use and cover and a linear-type one for hedge-rows and tree-rows. The detail level is equal to an information scale of 1:10,000 with a minimum mapping unit size of 0.16 hectares for the polygonal elements and greater than 40 m for the representation of linear elements. Only the land use and land cover map of the 1980 presents a lower level of details with a minimum mapping unit size of 4 hectares and scale of 1:50,000. However, some subthreshold polygons were added during the digitalization process of the original paper map. The overall accuracies of the DUSAF products were reported in Zaffaroni (2010) as approximately 95%. The DUSAF legend is adopted in conformity with the European nomenclature system of the CORINE Land Cover (CLC) and it is structured in a hierarchical way based on five levels of
investigation: the first three levels comply with the CLC nomenclature while the fourth and fifth ones represent additional levels specifically intended for identifying local characteristics.

The urban region was determined as follows: in a first step, we divided the territory of Lombardy into a 100 x 100-meter grid and calculated the proportion of urban surface (Pu) for each cell of the grid. The urban surface corresponds to class 1 of the DUSAF. Pu was then aggregated into five classes (Table 1) and we obtained the gridded map of urban land-use (Figure 2a,b). Population density (Popd) was first calculated for each municipality (Figure 2c) and then classified into three classes of density (Table 1) based on the distribution of the values (Pareglio, 2013). By intersecting a grid of 100 x 100 meters with the map of population density at municipality levels, we obtained the gridded map of population density (Figure 2c,d). For cells shared by two or more municipalities, population density of the municipality most represented in the cell ( % of surface) was assigned.

Table 1. Classes in which the proportion of urban surface (Pu) and population density (Popd) were classified.

<table>
<thead>
<tr>
<th>Classification of proportion of urban surface and population density</th>
<th>Pu (%)</th>
<th>Class</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt; 10</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>10 – 20</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>20 - 40</td>
<td>2</td>
<td></td>
</tr>
<tr>
<td>40 – 60</td>
<td>3</td>
<td></td>
</tr>
<tr>
<td>&gt; 60</td>
<td>4</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Popd (ab/km²)</th>
<th>Class</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt; 200</td>
<td>Low</td>
</tr>
<tr>
<td>200 - 1000</td>
<td>Medium</td>
</tr>
<tr>
<td>&gt; 1000</td>
<td>High</td>
</tr>
</tbody>
</table>

In a second step, we combined the two gridded maps obtained from the previous calculation (Pu and Popd maps) and obtained a new map with two measures for each cell (one measure relative to the proportion of urban surface and one relative to population density, Figure 2e). We then applied a final
classification depending on the combination of these two measures, as is shown in Table 2. Final classes were named as ‘Natural’, ‘Rural’, ‘Peri-urban’ and ‘Urban’ (Figure 2f) (Pareglio, 2013). To determine Milan’s spatial extension of the UR we examined the continuity of the urban and peri-urban tissue. Starting from the core of the city of Milan, we selected continuous urban and peri-urban cells (Figure 2g). Some cuts were made in correspondence to the major streets (highways), when these were not surrounded by urban centres, otherwise approximately all urban and peri-urban cells of the study area would result as continuous, with no meaning for our purpose (Figure 2h, see also Appendix Figure 1 for a detailed explanation of this procedure). Finally, we added a 2 km buffer to fill the holes left in the area. We thus obtained the urban region of Milan, with its urban and periurban surface, alongside with rural and natural areas, satellite cities and other minor centres (Figure 2i, 3).

Table 2. Criterion followed in classifying each cell of the grid, considering the combination of $Pu$ and $Popd$ values. Final classes are named as N (natural), R (rural), PU (peri-urban) and U (urban).

<table>
<thead>
<tr>
<th>Proportion of urban surface ($Pu$)</th>
<th>Population density ($Popd$)</th>
<th>Final classification based on the combination of $Pu$ and $Popd$ values</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>low</td>
<td>N</td>
</tr>
<tr>
<td></td>
<td>medium</td>
<td>R</td>
</tr>
<tr>
<td></td>
<td>High</td>
<td>PU</td>
</tr>
<tr>
<td>1</td>
<td>Low</td>
<td>R</td>
</tr>
<tr>
<td></td>
<td>medium</td>
<td>R</td>
</tr>
<tr>
<td></td>
<td>High</td>
<td>PU</td>
</tr>
<tr>
<td>2</td>
<td>Low</td>
<td>PU</td>
</tr>
<tr>
<td></td>
<td>medium</td>
<td>PU</td>
</tr>
<tr>
<td></td>
<td>High</td>
<td>PU</td>
</tr>
<tr>
<td>3</td>
<td>Low</td>
<td>PU</td>
</tr>
<tr>
<td></td>
<td>medium</td>
<td>PU</td>
</tr>
<tr>
<td></td>
<td>High</td>
<td>PU</td>
</tr>
<tr>
<td>4</td>
<td>Low</td>
<td>U</td>
</tr>
<tr>
<td></td>
<td>medium</td>
<td>U</td>
</tr>
<tr>
<td></td>
<td>High</td>
<td>U</td>
</tr>
</tbody>
</table>

26
Figure 2. Methodology to define the spatial extension of the urban region is schematically represented in this figure.

To compute statistics on variables accessible at administrative levels (e.g. resident population in the UR), we also selected the municipalities included in the UR. Only municipalities with 60% or more of their area enclosed in the boundaries of the UR were considered included in it (Appendix Figure 2).
2.2 Landscape fragmentation

Fragmentation was assessed with effective mesh size ($m_{\text{eff}}$; Jaeger, 2000). $m_{\text{eff}}$ expresses the probability that any two points chosen randomly in a region are connected; that is, not separated by barriers such as transport routes or built-up areas (Jaeger, 2000). The more the barriers fragmenting the landscape the lower is the probability of two points being connected (and the lower the $m_{\text{eff}}$). The minimum value of the effective mesh size is 0, and it corresponds to a completely fragmented landscape (that is, completely covered by urban surface), while the maximum value of $m_{\text{eff}}$ equals the size of the area studied and it corresponds to a completely unfragmented landscape. Effective mesh size assigns greater weight to large patches than to small patches and thus it is not usually equal to the average size of the patches. We calculated $m_{\text{eff}}$ following the CBC procedure (Cross Boundary Connections; Moser, Jaeger, Tappeiner, Tasser, & Eiselt, 2007), as follows:

Figure 3. Extension of the urban region (UR) of Milan.
Chapter I

\[
m_{\text{eff}} \text{CBC} = \frac{1}{A_{\text{total}}} \sum_{i=1}^{n} A_i A_i^{\text{compl}}
\]

Where \(n\) = the number of patches, \(A_{\text{total}}\) = the total area of the reporting unit, \(A_i\) = size of patch \(i\) inside the boundaries of the reporting unit \((i = 1, 2, 3, ..., n)\), \(A_i^{\text{compl}}\) = the area of the complete patch that \(A_i\) is a part of, i.e., including the area on the other side of the boundaries of the reporting unit up to the physical barriers of the patch (Appendix Figure 3). This procedure was introduced because the original computation of \(m_{\text{eff}}\) (the cutting-out procedure) suffered from the boundary problem. In fact, the boundaries of the reporting units were considered to be additional barriers, while the CBC procedure is independent of the size and administrative boundaries of the reporting units. This is advantageous because lead fragmentation values to refer to municipalities boundaries in this study, making this measure important for communication with decision makers, but does not allow them to erroneously influence fragmentation values (Moser et al., 2007). Landscape fragmentation was assessed for the municipalities within the Milan’s UR. We identified the landscape elements that were relevant to fragmentation. The choice of a specific set of fragmenting elements allow us to define the so-called “fragmentation geometry” (Jaeger, Soukup, Madriñán, Schwick, & Kienast, 2011). The delineation of the fragmentation geometry was based on GIS databases which included man-made barriers (urban areas, main roads and railways) and also natural barriers (rivers and lakes) and was represented by 2-dimensional features. The land cover and land use map (DUSAF) of the year 2012 was used to select urban areas, rivers and lakes (Table 3). The regional GIS database (CTR) of transportation network of the Lombardy region (www.dati.lombardia.it) was used to select main roads and railways as linear elements (Table 4). CTR (Regional Technical Map) is a vector database with a scale of 1:10,000. Linear elements were buffered based on road and railway classes in order to reflect the surface occupied (Jaeger et al., 2011). The two datasets used were intersected using ArcGIS 10.0 and any feature identified in either of the datasets as being a barrier, was assumed to be a barrier.
for the fragmentation geometry (Girvetz, Thorne, Berry & Jaeger, 2007). Reporting units considered in assessing fragmentation values were municipalities.

Table 3. Attribution of the land uses and land covers from the DUSAF project to the fragmentation geometry. Class ID (DUSAF) = represent the univocal code assigned to land cover classes in the DUSAF map; ID congruent with CLC = if Yes, the same univocal code for the land cover class is shared with Corine Land Cover maps.

<table>
<thead>
<tr>
<th>Land Use and Land Cover Class</th>
<th>Class ID (DUSAF)</th>
<th>ID congruent with CLC</th>
<th>Included in F.G.¹</th>
</tr>
</thead>
<tbody>
<tr>
<td>Continuous urban fabric</td>
<td>111</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Discontinuous urban fabric</td>
<td>112</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Industrial or commercial units</td>
<td>121</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Road and rail networks and associated land</td>
<td>122</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Port areas</td>
<td>123</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Airports</td>
<td>124</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Mineral extraction sites</td>
<td>131</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Dump sites</td>
<td>132</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Construction sites</td>
<td>133</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Degraded areas with no vegetation</td>
<td>134</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>Green urban areas</td>
<td>141</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Sport and leisure facilities</td>
<td>142</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Water courses</td>
<td>511</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Water bodies</td>
<td>512</td>
<td>Yes</td>
<td>Yes</td>
</tr>
</tbody>
</table>

¹ Fragmentation geometry

Table 4. Attribution of the linear fragmenting elements to the fragmentation geometry.

<table>
<thead>
<tr>
<th>Linear Elements</th>
<th>Buffer (m)</th>
<th>Included in F.G.¹</th>
</tr>
</thead>
<tbody>
<tr>
<td>Highways</td>
<td>50</td>
<td>Yes</td>
</tr>
<tr>
<td>Main roads</td>
<td>10</td>
<td>Yes</td>
</tr>
<tr>
<td>Secondary roads</td>
<td>5</td>
<td>Yes</td>
</tr>
<tr>
<td>Railways</td>
<td>2</td>
<td>Yes</td>
</tr>
</tbody>
</table>

¹ Fragmentation geometry

The degree of fragmentation assessed with \( m_{\text{eff}} \) can be alternatively expressed through the effective number of patches per unit (Jaeger et al., 2008; Moser et al., 2007). This is the effective mesh density \( (s_{\text{eff}}) \) and is related to effective mesh size according to \( s_{\text{eff}} = 1/ m_{\text{eff}} \). We chose to show the \( s_{\text{eff}} \) (Figure 6) because of its more intuitive reading in relation to fragmentation trends: an increase in \( s_{\text{eff}} \) indicates an increase in landscape fragmentation.
2.3 Urban sprawl

Urban sprawl was assessed by computing Shannon’s entropy (Yeh & Li, 2001), which is given by equation (2):

\[ H_n = -\sum_{i=1}^{n} P_i \ln P_i \]

where, \( P_i \) is the probability or proportion of a phenomenon (variable) occurring in the \( i \)th zone \( p_i = \frac{x_i}{\sum_{i=1}^{n} x_i} \), where \( x_i \) is the observed value of the phenomenon in the \( i \)th zone. The value of entropy ranges from 0 to \( \log_e(n) \). A value of 0 indicates that the distribution of built-up areas is very compact, while values closer to \( \log_e(n) \) reveal that the distribution of built-up areas is dispersed. Higher values of entropy indicate the occurrence of sprawl. The half-way mark of \( \log_e(n) \) is generally considered as a threshold: if the entropy value crosses this threshold the city is considered as sprawling (Bhatta 2010). Relative entropy can be used to scale the entropy value into a value that ranges from 0 to 1. Relative entropy \( H'_n \) can be calculated as (Thomas, 1981): \( H'_n = H_n / \ln n \). For relative entropy, a value of 0.5 is considered a threshold in determining the occurrence of sprawl (Bhatta et al., 2010).

To detect an increase in urban sprawl in a given area of interest, the measurement of the differences in entropy between time \( t \) and time \( t+1 \) indicate the magnitude of change in urban sprawl (Yeh & Li, 2001): \( \Delta H_n = H_n(t_2) - H_n(t_1) \). We assessed Shannon’s entropy for four years (1954, 1980, 2000 and 2012) and three different areas: (i) Lombardy, (ii) the UR – intended as the territory corresponding to the UR of the year 2012- and (iii) the area external to the UR. The classified land use and land cover maps (DUSAF) were reclassified into two categories: built-up land and non-built-up areas (Appendix Table 1). We then obtained a map for each of the three study areas where sprawl was investigated with two classes of land use. Each of the maps obtained was intersected with a regular grid of 1 x 1 km and the proportion of built-up surface was calculated for each cells. Shannon’s entropy was calculated from the built-up area for each cells, considered as an individual zone (\( n \) is the total number of cells). Variation in entropy between different areas provides evidence of different effects induced by land-use patterns. Moreover, by analysing urban sprawl in a temporal series, we
were able to observe whether Milan’s urban development followed a more dispersed (sprawled) or compact pattern of urban expansion.

In a second step, we analysed sprawl on a more detailed scale to ascertain the emergence of patterns of urban sprawl. To do this, we analysed Shannon’s entropy in 7 transects (Figure 5). Transects were drawn following the main infrastructures that connected the city of Milan with satellite cities and satellite cities with each other. Sprawl normally takes place in radial direction around the city centre or in linear direction along the highways (Sudhira et al., 2004). In this study, five transects were drawn congruent with the direction of expansion of the urban tissue (transects MI-VA, MI-LO, MI-BG, BG-BS, BS-MN) and two transects followed opposite directions (transects MI-PV and BS-CR; for the expansion of urban tissue see Appendix Figure 4).

Figure 5. Transects within which entropy was assessed. They follow the main infrastructure that connect the city of Milan with satellite cities and satellite cities with each other: Five transects were congruent with the direction of expansion of the urban tissue (transects MI-VA, MI-LO, MI-BG, BG-BS, BS-MN) and two transects (MI-PV, BS-CR) followed opposite directions.
In order to compute Shannon’s entropy, we used the land use and land cover map reclassified in built-up and non-built-up lands. Each transects represented an independent study area where sprawl was determined. Transects were drawn in ArcGIS 10.0 as linear elements connecting two points (i.e. two towns) or following the direction of the main infrastructures and were subsequently buffered on each side with a 5 km buffer. In order to compute Shannon’s entropy, we divided each linear transects into regular segments (of approximately 5 km of length) and the proportion of built-up surface was calculated for each segments which represented an individual zone ($n$ in the equation). Entropy within these transects was analysed for years 1954, 1980, 2000, 2012 and temporal trends were described as well as changes of sprawl values within the transects.

3. Results and Discussion

3.1 Extension of the Urban Region of Milan

Population of a city can be used as indicator of size, because somewhat consistent and reliable data are available for every cities (Turner, 2005; Forman, 2008). Following Forman (2008), Milan is considered as a medium city (the range is from 1 to 4 million inhabitants). The UR around the city of Milan covers around a third of the Lombardy region (Figure 3). It is clear that UR representation goes beyond administrative boundaries of municipalities and provinces and allows us not to restrict the analysis of urban processes (like sprawl) to pre-constituted limits. However, some statistics of the UR were presented in this study at administrative levels, in order to be informative for planning purposes (Table 5).

In an urban region, satellite cities located around the major city are defined as normally having <250,000 inhabitants (Forman, 2008). In our study, several satellite cities were included in the UR area: three cities between 100,000-200,000 inhabitants (Brescia “BS”, Monza “MB” and Bergamo “BG”) and other minor cities of less than 100,000 inhabitants (Como “CO”, Varese “VA”, Lecco “LC” and Lodi “LO”).
Table 5. Summary statistics of the urban region (data refer to the year 2012).

<table>
<thead>
<tr>
<th>Urban Region of Milan</th>
</tr>
</thead>
<tbody>
<tr>
<td>UR surface</td>
</tr>
<tr>
<td>% surface of Lombardy</td>
</tr>
<tr>
<td>n° of municipalities included</td>
</tr>
<tr>
<td>% of Lombardy municipalities included in the UR</td>
</tr>
<tr>
<td>Population residents in the UR</td>
</tr>
<tr>
<td>Population resident in Lombardy</td>
</tr>
<tr>
<td>Average population density in the UR</td>
</tr>
<tr>
<td>Average population density in Lombardy</td>
</tr>
</tbody>
</table>

Municipalities that are included in the UR (Appendix Figure 2) represent almost half of the total municipalities of Lombardy. The core of the region is the city of Milan, where the density of urban settlements and population is higher and where the major infrastructure (motorways, railways and main roads) converge. Overall, land use and land cover within Milan UR is mainly represented by anthropic or agricultural areas (together covering approximately 70% of the area), while forest and semi-natural lands together with water bodies and wetlands cover the rest of the area of the UR (Figure 4b). Within other urban regions, Forman (2008) found that agriculture is often more prominent close to the metropolitan area than across the urban-region ring as a whole, where instead natural land usually predominates. In these regions, urbanization spread of the metropolitan area mainly covers valuable agricultural land and studies indicated that, since the original community began by prime agricultural soil, urbanization over time might cause a significant loss of the region’s best soils (Forman, 2008). The same concern is shared for lands around the Milan UR, where the recent urban expansion has led to the consumption and degradation of the soils regardless to their quality (ISPRA, 2016). Previous findings showed that most urban regions have no towns in natural areas (Forman, 2008). However, urbanization that spread from satellite cities within the urban region may degrade proximal natural lands and this pressure may be quite significant depending on the amount and location of these lands (Forman, 2008). In our study, most of the satellite cities are close to natural and semi-natural areas and their urban expansion will lead to a significant consumption of
these lands especially for those cities located in the northern part of the UR (Varese, Como, Lecco, Bergamo and Brescia). Milan UR area is morphologically characterized by a northern band of natural and semi-natural lands which is separated from the southern agricultural lands by a semi-continuous urban “belt” (Figure 4a), where the remnant fringes of natural and agricultural lands intersected. The spatial configuration of some towns that we found in the Milan UR is consistent to what found in other regions, where quite often towns were located near the boundary between agricultural and natural areas (Forman, 2008). Because of the ecological value of these lands, limiting growth of the existing towns located nearby would be a valuable investment.

Figure 4. Land use and land cover for a) the territory of Lombardy excluded by the UR and b) the UR. LULC classes correspond to the DUSAF classification: artificial surfaces (class 1), agricultural areas (class 2), forest and semi natural areas (class 3) and wetlands and water bodies (classes 4 and 5).

The shape of the Milan UR expands in two directions starting from the city of Milan: one axis of expansion is towards the north-west and the other towards the east. Previous studies (Lanzani, 2011) already emphasized the considerable importance of the dry lowland area to the north of Milan and of the lowland towns arranged along a north-west to south-east line descending from Bergamo to Brescia
and then towards Mantova. Factors affecting the shape of the UR are not analysed in this study but further analysis aiming to take into consideration both geographic and socio-economic drivers in the process of shaping the UR would certainly improve our understanding of this urban area. The role of higher elevation areas located in the northern part of the region (which are less accessible and limit the infrastructure development) together with the presence of a plain topography in the southern lands (which on the contrary facilitates transportation infrastructure development) as well as different agricultural development (past non-irrigated crops in the north, that were less profitable compared to the south) are all aspects on which attention should be placed.

3.2 The current status of landscape fragmentation in the UR of Milan

Results of this study lead us to represent the degree of landscape fragmentation operated by urban tissue to the lands still not converted to urban uses (agricultural, natural and semi-natural lands). Industrialisation, urban development, and the construction of transportation infrastructure are more concentrated in the municipalities in the north of the city of Milan and to a lesser degree, around some satellite cities. Results of $S_{eff}$ were mapped and provided a picture of the current status of landscape fragmentation within the UR (Figure 6). The metric produces a map of the spatial distribution of fragmentation, as well as quantitative data on the level of fragmentation present in different planning areas relevant to planners (Girvetz et al., 2007), that are represented by municipalities in this study. The degree of fragmentation varies among municipalities within the UR. Higher fragmented municipalities are more concentrated in the areas to the north and north-west of the city of Milan (close to the cities of Monza, Varese, Como, Lecco). Municipalities close to the pedemontan area (north belt of the UR) and in the east part of the UR showed a lower degree of fragmentation. This pattern may be understood by considering that these areas underwent different urban pressures caused by their development (Lanzani, 2011). In particular, municipalities in areas to the north and north-west of Milan correspond to those that were subjected to the first (and much stronger) expansion of the urban tissue (Appendix Figure 4). Results reveal that the provinces with the highest degree of
landscape fragmentation are those of Milano, Monza, Varese and to a lesser degree Como, Bergamo and Lodi. Around the south of Milan, especially in the last 30 years, it has been observed a residential suburbanization which involved not only the first belt but also the second belt in the areas of the provinces of Lodi and Pavia (as a result of the first signs of congestion in the first belt close to the city of Milan) (Lanzani, 2011). For the Lombardy region, it has been highlighted in other studies a diffuse shift in residential consumption linked to the fragmentation of family nuclei and therefore to the growing number of homes and of area per capita, with a clear prevalence of preferences for certain kinds of building i.e. (the detached house) (Lanzani, 2011), which has contributed to the diffuse increase of landscape fragmentation.

![Figure 6. Landscape fragmentation expressed by effective mesh density ($s_{eff}$) in the municipalities included in the urban region. High values of $s_{eff}$ correspond to high fragmentation. The categories show the sizes of the remaining patches.](image)

Figure 6. Landscape fragmentation expressed by effective mesh density ($s_{eff}$) in the municipalities included in the urban region. High values of $s_{eff}$ correspond to high fragmentation. The categories show the sizes of the remaining patches.
3.3 Spatio-temporal patterns of urban sprawl

The spatiotemporal pattern of urban sprawl was analysed by calculating Shannon’s entropy for different areas and times. For computation of urban sprawl low values of Shannon’s entropy (H’) indicate compact distribution of built-up areas, while high values show a dispersed distribution and thus an occurrence of urban sprawl (Sudhira et al., 2004). Table 6 gives entropy values for three different areas of investigation: Lombardy, the UR and the area outside the UR. Relative entropy H’\textsubscript{n} was used to compare entropy for areas of different spatial extension. Results show that entropy was considerably high in each area considered (mean value: 0.89, standard dev.: 0.03) and relative entropy reveals an occurrence of sprawl (H’\textsubscript{n} > 0.5) both in the UR and in other areas (Figure 7). Entropy within the area of the UR was higher than in Lombardy and in the area outside the UR, thus revealing that this area is the core of the sprawl phenomenon, but all Lombardy experienced a high degree of urban sprawl. Relative entropy H’\textsubscript{n} in the UR passed from 0.89 to 0.95 (+ 0.06) in the period 1954-2012, thus showing an increasing trend. The trend was positive also for Lombardy and the area external to the UR, although these increases were less marked than for the UR (H’\textsubscript{n} increase respectively of + 0.04 and + 0.02). Entropy was also calculated in seven transects and results are shown in Table 7. It is known that sprawl usually takes place on the urban fringe, at the edge of an urban area or along the highways (Sudhira et al., 2004).

Table 6. Shannon’s entropy (H’) and relative entropy (H’\textsubscript{n}).

<table>
<thead>
<tr>
<th></th>
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</thead>
<tbody>
<tr>
<td><strong>Lombardy</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>H’</td>
<td>8.79</td>
<td>8.66</td>
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<td>9.15</td>
</tr>
<tr>
<td>H’\textsubscript{n}</td>
<td>0.87</td>
<td>0.86</td>
<td>0.9</td>
<td>0.91</td>
</tr>
<tr>
<td><strong>Urban region of Milan</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>H’</td>
<td>7.95</td>
<td>8.13</td>
<td>8.44</td>
<td>8.49</td>
</tr>
<tr>
<td>H’\textsubscript{n}</td>
<td>0.89</td>
<td>0.91</td>
<td>0.94</td>
<td>0.95</td>
</tr>
<tr>
<td><strong>Territory of Lombardy excluded to the UR</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>H’</td>
<td>8.36</td>
<td>8.36</td>
<td>8.49</td>
<td>8.53</td>
</tr>
<tr>
<td>H’\textsubscript{n}</td>
<td>0.87</td>
<td>0.82</td>
<td>0.88</td>
<td>0.89</td>
</tr>
</tbody>
</table>
In this study, five transects were drawn congruent with the direction of expansion of the urban tissue and two were not. Entropy values were comparable for all the transects, but entropy magnitude of the transects that did not follow the direction of expansion of the urban tissue was negative (-0.04 and -0.05), thus indicating that the sprawl in these transects followed an opposite trend. A partial explanation of this fact can again be found in the importance of agricultural production in the lands south of Milan. To preserve agricultural activity, the plan of the territorial administration provides protection for these lands and confines urban expansion to areas close to urban centres. However, as we stated before, this may have determined only partially the trend towards a more compact urban development, and other causes need to be investigated (Bencardino, 2015; Pareglio, 2013).

Table 7. Shannon’s entropy calculated in seven transects. Five transects followed the main direction of expansion of the urban tissue and two transects followed opposite directions (*). ∆H indicate an increase (+) or decrease (-) of sprawl between two time periods.

<table>
<thead>
<tr>
<th>Transects</th>
<th>1954</th>
<th>1980</th>
<th>2000</th>
<th>2012</th>
<th>∆H 54-12</th>
<th>∆H' 54-12</th>
</tr>
</thead>
<tbody>
<tr>
<td>MI-VA</td>
<td>2.14</td>
<td>0.89</td>
<td>2.28</td>
<td>0.95</td>
<td>2.33</td>
<td>0.97</td>
</tr>
<tr>
<td>MI-BG</td>
<td>1.78</td>
<td>0.81</td>
<td>1.85</td>
<td>0.84</td>
<td>2.12</td>
<td>0.97</td>
</tr>
<tr>
<td>BG-BS</td>
<td>1.98</td>
<td>0.90</td>
<td>2.09</td>
<td>0.95</td>
<td>2.14</td>
<td>0.98</td>
</tr>
<tr>
<td>BS-MN</td>
<td>2.37</td>
<td>0.95</td>
<td>2.33</td>
<td>0.94</td>
<td>2.38</td>
<td>0.96</td>
</tr>
<tr>
<td>MI-LO</td>
<td>1.67</td>
<td>0.76</td>
<td>1.85</td>
<td>0.84</td>
<td>1.97</td>
<td>0.90</td>
</tr>
<tr>
<td>MI-PV *</td>
<td>2.04</td>
<td>0.88</td>
<td>1.81</td>
<td>0.79</td>
<td>1.93</td>
<td>0.84</td>
</tr>
<tr>
<td>BS-CR *</td>
<td>2.14</td>
<td>0.97</td>
<td>1.93</td>
<td>0.88</td>
<td>2.00</td>
<td>0.91</td>
</tr>
</tbody>
</table>

Observing the results (Tables 6, 7), some data differ a bit from the general trend (data referring to the year 1980). This is explained by the fact that the land-use and land-cover map of the year 1980 have a minor scale of detail with respect to the other maps. Even though some subthreshold polygons were added during the digitalization process of the original paper map, the resulting map of the year 1980 has scarce quality in terms of detail and many polygons are missing (which are instead present in the 1954, 2000 and 2012 maps). In general, we observed that sprawl has followed a linear trend (positive
or negative depending on the transects) during the 58-years period investigated. This can inform us about the direction of the process and thus be a useful indication for urban planners who aim to monitor sprawl. As stated by Brezzi et al. (2012), in the period 2000-2006, the rate of population growth in the cores of metropolitan areas in many countries (including Italy) was lower than the rate of urbanised land growth. This phenomenon is intrinsic in the process of sprawl occurring in large metropolitan areas, with proliferation of dispersed buildings and under-utilization of infrastructures accompanied by inefficient resources utilization. Land takeover may also be promoted by increasing urbanization pressures (Guastella & Pareglio, 2014). It is important to consider that in Italy urbanization processes suffer from the need of local administrations to balance current expenditures with land-use rights (Guastella & Pareglio, 2014), and not only socio-demographic trends or unplanned expansion are the causes of urban expansion. Unfortunately, therecent economic crisis may consolidate the trend of expanding urbanization.

Figure 7. Urban sprawl expressed by relative Shannon’s entropy ($H'_n$) for the UR and for the areas outside the UR for four temporal steps.
Chapter I

4. Conclusions

This study gives an explicit spatial definition of the urban region of Milan and analysed urban sprawl and landscape fragmentation at this scale, comparing different areas and times. The derivation of methodologies able to describe urban areas can help in responding to relevant policy questions (Brezzi et al., 2012) and can be useful in informing planners in implementing a sustainable use of resources. A recent analysis (OECD, 2010) argues that policy-makers concerned with sustainable development should focus more on the form and quality of urbanisation processes rather than simply on the volume and speed of urbanisation. Low urban densities accompanied by the expansion of the urban space it is known to lead to loss of green space, high cost of infrastructure and energy, increased social segregation and functional land-use divisions, which both reinforce the need to travel and increase dependence on the private motorised transport model, leading in turn to increased traffic congestion, energy consumption, and polluting emissions (Travisi et al., 2010; OECD, 2004; CEC, 2000). Thus, an appropriate estimation of the amount of land which will be needed by urban development is needed in order to implement adequate spatial strategies that can be achieved with the lowest cost and highest efficiency possible (Inostroza et al., 2013). The measurement of urban form and of the changes in shape, size, and configuration of the built environment can provide a more systematic analysis of the relationships between urban form and process (Yeh and Li, 2001). The relationship between landscape fragmentation and urban sprawl has not been examined in this paper. However, the issue constitute an interesting research topic that deserve to be investigated in future studies, giving also the scarcity of studies that aimed to explore it in details made so far (Torres et al., 2016). As recently highlighted by Torres et al., (2016) the anthropogenic landscape fragmentation appear to be only partly explained by urban sprawl patterns and the relationship is scale-dependent, with sprawl and fragmentation patterns matching more closely at finer scales (i.e. an urban region scale may be a suitable scale). Urban sprawl, with its wide dispersion of metropolitan areas and the spread of cities with high consumption of scarce resources, is a relatively recent phenomenon in
Europe (Travisi et al., 2010). In the current scenario of constant urban expansion, the impelling question for the urban planning is what the acceptable degrees of sprawl and fragmentation are (Insotroza et al., 2013). Land cover changes analysed with detailed spatial information on a regional scale may help in planning urbanization processes that are of concern. As urban expansion will inevitably take place in most of the urbanized areas worldwide, there should be concern about its spatial configuration and about the amount of land which will be needed by urban development in order to implement adequate (low cost and high efficient) spatial strategies (Insotroza et al., 2013). The present study is important in identifying not only general trends, but also in recognizing which areas suffer from the higher pressures of urbanization and thus help in implementing targeted policies to limit threats to the environment. It is of fundamental importance to integrate the assessment of urban processes into urban planning, and thus specific studies aiming to monitor the degree of fragmentation and sprawl of our urban regions are essential. In urban areas, every unit intervention further expanded the existing urbanizations and for the Italian territory a cycle does not yet seem to have begun of re-use and densification of already urbanized areas, with the exception for the re-use of some large abandoned areas in towns (Lanzani, 2011). In Lombardy as in the rest of Italy, an excess of bureaucratization and legislation in the sector of planning (together with the applicability by discretion of many rules) was not matched by a capacity to govern their actual design and their impact on the territory. In Lombardy, a too weak presence and policy of parks (regional and municipal) determined a continuous interclusion and fragmentation of the open spaces in the territory which (except in rare areas) shows a low profitability from agriculture and therefore low resistance not only to diffuse building but also to improper uses for dumping, deposits, fencing in etc. (Lanzani, 2011). Furthermore, in comparison to other large European urban areas what differentiate the UR of Milan is a complete absence of a strong policy of reorganization of public transport with enhancement of the networks and services and also an effective governance of the location of high-attraction functions (i.e. new public services and many commercial facilities) (Lanzani, 2011). For what concern the governance of the residual open spaces, which are mainly destined to further fragmented and
subjected to degrading uses, an active policy which upgrade in sustainability is needed the set up (Lanzani, 2011). Despite these concerns, in the recent report on soil consumption of the Italian Institute for Environmental Protection (ISPRA, 2016) the province of Milan has been rank as one with the highest increase of land consumption for the period 2012-2015 (<0,9%). The measure of the amounts of land in urban use and its spatial configuration is essential in order to provide useful information to project future needs, and to ensure the adequate supply of public good e.g. infrastructure, open space, and common facilities for urban expansion (Angel 2010).

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Pileri, P. (2012). Land cover changes in Lombardy over the last 50 years. Publisher: *Regione Lombardia*; ERSAF, pp.185-204.


Appendix of Chapter I
Figure 1. To have a proper representation of the urban region and avoid the presence of long strips of territory that have no meaning for our purposes, we proceeded to identify ‘cutting points’ using a standardize methodology. We visually identified the major roads where a cut was needed (a). We then divided those areas into sections having the same length by identifying a buffer of 2 km around the major roads on either side (b). In each section, we calculated the portion of urban surface and ‘cut’ the urban region when a section with less than 50% of urban surface was found (b). This figure shows two sections as examples (b): the urban region was cut in correspondence to the border of the ‘Sez1_A1’ (urban surface 69%), because the successive section (“Sez2_A1”) had 36% of urban surface.
Figure 2. Municipalities included in the urban region. Only municipalities with 60% or more of their area enclosed in the boundaries of the UR were considered included.

Figure 3. Calculation of effective mesh size ($m_{\text{eff}}$) following the CBC procedure (Cross Boundary Connections; Moser et al., 2006). In our study, Reporting units were municipalities. In this example, the continuous red line represents the boundaries of the reporting unit ($A_{\text{total}}$ = the total area of the reporting unit) which intersect two different patches (dark brown and light brown): $A_i$ represents the proportion of the patch light brown included in the reporting unit and $A_{i,\text{empl}}$ the total area of the patch light brown that $A_i$ is a part of (and that goes beyond the boundaries of the reporting unit).
Figure 4. Expansion of the urban tissue (the arrows indicate the directions) around the city of Milan from 1954 to 2012.
Chapter II

Urban parks as habitat providers for biodiversity: a multi-scale analysis in the urban area of Milan

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Submitted to Urban Forestry and Urban Greening
Abstract

Actual trends draw attention to a significant biodiversity decline associated with current and upcoming degree of urbanization. In this context, urban parks can play a crucial role for the conservation of many species within cities. Urban systems are highly dynamic and complex human-shaped ecosystems, leading the maintenance of high biodiversity levels within them a challenging goal to undertake. In this study, we conducted a multi-scale analysis in order to detect how environmental features in an urban area affect biodiversity and we provided suitable information for implement an effective management of green spaces with the aim of support high level of biological diversity. Fifteen urban and peri-urban parks in the metropolitan area of Milan (Italy) were investigated and 93 sampling plots for birds (species and number of individuals) and vegetation (trees Diameter Breast Height) sampled. Results of GLMs at landscape scale showed how bird species richness and abundance are significantly affected by park area and three land cover types and the importance of the environmental variables was determined using RDA components. In a finer level of analysis we described how 28 birds species respond differently to land cover types, presence of water and distance from park's border within sampling points. Here, wooded land cover type and water bodies resulted to be the variables that mostly affect birds presence. For forest species, we further investigated the effect of vegetation structure. Results showed unusual pattern for some species (preference of little trees) that can be explained by considering the effects of environmental features acting at different scales. A multi-scale approach is essential to proper understand biodiversity patterns and concerted conservation actions are necessary.

1. Introduction

Actual trends draw attention to a significant biodiversity decline associated with current and upcoming degree of urbanization (McDonald et al., 2008). Evidences demonstrated how urban areas have been already responsible by a notable extent to the threats to the survival of many species (Venter
et al., 2006; Turner et al., 2004; McKinney, 2002). McDonald et al. (2008) estimates that urbanization is implicated in the listing of around 8% of the IUCN vertebrate species. Urbanization also reduces the quantity of native vegetation and alters its local structure and regional spatial pattern (Donnelly and Marzluff, 2006) by inducing habitat fragmentation and by favour invasion of exotic species. The loss of biodiversity is of critical concern, given that diversity plays an important role in long-term ecosystem functioning and in ecosystems services provisioning (Mace et al., 2012; Haines-Young and Potschin, 2010). Concern arising from urbanization is also due to the increase of biotic homogenization (Alvey, 2006; McKinney and Lockwood, 1999) that lead the same “urban-adaptable” species becoming increasingly widespread and locally abundant in cities across the planet (McKinney, 2006), with a loss of less adaptable species. As a consequence, the urban avian community is going to be composed by few species that may dramatically differ from those of local natural environments (Chace and Walls, 2006; O’Connell et al., 2000). Consequences of biotic homogenization act at local as well as global scale by leading to local extinctions and decrease the overall global biodiversity (McKinney, 2006). Protected areas are not sufficient to safeguard biodiversity by themselves (Rodrigues et al., 2004) and it is of fundamental importance to promote conservation actions also in non-protected areas. In addition, the distance between protected areas and cities is predicted to shrink dramatically in some regions (McDonald et al., 2008) because of the expansion of urban areas, and thus conservation actions that go beyond the boundaries of natural protected areas in a more comprehensive strategy are required. In this context, green spaces within urban areas may provide suitable refuges for species whose native habitats have been greatly diminished (Angold et al., 2006). Green spaces of good ecological quality occurring in cities may also function as dispersal corridors for various organisms (Sandström et al., 2006; Bolger et al., 2001) and even allow the occurrence of specialised forest species in urban landscapes (Park and Lee, 2000; Mörtberg and Wallentinus, 2000). Moreover, vegetation in cities can comprise a significant percentage of a nation’s tree canopy, leading their management to have important effects not only to a local but also to a regional or national scale.
It has been shown that, in general, the structural simplification of vegetation in green areas has negative impacts on biodiversity (McKinney, 2006). In fact, structural complexity can provide the within-stand variation in habitat conditions required by some taxa (a ‘habitat heterogeneity’ function, Lindenmayer et al., 2006; Sanesi et al., 2009; Savard et al., 2000). However, ecological value of urban forests may be deflated by anthropogenic actions such as, for example, frequent cuts, leaf litter removal, plantation of exotic species, etc., or by stressors derived from the external urban matrix. Despite the primary importance played by vegetation in harvest species, it is important to remind how the complexity of urban systems need an effort to understand the importance of the factors that act locally (Melles et al., 2003). In fact, forest fragments of similar size and vegetative structure may not be ecologically equal because of differences in their surrounding landscapes (Friesen et al., 1995). For example, in small patches it is possible that ecosystem dynamics are driven predominantly by external rather than internal forces (Saunders et al., 1991) and urbanization processes may lead to detrimental influences (i.e. population decline) on species even when forest patches are maintained (Engels and Sexton, 1994; Herkert et al., 1993). External effects may severely deflate the ecological value of adjacent forests and anthropogenic stress might act in different ways on forests patches. Moreover it should be considered that the influences of different environmental features on biodiversity operate also at different spatial scales. These spatial scales are not independent from one to another but linked in a hierarchical way (Allen and Starr, 1982): the effects of an action at a given scale must be considered on higher and lower scales (Savard, 1994). For this reasons a multi-scale approach should be considered the proper way to address biodiversity questions (Savard et al., 2000) also in urban landscapes, where on the contrary it appears to be often neglected. Cities are highly dynamic and complex human-shaped ecosystems, rendering the maintenance of high biodiversity levels within them a challenging goals to undertake. A deeper understanding of what biodiversity needs to be maintained and enhanced in cities is of fundamental importance to plan effective conservation strategies aiming to reduce the ecological footprint and ecological debt of cities towards nature. In this study we investigated: i) how environmental features at landscape scale affected
biodiversity by studying 15 urban and peri-urban parks in one of the most urbanized areas of Europe (the metropolitan area of Milan), ii) how biodiversity is affected by site-scale environmental variables by studying 28 bird species over 93 sampling points within the parks and iii) how vegetation structure within sampling points affected forest species abundance. We choose to use birds as reference taxa to evaluate the level of biodiversity in urban environment (Sandström et al., 2006). They show several useful features which made them a suitable indicator to explore habitat quality in urban landscapes (Fernandez-Juricic, 2004; Bolger et al., 2001; Hostetler and Holling, 2000; Mörtberg and Wallentinus, 2000): they are mobile organisms, which require the existence of a wide variety of habitat at different spatial scales and they are relatively conspicuous and fairly easy to survey (Sandström et al., 2006).

2. Materials and Methods

2.1 Study area

The study area comprises the city of Milan and its metropolitan surrounds. Milan city has a population of 1,345,851 million (ISTAT, 2015) with a surrounding province of 3,208,509 million (ISTAT, 2015). Compared to other Italian cities, Milan has a considerable amount of urban green spaces. However, the area around the city (in particular in the north) experienced a high degree of urban development in the last decades, leading to be one of the most urbanized area of the country and of Europe (Trono and Zerbi, 2002). Geographically, the metropolitan area of Milan extends between the alluvial plain of the Po river and the mountainous area of the Pre-Alps. For this study, we selected 15 urban and peri-urban parks located in the metropolitan area of Milan, ranging from small, structural simple and city central parks to larger, structural complex and periurban parks (Table 1).
Table 1. Parks selected for this study.

<table>
<thead>
<tr>
<th>Park name</th>
<th>Area</th>
<th>Distance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Monte Stella</td>
<td>38.6</td>
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</tr>
<tr>
<td>Parco Guido Vergani</td>
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<td>Parco Lambro</td>
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<td>Parco Forlanini</td>
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<td>Parco Sempione</td>
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<td>Boscoincittà</td>
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<td>8.17</td>
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<td>Parco di Trenno</td>
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</tr>
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<td>Parco Solari</td>
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</tr>
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<td>Parco delle Cave</td>
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<td>Parco Ravizza</td>
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<td>Parco Nord</td>
<td>596.5</td>
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</tr>
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<td>Parco di Monza</td>
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</tr>
<tr>
<td>Bosco delle Querce</td>
<td>42.8</td>
<td>20.50</td>
</tr>
</tbody>
</table>

2.2 Bird surveys

Birds surveys were carried out using point-counts (Raplh et al., 1998) in 93 sampling points randomly distributed in 15 urban parks (the number of point-counts in each park was proportional to the parks’ area). Minimum distance between two points was set to at least 200 m in order to prevent overlapping observations (Sandström et al., 2006). Surveys were conducted during spring from April to June in the year 2014 in days with no adverse meteorological conditions (wind, clouds or rain). Each point-counts was surveyed two times. For each species, we recorded both occurrence and number of individuals detected in each point distinguishing between birds contacted within and beyond the point area (a circular buffer of 100 m of ray around the point). Overall data were used to estimate species richness at park’s level, while data referred to birds detected within the point area were used for further analysis.
2.3 Environmental features

Data on environmental features were recorded directly during the field surveys or derived from a Geographic Information System (elaborated with ArcMap 10.2.2) or combining either methodologies when need to validate GIS information. Landscape variables were recorded for the whole park, while site-scale variables and DBH were collected within the point-counts area. The following seven variables were recorded at landscape level: surface covered by trees (m$^2$) in the park; surface covered by grassland (m$^2$) in the park; other coverings (usually represented by paving or buildings; m$^2$) in the park; presence of water (rivers or lakes) in the park; age of the park (years from the establishment); unbuilt surface in a buffer of 1000 m around the park (m$^2$). Within each point-count we recorded the following environmental variables: surface covered by trees (%); surface covered by grassland (%); other coverings (%); presence of water (rivers or lakes); minimum distance from the park’s border (m). Moreover, we conducted surveys on the structure of the vegetation recording the DBH (Diameter at Breast Height taken at 1.3 m above the ground) in each site. The number of trees measured was proportional to the amount of surface covered by trees in that site. A maximum of 100 trees per site were randomly measured in site occurring in forested areas (where the wooded land cover type was the 100%) and a minimum of 0 trees were measured in grassland sites (wooded land cover type was 0%).

3. Data analysis

3.1 Detectability and abundance of birds

Detection of animals is rarely either perfect or constant due to many reasons like observer error, species rarity or because detection varies with confounding variables such as environmental conditions (Kellner and Swihart, 2014). Failure to correct for imperfect detection may result in bias in, among others, estimation of relationships with ecological covariates (Zipkin et al., 2010; Gu and Swihart, 2004). In order to account for that, we assessed the probability of occupancy (psi) of the
sampling points for every bird species recorded using PRESENCE 11.2 (Hines, 2006). PRESENCE describes the probability of detecting a species using a probabilistic argument to describe the observed detection history for a site over a series of surveys (MacKenzie et al., 2002). This method leads to estimate the probability of site occupancy in situations where a species is not guaranteed to be detected even when it is present, reducing therefore the risk of underestimation of the occupancy. On the basis of the occupancy results for every species we calculated the misdetection rate as the percentage difference between the observed occupancy and the occupancy estimated by PRESENCE. We then selected only the species observed at least in the 10% (n = 28) of the point-counts surveyed. The probability of occupancy of each point (conditional psi) was used to assess the relationships between the 28 bird species occurrence and the environmental features at site-scale. For the species with a low misdetection rate, we have also taken into account the maximum number of individuals recorded for each point between the two sampling sessions performed in order to assess the relationships between the single species abundance and some of the environmental features recorded.

3.2 Relationships between species richness and abundance at landscape scale

To disentangle the importance of environmental variables on birds species richness and abundance we performed a series of constrained redundancy analyses (RDA) using as endogenous dataset the total number of species recorded and the maximum number of birds recorded and as the exogenous dataset the environmental variables. The RDA is a canonical analysis that combines the proprieties of regression and ordination techniques and that evaluates how much of the variation of the structure of one dataset (e.g., community composition in a forest, endogenous dataset) is explained by the independent variables (e.g., habitat features, exogenous datasets) (Borcard et al., 2011). To assess the significance of the explained variance by the RDAs and avoid type-I error, ANOVA-like permutation tests (10,000 permutations) were performed. Moreover we extracted the scores of the two first components of the RDA (RDA1 and RDA2) for each park. To assess the relative role of the landscape environmental variables we built separated linear mixed models using RDA1 and RDA2 scores as
independent variables and birds species richness and birds abundance as dependent variable. This method allowed us to reduce the correlated variables and to perform Linear Models (LMs) on a lower number of uncorrelated components. Analysis were run using R Studio version 0.98.1091.

3.3 Relationships between species occurrence and abundance at site scale

In a finer level of analysis, we wanted to find the relationships among species occurrence and sites features. RDA analyses were performed on a subset of 28 species observed in at least the 10% of the sampling plots. The probability of occupancy (psi) at a given sector as estimated by PRESENCE was assumed for each species (endogenous dataset). The environmental variables of the sites were the exogenous dataset. As for previous analysis, ANOVA-like permutation tests (10,000 permutations) were performed to assess the significance of explained variance by RDA.

In wooded areas, structural complexity of vegetation can provide the habitat conditions required by some birds and these requirements may vary among different species. For birds species that has forest habits, we tested the presence of linearity in the relationship with mean DBH and the maximum number of individuals recorded. For this analysis we selected the Eurasian blackcap (Sylvia atricapilla), the great tit (Parus major), the common chaffinch (Fringilla coelebs), the common blackbird (Turdus merula), the great spotted woodpecker (Picoides major), the eurasian blue tit (Parus caeruleus) and the european green woodpecker (Picus viridis) because they showed a good detection probability. We used Generalized Additive Models (GAMs) assuming the park as random factor and a Poisson error distribution. In GAMs, increasing values for the effective degrees of freedom (edf) indicate an increased complexity and non-linearity of the response curve (Wood, 2006); we therefore considered edf of 1 as an evidence of a linear relationship, while values higher than 1 indicated a non-linearity (Digiovinazzo et al., 2010).
4. Results

4.1 Birds surveys

A total of 63 species of birds were detected in the study area and among these 18 are listed in a protection list (Table 2). In total 3343 individuals in the first survey and 3541 in the second were observed. Birds contacted comprise not only common species in urban environments (i.e. Rock dove (*Columba livia*), Common wood pigeon (*Columba palumbus*), Eurasian collared dove (*Streptopelia decaocto*), Barn swallow (*Hirundo rustica*), European robin (*Erithacus rubecula*), Common blackbird (*Turdus merula*), Hooded crow (*Corvus cornix*) but also elusive species (Common firecrest (*Regulus ignicapillus*), Red-backed shrike (*Lanius collurio*) or Eurasian tree sparrow (*Passer montanus*) and aquatic birds (i.e. Little grebe (*Tachybaptus ruficollis*), Great crested grebe (*Podiceps cristatus*), Little bittern (*Ixobrychus minutus*) or Mallard (*Anas platyrhynchos*) or birds usually associated with agricultural environments (i.e. Grey heron (*Ardea cinerea*), Common pheasant (*Phasianus colchicus*)). Most frequent species observed in the point-counts and number of individuals are showed in Figure. 1.

![Figure 1](image)

Figure 1. Frequencies of observation of the most common species (detected in more than 20% of sampling points) and number of individuals (above the columns, here is reported only number of individuals of the first survey as example).
Table 2. Conservation priority are assessed as following: “V”, listed in Annex V of Birds and Habitats Directive; “VU”, vulnerable (IUCN); “NT”, near threatened (IUCN); “S”, SPEC3 (BirdLife International).

<table>
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<td>Budgerigar</td>
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<td>Eurasian golden oriole</td>
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<td>6840</td>
<td>Streptopelia decaocto</td>
<td>Eurasian collared dove</td>
<td></td>
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<tr>
<td>6700</td>
<td>Columba palumbus</td>
<td>Common wood pigeon</td>
<td></td>
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<td></td>
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<td>Sturnus vulgaris</td>
<td>Common starling</td>
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(follow Table 2)

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<td><em>Parus palustris</em></td>
<td>Marsh tit</td>
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<td>Eurasian wren</td>
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<td>10010</td>
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<td>15980</td>
<td><em>Passer montanus</em></td>
<td>Eurasian tree sparrow</td>
<td>VU</td>
<td>11870</td>
<td><em>Turdus merula</em></td>
<td>Common blackbird</td>
<td></td>
</tr>
</tbody>
</table>

4.2 Relationships between species richness and abundance at landscape scale

One goal of this study was to establish how landscape environmental features affected the number of species and the abundance of birds (maximum number of individuals). Most of the recorded environmental variables presented correlations among them. For instance, park area was correlated with wooded surface, because biggest parks are most likely to have forested patches than central and smallest park. Biggest peri-urban parks were also more recent, while smallest central parks were usually oldest. The relationships among variables made it difficult to unravel their relative importance for birds. Due to the high collinearity of variables, we performed a RDA analysis and built the best linear model to explain our data using the RDA1 and RDA2 scores obtained for each point-count. Redundancy analysis was significant (P<0.01) and explained 89% of variation (Table 3). The first RDA component (RDA1) explained 88% of the variance described by the RDA. RDA1 was essentially represented by small parks with scarce wood cover, while RDA2 by younger parks (the scores of variables are shown in Table 3). Both the dependent variables (species’ richness and abundance) shown a negative relationship with component RDA1, while only birds’ abundance
presented a negative relationship with RDA2 (Fig. 2). Linear models built for species’ richness and birds’ abundance showed a significant correlation with RDA1 and RDA2 (P< 0.001, Table 4).

Table 3. Coefficients of environmental variables represented in RDA analysis of the relationships between birds and environmental features at the site-scale. Area and land cover types (trees, grasslands or other covers) have the highest influence on the first component of RDA, while the variable age of the park represents 60% of the second RDA component.

<table>
<thead>
<tr>
<th>Variable</th>
<th>RDA1</th>
<th>RDA2</th>
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</thead>
<tbody>
<tr>
<td>Area</td>
<td>-0.93</td>
<td>0.16</td>
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<tr>
<td>Unbuilt area around the park</td>
<td>-0.70</td>
<td>0.50</td>
</tr>
<tr>
<td>Age</td>
<td>0.22</td>
<td>-0.59</td>
</tr>
<tr>
<td>Wooded area</td>
<td>-0.87</td>
<td>-0.01</td>
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<tr>
<td>Grassland area</td>
<td>-0.88</td>
<td>0.17</td>
</tr>
<tr>
<td>Other coverings</td>
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<td>0.06</td>
</tr>
<tr>
<td>Water bodies occurrence</td>
<td>-0.73</td>
<td>0.07</td>
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</table>

Table 4. Results of the GMLs analysis for species richness and birds abundance performed on the variables RDA 1 and RDA 2 extracted from RDA analyses (A = analysis on species richness; B = analysis on birds abundance). NumDf = degrees of freedom in the numerator; DenDF = degrees of freedom in the denominator.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Estimate</th>
<th>NumDf</th>
<th>DenDf</th>
<th>F</th>
<th>P</th>
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<td>A. Species’ richness</td>
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<td></td>
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<tr>
<td>rda1</td>
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<td>50729</td>
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<tr>
<td>rda2</td>
<td>0.14</td>
<td>1</td>
<td>12</td>
<td>12036</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>B. Birds abundance</td>
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<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>rda1</td>
<td>-1.16</td>
<td>1</td>
<td>12</td>
<td>136932</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>rda2</td>
<td>-0.05</td>
<td>17</td>
<td>12</td>
<td>408.16</td>
<td>&lt;0.001</td>
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</table>

62
Figure 2. Constrained redundancy analysis showing the relationship between birds species richness and abundance and the environmental variables tested. Constraining variables are represented by blue arrows. Richness = number of bird species; Abundance = number of individuals; Wooded = surface covered by trees; Grassland = surface covered by grassland; Other lc = other coverings usually represented by paving or buildings; Water = presence of water bodies (rivers or lakes); Area = area of the park; Age = years from the establishment of the park; Buffer_unbuilt = unbuilt surface in a buffer of 1000 m around the park.

4.3 Relationships between species occurrence and abundance at site scale

Considering the relationships between birds presence and environmental features at site scale (Fig. 3, Table 5), the 14% of variation in species presence is explained by the variables considered (P<0.001). First component of the analysis (RDA_{bird1}) is mostly represented by surface covered by trees (Table 6) and explains 48% of variance described by RDA. Second component (RDA_{bird2}) is mostly represented by presence of water bodies and explains 26% of variance described by RDA. GAMs analysis reveals that two forest species (Eurasian blackcap (Sylvia atricapilla) and the Great tit (Parus major) presented a significant linear relationship with mean DBH (Fig. 4) in urban woods. In particular the maximum number of individuals decreases with increase in mean trees' diameter.
Figure 3. Constrained redundancy analysis showing the relationship between 28 species and environmental variables tested. Wooded = surface covered by trees; Grassland = surface covered by grassland; Other lc = other coverings; Water = presence of water bodies (rivers or lakes); Distance border = minimum distance from the park border. Constraining variables are represented by blue arrows. Group C* = central group* composed by the following species: *S. serinus, S. decaocto, A. caudatus, E. rubecula, A. apus, F. coelebs, C. monedula, P. krameri, D. urbica* and *F. tinnunculus*

Figure 4. Results of Generalized Additive Models for (a) the Eurasian blackcap (*Sylvia atricapilla*) (P <0.001) and (b) the Great tit (*Parus major*) (P<0.01).
Table 5. Species correlations with the RDA scores extracted by the RDA analysis on the relationships between birds presence and environmental features at site scale.

<table>
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<tr>
<th>Species</th>
<th>RDA1</th>
<th>RDA2</th>
<th>Species</th>
<th>RDA1</th>
<th>RDA2</th>
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<td>Ardea cinerea</td>
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<td>Parus caeruleus</td>
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<td>Picoides major</td>
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<td>0.02</td>
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<td>-0.04</td>
<td>Columba livia</td>
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<td>-0.06</td>
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<td>Hirundo rustica</td>
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<td>0.04</td>
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<td>Apus apus</td>
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<td>0.01</td>
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<td>0.33</td>
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Table 6. Biplot scores for constraining variables of the RDA analysis on the relationships between birds presence and environmental features at site scale.

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<td>grassland</td>
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<td>other lc</td>
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<td>water</td>
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<tr>
<td>dist from</td>
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</table>
5. Discussion

Birds provide a suitable method to explore urban effects and responses to different urban designs (Sanesi et al., 2009; Chace and Walsh, 2006). An essential first step to manage urban environments more effectively is a fuller understanding of the interplay between landscape (matrix effects) and local factors (patch effects) that affect urban biodiversity (Angold et al., 2006). Birds community observed in this study was clearly affected by environmental variables considered at different scale of analysis. It is known that parks surface is one of the features that mostly affect species abundance and richness: the biggest the park, the higher is the number of individuals that may contain and the higher is generally the heterogeneity of habitats within the park (Alvey, 2006; Cornelis and Hermy, 2004; Godefroid and Koedam, 2003). However, not always is feasible to establish large parks (i.e. in high-density urban context) or already existing green areas cannot be enlarged. Our results at both landscape and site level highlight how other environmental features describing the mixing of different land cover types are of fundamental importance (Table 3). In fact, both the three different land cover types (wooded, grassland and other coverings) have a great representation in the RDA component (Table 3) explaining the variance of birds observed. Smallest parks can contain relative high level of biodiversity when presence of wooded patches is guaranteed (while grassland and other covering are usually always present in this type of parks). Presence of water bodies (rivers or lakes, also of small dimension) is resulted to be highly influential at landscape scale (Table 3) and even more at local scale (Table 6). Water elements may favour aquatic but also non-aquatic species. At local scale, alongside some aquatic birds, also other species like Eurasian blackcap (Sylvia atricapilla) and Common nightingale (Luscinia megarhynchos) showed a preference for the presence of water (Fig. 3, Table 5). Among the parks we studied, there was one (Parco delle Cave) constituted of five restored quarries that represents an example of a mosaic (of rivers, lakes and wetlands, overall covering approximately 30 ha) well integrated in a peri-urban landscape. It is interesting to notice that despite it is a recent park (the year of establishment is 2002) Parco delle Cave contributes with five species (all of them are aquatic) to the total checklist of birds (Great reed warbler (Acrocephalus
arundinaceus), Eurasian coot (Fulica atra), Little bittern (Ixobrychus minutus), Little grebe (Tachybaptus ruficollis) and Eurasian reed warbler (Acrocephalus scirpaceus)). For this reason, this park may be considered as a hot spot for biodiversity in our study area. Also the amount of un-built areas around the park appears to favour both number of species and populations abundance. Urban tissue is a low-permeable matrix for the dispersal of animals and urban parks represent islands where they may find suitable habitats and resources. Biodiversity inside urban parks is favourite when the urban surrounding presents open-areas or other green infrastructure (i.e. street trees, private gardens). These patches may work as functional corridors or stepping-stones in order to favour the colonization and maintenance of species inside urban parks. Age of the park also resulted to influence bird community in our study area. Age is partially correlated with dimension and position of the parks: city central parks are usually older while peripheral and more extended parks are more recent. Alongside with this, oldest park have been often designed with a different conception, more prone to fit decorative and aesthetic needs than to reproduce the features of natural vegetation structure.

By analysing the effect of environmental variables at local scale, wooded land cover type and water bodies resulted to be of great importance for the overall avian community investigated. Also grassland lands and distance from park border (Table 6) influenced the presence of birds within the sampled points. Other features were less represented, meaning that their role in explaining the variance in species presence is negligible. Common starling (Sturnus vulgaris) and Barn swallow (Hirundo rustica) resulted influenced by presence of grasslands as well as the Italian sparrow (Passer italicae) which prefers sites with also presence of other coverings (i.e. buildings inside the park). The correlation between bird species richness and presence of water and wooded surface as well as grasslands and less natural areas emphasised the importance of urban green space containing heterogeneous elements capable of providing suitable habitats for a large number of species with different ecological requirements. In fact, the mixing of these different elements (when feasible) may lead even a single park (i.e. Boscoincittà, Parco Lambro), or few parks, to harbour high level of biodiversity. Among the species observed, a considerable portion (30%) is listed in some protection
list leading their presence of particular conservation interest. This result emphasizes the importance of the role that green urban spaces can have in support wildlife conservation.

In a previous study conducted in Milan it has been described how differences in structural attributes of forests support different abundance of birds’ population by build models based on distribution of trees diameter (Sanesi et al., 2009). By studying target species of birds in relation to DBH, Sanesi et al. (2009) provided evidence on the response of bird species to the presence of mature and heterogeneous forest stands.

Figure 5. Pictures taken from two different type of parks in our study area: (a) a typical forested environment in a peri-urban park (Parco Nord) and (b) big isolated trees in a central park (Parco Solari).

More in general, it is known how the maintenance of stand structural complexity is critical for forest biodiversity conservation (Lindenmayer et al., 2006) and old trees are demonstrated to be of great importance for some species and for biodiversity in general (Cowie and Hinsley, 1988; Andersson and Östlund, 2004). In this study, we tested if forests species abundance have a relationship with the presence of mature trees (high mean DBH). Only two species on seven investigated presented a significant relationship (Eurasian blackcap, *Sylvia atricapilla* and the Great tit, *Parus major*): number of individuals observed decreased with the increase of mean trees diameter, suggesting an opposite trend compared to what observed in other studies. However, the vegetation structure of the urban
parks studied presents some peculiarities due to vegetation management that differ from natural forest patches. In fact, oldest and biggest trees in urban parks are usually distributed with low density, when not even isolated. While forest patches normally present few big trees and many medium and little trees at high densities (see Fig. 5).

In Fig. 6 it is shown how point-counts belonging to forested environments (boxplot on the left) have lower values of mean DBH than point-counts with no-forest cover (less than 20%) that have instead higher values of DBH. Forest species resulted to be associated with small trees and small trees resulted more frequent in forested areas.

![Figure 6. Boxplot of the differences between trees DBHs in forested patches (tree cover > 60% of the point area) and trees DBHs in unforested patches (tree cover ≤ 20% of the point area).](image)

However, considering the importance of old trees that has been demonstrated in previous studies (Andersson and Östlund, 2004; Wells et al., 1998; Berg et al., 1994), their presence in the studied forested patches would probably further increase species presence and should be promoted. In fact, the presence of old trees in isolated exemplary or with very low-density stands may be not sufficient to promote biodiversity by itself because of difficult exploitation from bird forest species. Bird species
richness in urban ecosystems is influenced by both local and landscape characteristics in different ways and a multi-scale approach is essential to its proper management (Savard et al., 2000). Within urban ecosystems, actions taken to preserve or enhance biodiversity should be promoted and managed at scales ranging from individual plants to the entire city itself and even surrounding areas in order to integrate different environmental requirements and effects. The concerted efforts at various scales can produce the best results (Goddar et al., 2010; Savard et al., 2000; Poiani et al., 2000) and conservation actions that neglect to consider the interplay relations between landscape and local features may fail or produce powerless effects on biodiversity conservation.

Enhancement of biodiversity in urban ecosystems can have a positive impact also on the quality of life and education of urban dwellers and thus facilitate the preservation of biodiversity in natural ecosystems (Savard et al., 2000) because of increase in awareness and sensitivity to environmental issues (Rohde and Kendle, 1994; Sebba, 1991). Some authors suggest that a challenging strategy to address the growing conflict between cities and biodiversity is to make urban growth compatible with biodiversity protection (McDonald et al., 2008), minimizing the conflict between people and nature at the urban-wildlands interface (Goldstein et al., 2006). Ecosystem services derived from urban forests improve resilience and quality of life in cities (Gómez-Baggethun et al., 2013) and social as well as ecological benefits will be gained through biodiversity protection. Undertake good practices of management of green spaces in the urban ecosystem is therefore a great strategy for the sustainability of human development, for cities resilience and for biodiversity conservation.

6. Conclusions

Our study shows that, despite traditional attitude would consider all green spaces associated with urban environment mostly designed to be exploited by citizen for recreational activities, urban forests can (and must) at the same time provide suitable habitats for wildlife fauna. Urban green areas can therefore be actively managed by foresters and city planners to preserve the biological diversity that they harbour. Local actions as well as regional actions are equally important on their respective scale
(Savard et al., 2000) and our study shows how different environmental requirements exist for biodiversity maintenance at different scales, which need therefore different management strategies. The results presented in this paper provide suitable information for implement an effective management of green spaces with the aim of support high level of biological diversity within urban environment. Undertake consistent conservation actions would have the double result to preserve biodiversity and to provide opportunity for residents to experience nature and its diversity.

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Chapter III

Diversity and hydrocarbon-degrading potential of epiphytic microbial communities on *Platanus x acerifolia* leaves in an urban area

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Abstract

Plants and their associated bacteria have been suggested to play a role in air pollution mitigation, especially in urban areas. Particularly, epiphytic bacteria might be able to degrade atmospheric hydrocarbons. However, phyllospheric bacterial communities are highly variable depending on several factors, e.g. tree species, leaf age and physiology, and environmental conditions. In this work, bacterial communities hosted by urban Platanus x acerifolia leaves were taxonomically characterized using high throughput sequencing of 16S rRNA gene, and their temporal and spatial variability was assessed by comparing samples collected from different locations in the city of Milan (Italy) and in different months. The diversity of alkane hydroxylase (alkB) phylotypes harboured by phyllospheric bacteria associated to urban Platanus trees was also evaluated. Results revealed that temporal changes, which are related to seasonality, acted as a stronger driver both on Platanus phyllospheric community structure and on alkB phylotype diversity than sampling location. Biodiversity of bacterial communities decreased along the growing season, leading to a strong dominance by the genus Stenotrophomonas. On the contrary, diversity of hydrocarbon-degrading populations increased over the months, although it resulted lower than that reported for other habitats. It was therefore hypothesized that atmospheric hydrocarbons might play a key role in the selection of phyllospheric populations in urban areas.

1. Introduction

Air pollution is a matter of global concern, especially in urban areas, due to the harmful effects of atmospheric pollutants on human health and on the environment. Current emission reduction methods and mitigation strategies are not adequate to fully meet the World Health Organization (WHO) guidelines for air pollutants (Ali et al., 2012; Weyens et al., 2015). Since most of the environmental problems in urban areas are generated at local level, often one of the most effective ways to deal with
them is through local solutions (Bolund and Hunhammer, 1999). Plants have been suggested to effectively contribute to reduce air pollution levels and offsetting greenhouse gas emissions in cities (Beckett et al., 1998; Dzierzanowski et al., 2011; McPherson et al., 1998; Nowak and Crane, 2002; Nowak et al., 2006; Paoletti, 2009; Redford et al., 2010; Yang et al., 2005; Zhao et al., 2010). In this context, the regulation of ecosystem services (the direct and indirect contributions of ecosystems to human well-being (TEEB, 2011)) provided by vegetation in urban areas is of great importance (Baró et al., 2014). Many studies, in fact, indicated that the management of urban forests to enhance ecosystem service supply can be a cost-effective strategy to meet specific environmental standards or policy targets (Escobedo et al., 2010, 2011).

Furthermore, it has been recognized that also plant-associated bacteria can play a crucial role in air bioremediation processes (Glick, 2015; Weyens et al., 2015). Particularly, the aerial parts of terrestrial plants, mainly leaves (i.e. the phyllosphere) host huge amounts of bacteria. In fact, although phyllospheric microorganisms comprise also fungi, yeasts, algae, protozoa and nematodes, bacteria are by far the most abundant inhabitants of leaf surfaces (Lindow and Brandl, 2003). Since phyllospheric bacteria are often found at an average of $10^6$-$10^7$ cells cm$^{-2}$ of leaf surface (Lindow and Brandl, 2003), the planetary phyllospheric bacterial population has been estimated to be as large as $10^{26}$ cells (Morris and Kinkel, 2015). Among them, epiphytic bacteria, which primarily live on leaf surfaces, are directly positioned to the interface with the atmosphere. Thus, they are exposed to several detrimental factors such as UV radiation, desiccation, severe temperature changes and, especially in urban areas, atmospheric pollutants (Lindow and Brandl, 2003). For this reason, they are expected to have developed metabolic abilities towards atmospheric hydrocarbons and therefore to play a potential role in air bioremediation processes. Indeed, several papers have already reported the ability of phyllospheric bacteria to degrade aliphatic (Al-Awadhi et al., 2012) and aromatic hydrocarbons, namely phenolic compounds, toluene, xylene and phenanthrene (De Kempeneer et al., 2004; Sandhu et al., 2007; Sangthong et al., 2016; Scheublin et al., 2014; Waight et al., 2007; Yutthammo et al., 2010).
Despite their continuous exchange with airborne populations (Lighthart, 2006; Lymeropoulou et al., 2016; Whipps et al., 2008), phyllospheric bacteria are not random assemblages but they rather form actual communities. In fact, some bacterial taxa are recurrently retrieved from leaf-associated habitats, leading to the hypothesis that, after recruitment, they undergo some selection processes (Delmotte et al., 2009; Rastogi et al., 2013; Vorholt, 2012; Yang et al., 2001). The relative abundance of a specific bacterial taxon in phyllospheric communities, however, can vary considerably. The main drivers that were suggested to shape community structure include host plant species, leaf age and physiology, season, geographical location, and environmental factors, such as solar radiation, humidity and nutrient availability (Laforest-Lapointe et al., 2016; Müller and Ruppel, 2014; Peñuelas et al., 2012; Rastogi et al., 2012; Redford et al., 2010; Vokou et al., 2012), Interactions between these factors can also affect bacterial communities. For example, Wagner and colleagues (2016) suggested that the plant genotype-by-sampling site interaction was a stronger driver than plant genotype only. Moreover, the occurrence of a contribution from stochastic processes was also observed (Maignien et al., 2014). Therefore, due to the high variability of phyllospheric community structure, a more profound knowledge about bacterial communities hosted by different plant species in different environments is needed to assess their potential contribution to air bioremediation. Among plant species that can be typically found in urban areas, Platanus trees are widespread in most cities of central and southern Europe. They are frequently planted along high traffic roads, since they are known to be considerably resistant to stresses caused by urban pollution (Yang et al., 2015). To the best of our knowledge, bacterial communities associated to Platanus leaves were characterized only by Zhang et al. (2015), who however limited their research to the assessment of functional diversity of the culturable fraction.

The aims of this work were: (i) a deep phylogenetic characterization of bacterial communities hosted by urban Platanus x acerifolia leaves using high-throughput sequencing (HTS) methods; (ii) an evaluation of the diversity of alkane hydroxylase (alkB) phylotypes harboured by phyllospheric bacteria associated to urban P. x acerifolia trees; (iii) the assessment of temporal and spatial
variability of bacterial phyllospheric communities associated to *P. x acerifolia* trees located in different areas of the city of Milan (Italy) and sampled in different months.

2. Materials and Methods

2.1 Sampling

Leaves were collected from eight different *Platanus x acerifolia* trees in the city of Milan (Italy). Four of them were located in an urban park (Parco Nord), next to a low-traffic secondary road, and the other four were planted along a high-traffic road (Viale Fulvio Testi), which is one of the major arterial roads in the northern part of the city (Fig. 1). Meteorological conditions and atmospheric pollutant concentrations for this area are reported in Fig. S1. Sampling was performed at the beginning (April 17, 2014) and in the middle of the growing season (July 11, 2014). For each tree, samples were collected in triplicates, for a total of 48 samples. Each sample was composed by three young leaves in April and by two mature leaves in July, collected at a height ranging approximately between 1.50 and 2.00 m. Leaves were handled with metal scissors and tweezers rinsed with ethanol and immediately put in sterile 120 mm Petri dishes to prevent DNA contamination from external sources.

Fig. 1 – Map of sampling locations.
2.2 DNA extraction

Total DNA of epiphytic bacteria was extracted with FastDNA Spin for Soil kit (MP Biomedicals, Solon, OH, USA). Leaves were thoroughly rinsed in sterile Petri dishes with approximately 4 mL of Sodium Phosphate Buffer supplied with the kit under a laminar flow hood. After rinsing, it was possible to recover approximately 2 mL of the used buffer. It was collected from the Petri dish with a micropipette and placed in the kit Lysing Matrix E Tube. Further steps were performed according to manufacturer's instructions.

2.3 Illumina sequencing

The V5-V6 hypervariable regions of the bacterial 16S rRNA gene were PCR-amplified using 783F and 1046R primers (Huber et al., 2007; Wang and Qian, 2009). For the characterization of alkB diversity, three different primer pairs were preliminarily tested on our samples (pairs (d), (e) and (f) of Jurelevicius et al. (2013)); detectable amplification was obtained with primer pair (f) only, which was therefore chosen for subsequent analyses. At the 5' end of each primer, a 6-bp barcode was included to allow sample pooling and sequence sorting. All amplicons were sequenced by MiSeq Illumina (Illumina, Inc., San Diego, CA, USA) with a 250 bp × 2 paired-end protocol. For each sample, 2 × 75 µL volume PCR reactions were performed with GoTaq® G2 Green Master Mix (Promega Corporation, Madison, WI, USA) and 1 µM of each primer. The cycling conditions for the amplification of the 16S rRNA gene fragment were: initial denaturation at 94 °C for 4 min; 28 cycles at 94 °C for 50 s, 47 °C for 30 s, and 72 °C for 30 s and a final extension at 72 °C for 5 min. The cycling conditions for the amplification of the alkB fragment were: initial denaturation at 96 °C for 4 min; 40 cycles at 96 °C for 45 s, 47 °C for 30 s, and 72 °C for 45 s and a final extension at 72 °C for 5 min. The amplicons were purified with the Wizard® SV Gel and PCR Clean-up System (Promega Corporation, Madison, WI, USA) and purified DNA was quantified using Qubit® (Life Technologies, Carlsbad, CA, USA). Groups of 9/12 amplicons bearing different barcode pairs were pooled together to build a single library. Further library preparation with the addition of standard
Nextera indexes (Illumina, Inc., San Diego, CA, USA) and sequencing were carried out at Parco Tecnologico Padano (Lodi, Italy).

2.4 Sequence analysis

Reads from both 16S rRNA and \textit{alk}B genes sequencing were demultiplexed according to the indexes. Uparse pipeline was used for the following elaborations (Edgar, 2013). In case of 16S rRNA genes, forward and reverse reads were merged with perfect overlapping and quality filtered with default parameters. Conversely, since \textit{alk}B reads were not overlapping, only one read was analysed. Suspected chimeras and singleton sequences (i.e. sequences appearing only once in the whole data set) were removed. Phylotypes were defined on the whole data set clustering the sequences at a 97% of similarity and defining a representative sequence for each cluster. Representative sequences of 16S rRNA gene phylotypes (Operational Taxonomic Units – OTUs) were classified using SINA with SILVA database (Pruesse et al., 2012) and sequences not classified as belonging to Bacteria domain (i.e. Archaea, chloroplasts and mitochondria) were discarded. Abundance of each OTU was estimated by mapping the sequences of each sample against the remaining OTU representative sequences at 97% of similarity. Representative sequences of \textit{alk}B gene phylotypes were translated into aminoacid sequences considering the proper frame and annotated with Blastp (Altschul et al., 1990). Sequences not annotated as \textit{alk}B were discarded; sequences of each sample were then mapped against the remaining representative phylotype sequences at 97% of similarity.

To assess the spatial and temporal variability both of the structure of phyllospheric bacterial communities hosted by \textit{Platanus} leaves and of \textit{alk}B phylotypes, samples were grouped according to their sampling location (urban park or high-traffic road) and to sampling month (April or July). Non-metric Multidimensional Scaling (NMDS) analyses based on Hellinger distances were performed using R (Vegan package) (Oksanen et al., 2009). Differences in abundance of the most abundant genera (≥ 2%) between months (April and July) or locations (park and road) were tested by t-tests. P-values were
corrected for multiple testing according to the False Discovery Rate (FDR) procedure (Benjamini and Hochberg, 1995) using the MULTTEST package in R.

3. Results and Discussion

3.1 Phylogenetic diversity

From NMDS analysis, two main groups could be identified, corresponding to the April and July samples respectively (Fig. 2). Within each of the two sampling months, samples from the urban park and from the high-traffic road were close but clearly distinguishable. Therefore, it can be hypothesized that temporal changes, which are in turn related to seasonality, acted as a stronger driver on the *Platanus* phyllospheric community structure than sampling location.

![Fig. 2 – NMDS analysis of bacterial phylogenetic diversity. Hellinger distances among samples were calculated on the basis of presence and abundance of OTUs. Empty symbols: April; filled symbols: July; squares: park; triangles: road.](image)
This is in agreement with the observations of several authors, e.g. Copeland et al. (2015), Rastogi et al. (2012) and Peñuelas et al. (2012), which identified seasonal changes as a major factor shaping bacterial phyllospheric communities associated to different plant species. Furthermore, environmental conditions and atmospheric pollutant concentrations are known to be substantially homogeneous in the Po Valley, where Milan is located; therefore, this area as a whole is generally considered as a pollutant hot-spot (Marcazzan et al, 2002; Maurizi et al., 2013; Vecchi and Valli, 1999). For this reason, it can be hypothesized that environmental variables may have been not sufficiently different at the two sites, which are approximately 2 km from each other, to cause appreciable dissimilarities in bacterial community composition.

The relative abundance of the main bacterial phyllospheric populations at the taxonomic levels of Class and Genus is shown in Fig. 3 (a and b, respectively). Overall, the most abundant classes were *Gammaproteobacteria, Alphaproteobacteria, Betaproteobacteria, Bacilli* and *Actinobacteria*. They have already been described by several authors as common classes in phyllospheric bacterial communities associated with different plant species (Dees et al., 2015; Rastogi et al., 2012; Vorholt, 2012; Whipps et al., 2008), although in some cases they were reported with very different relative abundances (Redford et al., 2010). In April, the *Platanus* phyllospheric communities were not clearly dominated by any class or genus. On the contrary, July communities exhibited a large prevalence of *Gammaproteobacteria*, with a relative abundance of approximately 50%. Within this class, most sequences belonged to the genus *Stenotrophomonas* (approximately 42% of total bacteria). This genus has already been reported to be one of the major genera commonly detected in phyllospheric communities, although at much lower percentages (Vorholt, 2012). Particularly, *Stenotrophomonas* has been generally described as a member of endophytic, rather than epiphytic, bacterial communities of different plant species (Ferrando and Fernández Scavino, 2015; Kgomotso et al., 2015; Mastretta et al., 2009; Romero et al., 2014; Taghavi et al., 2009). Several isolates belonging to this genus were demonstrated to possess plant-growth promoting properties (Calciolari and Silva, 2013; Islam et al., 2015). The same abilities were observed for a rhizospheric *S. maltophilia* strain and confirmed
through genome sequencing (Wu et al., 2015). Furthermore, some plant-associated *Stenotrophomonas* strains were reported as able to degrade oil hydrocarbons (Ali et al., 2012) and phenanthrene (Muratova et al., 2015). In a culture-independent study on endophytic communities of *Cucurbita pepo*, members of genera *Stenotrophomonas* and *Sphingomonas* showed a significantly higher abundance in the presence of DDE, the most common and persistent degradation product of the pesticide DDT, than in the absence of the molecule (Eevers et al., 2016). Thus, it can be hypothesized that the genus *Stenotrophomonas* may play a key role also in the ecology of phyllospheric communities associated to urban *Platanus* leaves. Table S1 reports the results of multiple t-tests on abundant genera that significantly varied between months. The genus *Hymenobacter* was identified as significantly more abundant in July phyllospheric communities, with average relative abundances of 11.1% and 4.3% in park and road samples, respectively. Some members of this genus have been described as radiation tolerant (Kim et al., 2016; Lee et al., 2014; Su et al., 2014) and psychrophilic or psychrotolerant (Klassen and Foght, 2011; Mi et al., 2014). Due to these features, it can be hypothesized that these bacteria may undergo a selection process, throughout the growing season, by the harsh conditions of the phyllospheric environment. Given the continuous exchange of bacterial populations between leaf surface and air, and the shared characteristics of high UV radiation and low temperature of the two environments, it is not surprising that the genus *Hymenobacter* was also reported in outdoor airborne communities (Fahlgren et al., 2011; Yooseph et al., 2013). The other genus identified as significantly more abundant in July samples was *Massilia* (Table S1). Members of this genus have already been described as commonly retrieved in phyllospheric epiphytic communities (Rastogi et al., 2013, 2012), as well as endophytes (Croes et al., 2015; Thijs et al., 2014). Therefore, it may have been enriched over time due to the selective conditions of the phyllospheric environment, which could favour it over other genera.
Fig. 3 – Relative abundance of phyllospheric bacterial taxa at Class (a) and Genus (b) level. Only taxa with an abundance ≥ 1% (Class) or ≥ 2% (Genus) in at least one of the four groups of samples are shown. Samples are grouped according to month and sampling location.

The genus *Buttiauxella* was the only one to be recognized as significantly more abundant in park samples, with average abundances of 8.3% and 2.0% in April and July samples, respectively (Table
S2). It is not reported to be one of the most common genera among phyllospheric bacteria (Bulgarelli et al., 2013; Vorholt, 2012). However, some *Buttiauxella* sp. strains were previously cultivated from atmospheric particulate matter (Fang et al., 2007; Gandolfi et al., 2011). On the contrary, the only genus identified as significantly more abundant in road communities was *Aeribacillus* (Table S2). Members of this genus have been often described as thermophilic bacteria, isolated from hot springs, geothermal reservoirs and different environments of sub-tropical areas (Aanniz et al., 2015; Filippidou et al., 2015; Yanmis and Adiguzel, 2014). Moreover, some strains can produce exopolysaccharides as a way to survive high temperatures (Radchenkova et al., 2013; Zheng et al., 2012). These features can possibly be also useful to deal with locally very high temperatures on leaf surfaces exposed to solar radiation.

Among the other most abundant genera, as reported in Fig. 2b, *Sphingomonas, Arthrobacter, Methylobacterium, Pseudomonas, Pantoea, Rhodococcus* and *Flavobacterium* have already been retrieved in phyllospheric environments (Delmotte et al., 2009; Maignien et al., 2014; Rastogi et al., 2013, 2012; Vorholt, 2012). Thus, a “core” of phyllospheric bacterial communities appears to exist (Laforest-Lapointe et al., 2016), although the relative abundance of each genus can show high variability both in different plant species and in different individuals of the same plant species (Bulgarelli et al., 2013).

The average number of OTUs detected in April samples was significantly higher than that in July samples (Fig. S2). Moreover, genera that were less abundant than 2% in all the four sample groups, indicated as “Others” in Fig. 2b, together constituted approximately 61% and 26% of April and July communities, respectively. Thus, the diversity of bacterial communities of young leaves appeared to be higher than that of the communities hosted by older leaves, as already observed by several authors (Copeland et al., 2015; Dees et al., 2015; Lindow and Brandl, 2003). This phenomenon is generally explained by a selection effect on biodiversity, which is due both to harsh environmental conditions typical of the phyllospheric habitat and to the plant characteristics determined by its genotype (Whipps et al., 2008). It has also been suggested that seasonality and/or leaf maturation may
determine a progressive decrease of nutrient availability (Dees et al., 2015), thus decreasing the number of bacterial populations that can be sustained. Nevertheless, this trend, although widespread, can not be considered to be the general rule, since in some cases phyllospheric communities remained stable over time (Delmotte et al., 2009), or even an increase in the richness of epiphytic bacteria was observed with increasing time of colonization (Peñuelas et al., 2012). Moreover, Laforest-Lapointe and colleagues (2016) observed that phyllospheric communities of five tree species in Canada underwent a succession during the growing season, although plant species was a stronger driver on bacterial diversity than sampling time. Therefore, more research is needed in order to better describe time-dependent shifts in phyllospheric community structures of an extensive range of plant species. This is particularly important for perennial plants, which can undergo a wide variability of climatic conditions throughout the year, especially in temperate areas.

3.2 Diversity of alkB phylotypes

In addition to the phylogenetic-based community structure, knowledge about potential metabolic abilities of phyllospheric bacteria and their functional diversity are of critical importance to assess their possible contribution to air remediation. Zhang et al. (2015) evaluated the carbon substrate utilization pattern through the BIOLOG method, in order to estimate the functional diversity of bacteria associated to leaves of urban trees in China, including a species of Platanus (P. orientalis). They found that phyllospheric communities associated with different trees significantly differed in their metabolic abilities. However, this method relies on laboratory cultivation. Thus, results are limited to the culturable fraction of bacterial communities. For this reason, it would be also necessary to explore a range of suitable marker genes in phyllospheric metagenomes. Up to now, only chiA, encoding a chitinase, was extensively studied through amplicon HTS (Cretoiu et al., 2012). More comprehensive approaches were chosen instead, in order to identify the main metabolic adaptations to phyllospheric life: shotgun metagenomic sequencing was applied to bacterial communities hosted by Tamarix aphylla leaves (Finkel et al., 2016) while metaproteomics was used on soybean, clover
and *Arabidopsis thaliana* communities (Delmotte et al., 2009). In this work, alkane hydroxylase (*alkB*) was selected as reference gene to roughly estimate the diversity of *Platanus* phyllospheric bacteria possessing the potential ability to degrade alkanes. Diversity of alkane hydroxylases has already been studied in the rhizosphere of different tree and grass species, both in isolates (Fatima et al., 2015; Tesar et al., 2002; Yousaf et al., 2010) and in whole bacterial communities through culture-independent methods (Mukherjee et al., 2015; Tsuboi et al., 2015). However, a characterization of the diversity of *alkB* phylotypes in phyllospheric communities is still lacking. From NMDS analysis, April and July samples were clearly distinguishable (Fig. 4). Moreover, while samples from the two sampling locations formed two separate groups in April, in July they showed a high overlapping. Therefore, although *alkB* phylotypes were different in the two sampling locations at the beginning of the growing season, they became highly similar over time. As already observed for phylogenetic diversity, it can be hypothesized that both environmental conditions such as temperature, humidity and solar exposure, and pollution levels were probably similar at the two locations. Thus, not only bacterial communities considered as a whole, but also hydrocarbon-degrading populations could have been subjected to the same selection drivers regardless the sampling location. However, in contrast with what observed for phylogenetic biodiversity, the number of *alkB* phylotypes was significantly higher in July (Fig. S3). This led to put forward the hypothesis that atmospheric hydrocarbons might play a key role in the selection of phyllospheric populations in urban areas. In fact, the selective pressure they exert would cause a decrease in phylogenetic diversity while increasing the diversity of hydrocarbon-degrading populations.
Fig. 4 – NMDS analysis of alkB diversity. Hellinger distances among samples were calculated on the basis of presence and abundance of OTUs. Empty symbols: April; filled symbols: July; squares: park; triangles: road.

The overall number of detected alkB phylotypes was 3036. A phylogenetic tree was built with the 51 phylotypes with a total abundance \( \geq 0.3\% \) (Fig. 5). Most of these phylotypes clustered together, and showed high similarities with alkB from different species of the genus *Rhodococcus*, particularly with *R. aetherivorans*, and with *Mycobacterium smegmatis*. Other 18 out of 51 phylotypes, which formed a separate cluster, revealed their best similarity with uncultured bacteria from various molecular studies. Although there are no indications on the taxonomy of these uncultured bacteria, the cluster to which they belong appears to be nearer to that including *Rhodococcus* sequences than to other reference strains. However, when comparing this cluster with sequences reported in a comprehensive alkB tree that was recently published, it was not possible to clearly identify its position on it (Nie et al., 2014). Conversely, only one of the considered phylotypes was highly similar to alkB belonging to a Gammaproteobacteria genus, i.e. *Shewanella*. The high prevalence of sequences from
Actinobacteria suggests the mainly terrestrial origin of potential alkane-degrading bacteria (Nie et al., 2014). However, the overall diversity of alkB phylotypes in bacterial communities hosted by *Platanus* leaves, although increased over time as observed above, appears to be still lower than that reported for other habitats (Nie et al., 2014). This may be possibly due to the harsher conditions in the phyllospheric environment than in other environments, which limit biodiversity.

Fig. 5 (following page). Phylogenetic tree of alkB phylotypes based on nucleotide sequence. Only phylotypes with a total abundance ≥ 0.3% were included. Sequences of alkB from some reference strains and from uncultured bacteria having a high similarity with phylotypes of this work were also included for comparison. The tree was built with the UPGMA method using MEGA7. The percentage of replicate trees in which the associated taxa clustered together in the bootstrap test (1000 replicates) are shown next to the branches. The evolutionary distances were computed using the Maximum Composite Likelihood method and are in the units of the number of base substitutions per site.
4. Conclusions

A proper management of vegetation has been suggested to be a promising strategy to decrease air pollution in urban areas. However, our understanding of the potential effectiveness of urban plants in air quality improvement is still affected by several uncertainties. Therefore, we need at least to be able to estimate the actual involvement of plants, and of plant-phyllospheric bacteria associations, in air pollutant removal.

On *Platanus x acerifolia* leaves, biodiversity of bacterial communities decreased along the growing season, while the diversity of hydrocarbon-degrading populations increased. This phenomenon might indicate that, in the phyllosphere of urban plants, selection effects on bacteria are driven more strongly by atmospheric hydrocarbons than by other environmental factors, such as temperature, humidity or solar radiation. However, the actual ability of phyllospheric bacterial communities to degrade hydrocarbons *in situ* still needs to be confirmed. Therefore, future research should be aimed at the quantification of the actual contribution of bacteria in air pollutant removal per unit of leaf weight or leaf area under different environmental conditions, and at the evaluation of the efficiency of different plant-bacteria systems in air quality improvement.

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Appendix of Chapter III
Supplementary Materials

Table S1. Genera which significantly varied between sampling months on the basis of multiple t-tests (p < 0.05). Only genera with an abundance ≥ 2% are reported. Significance of the obtained p-values was adjusted according to the False Discovery Rate procedure with Benjamini-Hochberg correction.

<table>
<thead>
<tr>
<th>Genus</th>
<th>t</th>
<th>Corrected p-value</th>
<th>Prevalence</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aeribacillus</td>
<td>2.214970</td>
<td>0.001767601</td>
<td>April</td>
</tr>
<tr>
<td>Buttiauxella</td>
<td>3.868329</td>
<td>0.001767601</td>
<td>April</td>
</tr>
<tr>
<td>Hymenobacter</td>
<td>-3.742334</td>
<td>0.004272642</td>
<td>July</td>
</tr>
<tr>
<td>Massilia</td>
<td>-2.420413</td>
<td>0.018212689</td>
<td>July</td>
</tr>
<tr>
<td>Methylobacterium</td>
<td>1.509056</td>
<td>0.027313227</td>
<td>April</td>
</tr>
<tr>
<td>Pseudomonas</td>
<td>2.671517</td>
<td>0.037046296</td>
<td>April</td>
</tr>
</tbody>
</table>

Table S2 – Genera which significantly varied between sampling sites on the basis of multiple t-tests (p < 0.05). Only genera with an abundance ≥ 2% are reported. Significance of the obtained p-values was adjusted according to the False Discovery Rate procedure with Benjamini-Hochberg correction.

<table>
<thead>
<tr>
<th>Genus</th>
<th>t</th>
<th>Corrected p-value</th>
<th>Prevalence</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aeribacillus</td>
<td>-1.4962479</td>
<td>0.0000004065</td>
<td>road</td>
</tr>
<tr>
<td>Buttiauxella</td>
<td>5.7981095</td>
<td>0.019961550</td>
<td>park</td>
</tr>
</tbody>
</table>
Fig. S1. Meteorological data and atmospheric pollutant concentrations from 1st January 2014 to 31st December 2014 in Milan, Italy (source: Regional Agency of Environmental Protection, ARPA Lombardia). All data were retrieved from the nearest available sensors to the leaf sampling locations. Meteorological parameters (mean daily temperature, mean daily global solar radiation, cumulative daily precipitations, mean daily relative humidity, mean daily wind speed) were obtained from the ARPA Lombardia sampling point “Cinisello Balsamo Parco Nord”. Atmospheric pollutant concentrations (PM10 mean daily concentration, benzene mean daily concentration) were obtained from the ARPA Lombardia sampling point “Milano Pascal Città Studi”. The two days of sampling are shown on the graph as vertical black lines.
Fig. S2 – Boxplot of average OTU number in April and July samples.

Fig. S3 – Boxplot of average number of alkB phylotypes in April and July samples.
Chapter IV

Urban soils properties and Organic Carbon stock in the urban area of Milan

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in preparation for Landscape and Urban Planning
Abstract

As urban areas dramatically increase globally, more studies on the effects of urbanization on C storage by urban soils are urgently needed. Urban areas, besides being major producers of atmospheric CO$_2$ emissions, may have a great carbon sequestration potential. However, they are normally excluded from the C stock estimation. Carbon sequestration capacity of urban soils is extremely variable depending on several natural and anthropogenic factors. In order to enhance the accuracy of C stock estimates, in this study we: i) describe the characteristics of urban soils in the study area; ii) estimate the organic C in the topsoil (0-40 cm) iii) test the influence of different environmental variables on C stocks and iv) explore the possibility to define the typical carbon stocks of different urban land cover and typology of areas for the urban area of Milan. The results showed high variability in soil properties and OC stock. Mean C stock found for the urban soils of Milan was comparable to values found for other soils (mainly agriculture soils) by previous studies. C stock was found higher for parks compared to other areas, but no clear differentiation has been found in order to define a typical C stock for each land cover or use type. The complex heterogeneity of urban soils together with the uncertainty of their origins, their history and management may summarize the reasons of the limitation in our capacity to model their characteristics.

1. Introduction

1.1 Carbon sequestration by soil

The atmospheric carbon dioxide (CO$_2$) concentration passed previous record every year, recently exceeding 400 ppm (2013) (Bae et al., 2014) and there is an urgent need to identify strategies of stabilizing atmospheric concentration of CO$_2$ (WMO, 2006). Globally, the amount of CO$_2$-C emitted into the atmosphere is estimated at 136 ± 55 Pg (1 Pg = 10$^{15}$ g) from terrestrial ecosystems, of which emission from world soils is estimated at 78 ± 12 Pg (Lal, 2008). One of the two principal sources of the increase in CO$_2$ concentration is attributed to land use conversion and deforestation (the other source is fossil fuel combustion) which presently contribute 0.6 to 2.5 Pg C/yr (Lal, 2008). Organic
carbon stock provided by soil (SOC) is a key component in carbon capture and storage to mitigate the increasing atmospheric CO\textsubscript{2} concentration and to regulate climate change (Morel et al., 2015, Ingram & Fernandes, 2001; Lal, 2004). In fact, to effectively offset the C emissions for climate change mitigation, C sequestration implies that C must remain stored not just for 100 years, but probably for more than 10,000 years due to the long atmospheric residence time of some of the emitted C (Lorenz and Lal, 2015, Hansen et al., 2013, Hutyra et al., 2014) and this long-term terrestrial C storage occurs in particular in soils (Lorenz and Lal, 2015). Moreover, soil is the largest terrestrial C pool in the biosphere, storing 1.5 to 3 times more C compared to that stored in vegetation (Wang et al., 2004) or in the atmosphere. Conversion of natural to managed ecosystems, and susceptibility to soil degradative processes can lead to depletion of soil organic carbon pool with attendant release of CO\textsubscript{2} and other GHGs into the atmosphere (Lal, 2008). One of the most important recent research themes of international interest is the anticipated change in terrestrial carbon stock under changing land use and climate (Taalab et al., 2012; Zaehle et al., 2007). Urban land-use and land-cover changes can directly alter above- and belowground biomass C, and soil C that is comprised of soil inorganic carbon (SIC) and soil organic carbon (SOC) (Lorenz and Lal, 2015).

Urban areas are increasing in extent at a greater pace than any other land-use type and the resulting impacts on ecosystem services, including on soil organic carbon stocks, remain poorly characterised (Edmondson et al., 2012). Soil organic matter is a key component of soil, influencing a number of ecosystem services including, apart from the cited carbon storage also nutrient cycling, water retention, soil structure and erosion (Hester and Harrison, 2010). However, despite its fundamental role for many ecosystem services, the majority of the studies on C sequestration in urban areas focused on net primary production by the urban vegetation despite the potential of urban soils to store large amounts of C and thus to cope with the great challenges met by cities (Morel et al., 2015; Lorenz and Lal, 2009; Pouyat et al., 2006). For example, Mestdagh et al., (2005) found that SOC stocks of grassy roadsides, waterways, and railways in urban areas accounted for 15% of the total SOC stocks
in a city. Moreover, also the soils beneath impervious surfaces in urban areas offered another, often overlooked, source of SOC (Edmondson et al., 2012; Raciti et al., 2012).

1.2 Urban soils

Urban soils face the paradox of being of highest interest regarding property and building issues, and being almost totally ignored with regard to consideration of its functions and roles for the management of urban ecosystems (Morel et al., 2015). Soils of urban areas may be strongly affected by human activities (de Kimpe and Morel, 2000). Human activities related to urban development are associated with the construction of artificial soils, sealing of natural soils, and extraction of material normally not affected by surface processes (IUSS Working Group WRB, 2006). Urbanization influences the natural soil template through disturbance, shifts in resource availability, and alterations in physicochemical conditions, resulting in urban ecosystems that are fundamentally distinct from their nonurban counterparts (Pavao-Zuckerman, 2008; Kaye et al. 2006). Their composition and functions and, thus, their ability to provide ecosystem services resulted in turn different from those of natural soils and often impaired (Morel et al., 2015). Soils of urban areas are mostly considered two-dimensionally: they are a surface that can harbor human activities, while their three-dimensional extension and functions are generally ignored by urban planning and management (Morel et al., 2015). Despite the poor attention that has been paid to them, urban soils can be managed in order to provide specific services and they can cover a fundamental role in the sustainability of cities development.

In contrast to natural soils, the properties and pedogenesis of urban soils are dominated by their anthropogenic origin (Lehmann and Stahr, 2007). Soils are formed through the combined effect of physical, chemical, biological and anthropogenic processes on soil parent material and these factors affect soil formation in different ways across the landscape, resulting in the spatial variation that we observed (Taalab et al., 2012). The urban environment has unique features that have direct and indirect impacts on soil properties and processes: cities mediate climate, topography, organisms, and
time in the formation of urban soils, as well as contribute to novel anthropogenic parent materials (Pavao-Zuckerman, 2008). As a consequence, compaction of soil, soil microclimates, the availability of water, the activity of soil organisms, the chemical properties (i.e. heavy metal concentration or rates of nitrogen and sulfur) may result altered in urban soils (Groffman et al. 1995; Pouyat et al. 2003; Pavao-Zuckerman & Coleman 2007). Urban soils may be more or less composed of coarse natural and anthropogenic materials (e.g., bricks, concrete, asphalt), and coarse constituents may contain high concentrations of pollutants in contrast to non-urban soils (Morel et al., 2015). The net effect of these urban effects on the physical, chemical, and biological properties of soils is an alteration of the fundamental nature of the belowground component of urban ecosystems, which ultimately shifts ecosystem functions and processes related to biogeochemical cycling (Goldman et al., 1995; Kaye et al., 2005; Pavao-Zuckerman and Coleman, 2005). Regarding the impact on carbon, urban soils may have input of organic material (i.e. hydrocarbons) or can happen that activities like aggressive land clearing and replanting can affect SOC stocks by changing soil properties and the dominant plant functional types (Pongratz et al., 2009; Bae et al., 2014).

1.3 Estimate Carbon stock for urban soils

Until recently, urban soils and their biogeochemical cycles as well as their ecosystem services have not been studied extensively (Pouyat et al., 2006; Lehmann and Stahr, 2007). The total quantity of organic carbon stored in urban soils (under both impervious and non-impervious surfaces), together with that in above-ground vegetation, has never been systematically measured or has been often considered negligible (Tao et al., 2014). This highlights a major gap in the understanding of this crucial ecosystem service, for which Kyoto protocol signatories are required to provide accurate inventories in national estimates of C storage (Edmondson et al., 2012).

In Italy, the C stock of the Lombardy region was estimated within a regional monitoring project on climate changes and greenhouse gases (called ‘KYOTO Project’) by the Regional Agency for Agriculture and Forests of Lombardy (ERSAF). As happened for other inventories (Edmondson et
urban areas have been excluded from the C stock estimation, based on the misleading assumptions that they do not store carbon or because of the lack of data available on the typical C storage of the urban soils. The common focus on C stock of agricultural and natural ecosystems (Don et al., 2011; Guo & Gifford, 2002) has led to the lack of C stock data for urban areas, with a consequent presence of empty spaces in regional soil monitoring systems (Rawlins et al., 2008) and the difficulty to accurately estimate or predict the regional carbon budget accounting for urban areas. This is particularly relevant if we considered that in densely urbanized areas such as Europe, urban land is estimated to cover 9% of the continent (Scalenghe and Marsan, 2009).

Carbon storage by soils can be directly measured or predicted with deterministic models (Morel et al., 2015). A common approach to estimate terrestrial Carbon stocks is to use models based on the assumption that the only changes in carbon stocks are due to changes in land covers (i.e. the InVEST Carbon Storage and Sequestration model developed by the Natural Capital Project team (Daily et al., 2009; Tallis et al., 2013). These models allowed to produce maps of carbon stocks and to compare different areas and historical land cover maps (Tao et al., 2014). Input data required are the estimates of the amount of carbon stored in a given carbon pool (i.e. soil organic matter) for each land cover type (Tao et al, 2014, Chan et al., 2006). This implies that the results of the model depends greatly on the accuracy of land use and land cover map and on the reliability of estimation of carbon stocks for each land use and land cover type (Munoz-Rojas et al., 2011). However, due to the unique features of urban soils, accurate estimation of C stock for different land cover types in urban areas is not available or it is difficult to assess. As accurate assessments of ecosystem C stocks are crucial to understand anthropogenic changes to the global carbon cycle, and to guide effective management of urban soils (Guo and Gifford 2002). Studies aiming to enhance the accuracy of C stock estimates for urban areas are needed and the study presented here is an attempt. In order to disentangle some of the uncertainties related to this anthropogenic-altered system and to enhance the accuracy of C stock estimates, in this study we: i) describe the characteristics of urban soils in the study area; ii) estimate the organic C in the topsoil (0-40 cm) iii) test the influence of different environmental variables on C
stocks and iv) explore the possibility to define the typical carbon stocks of different urban land cover and typology of areas for the urban area of Milan. A greater understanding of urban soils properties is urgently needed in order to assess their role in the global C cycle and to model their ecosystem services for the urban population (Tratalos et al., 2007).

2. Study area

The study area is the city of Milan (9.177224 E, 45.472098 N) and surrounded municipalities, that is one of the most extended and densely populated urban area of Italy. The population of Milan is of 1,345,851 inhabitants and the surface of 181.67 km². Green areas within the city (that comprised artificial, non-agricultural vegetated areas) cover an area of 25 m² representing almost 14% of city surface. Milan has a typical continental climate, with yearly average rainfall of 920 mm and average annual minimum and maximum temperatures of 8 °C and 17 °C respectively (Centro Meteorologico Lombardo). Metropolitan areas experience a 2-3 degree higher rise in temperature than in normality due to the urban heat island syndrome. The city of Milan is situated in the central-western area of the Po Valley. The topography is quite simple, with an average altitude of 100 m. The altitude sharply increases as it extends north beyond Brianza hills until reaching the Alps, over 3,000 m high and only 200 km from Milan.

3. Materials and Methods

3.1 Soil sampling

Soils sampling was performed during years 2014-2015 and soil samples were collected using a soil probe in 84 georeferenced sites (Figure 1) within 15 urban parks (selected in the framework of the PhD project) and in 26 additional urban green areas randomly selected from the land cover type “artificial, non-agricultural vegetated areas” (that comprised vacant sites, vegetated urban squares, private gardens, tree alley, …) (Table 1). In each site a topsoil layer of 40 cm depth was collected and
divided into three subsamples of different depth (0-10 cm, 10-20 cm and 20-40 cm labelled I, II and III layer respectively) collecting a total of 233 subsamples. Five replicate soil samples were taken at each site and composed in order to form the final sample (Figure 2). Many studies differentiate between topsoil and subsoil by depth (De Vos et al., 2005; Katterer et al., 2006). The data used in this study were sampled by pre-defined layers, meaning that there was a uniform sampling depth between sites and the number of samples taken at a given location was always of three subsamples (except for locations with a soil depth lower than 40 cm).

Soil bulk density (BD) was determined for each site for the first layer by the cylindrical core method on undisturbed core samples of 100 cm$^3$ volume, considering the volume of stones, when present. In order to estimate bulk density we used a pedotransfer function for compacted subsoils (Hollis et al., 2012) for the layer II (10-20 cm depth) and the layer III (20-40 cm depth) (equation 1) and a pedotransfer function for horizons “A_ot” (surface mineral layer showing distinct incorporation of organic matter of ‘other’ semi-natural vegetation, Hallett et al., 1998) for the first layer (0-10 cm depth) (equation 2), only when observed data were not available.

Equation (1):

$$\text{Bd} (\text{g/cm}^3) = 1.1257 - (0.1140245 \times \log_e (\text{OrgC})) + (0.0555 \times \log_e (\text{Horizon mid-point, cm})) + (0.002248 \times \text{Sand})$$

where OrgC = organic carbon in %, horizon mid-point = mean depth of the layer, Sand = total sand in %.

Equation (2):

$$\text{Bd} [A_{ot}] (\text{g/cm}^3) = 0.870 + (0.0710 \times \log_e (\text{Clay})) + (0.0930 \times \log_e (\text{Sand})) - (0.254 \times \log_e (\text{OrgC}))$$

where Clay = Clay in %, Sand = total sand in %, OrgC = organic carbon in %.

Table 1 (following page). Sampled sites with a short description of the area.
<table>
<thead>
<tr>
<th>n.</th>
<th>Cod. site</th>
<th>Area</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>BOSCOINCITTAb1</td>
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</tr>
<tr>
<td>2</td>
<td>BOSCOINCITTAb2</td>
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<td>Peri-urban park of big size (herbaceous vegetation cover)</td>
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<td>5</td>
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<td>7</td>
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<td>10</td>
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<td>Peri-urban park of big size (herbaceous vegetation cover)</td>
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</tr>
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<td>SEMP1</td>
<td>Urban park of medium-size small (grassland with few trees)</td>
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<td>Peri-urban park of big size (herbaceous vegetation cover)</td>
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<td>Peri-urban park of big size (tree cover)</td>
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</tr>
<tr>
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</tr>
<tr>
<td>46</td>
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<td>Urban park of big size (grassland with few trees)</td>
</tr>
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<td>TRENNO4</td>
<td>Peri-urban park of big size (herbaceous vegetation cover)</td>
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<td>PINI_PIOPPETO</td>
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<td>Vacant site (grassland with few trees)</td>
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<td>71</td>
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<td>Vacant site (tree cover)</td>
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<td>PZA_ARMI7</td>
<td>Vacant site (herbaceous vegetation cover)</td>
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<td>SANVITTORE1</td>
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<td>VIAMILANESE1</td>
<td>Other green urban areas</td>
</tr>
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<td>VILLALITTAb</td>
<td>Urban park of medium-size small (tree cover)</td>
</tr>
<tr>
<td>83</td>
<td>VILLALITTAp</td>
<td>Urban park of medium-size small (herbaceous vegetation cover)</td>
</tr>
<tr>
<td>84</td>
<td>WERTEINSTEIN1</td>
<td>Private garden</td>
</tr>
</tbody>
</table>

**Chapter IV**
Figure 1. Study area and sampling sites.

Figure 2. Soil sampling scheme.
3.2 Soil chemical analysis

Soil samples were air dried and sieved (2 mm mesh). We measured organic carbon (SOC), total nitrogen and C/N content (Flash EA 1112 NCSoil, Thermo Fisher Scientific CN elemental analyser, Pittsburgh, USA) and soil pH was determined potentiometrically in a soil-to-solution (water and KCl 1N) ratio of 1:2.5 (Jackson, 1958). The content of inorganic carbon was determined analysing carbonates (for samples with pH > 7.0) with gas volumetric method. Textural fractions were measured with particle size determination (Pipette Method). Diametric limits were cSa (coarse sand) 2-0.1 mm; fSa (fine sand) 0.1-0.05 mm; Si (silt) 0.05-0.002 mm; C (clay) <0.002 mm). Determination of available phosphorus (P) was carried out on the samples of the I layer (0-10 cm) using the Olsen method (Olsen et al., 1954). Heavy metals (Cr, Cu, Ni, Pb and Zn) were extracted with a solution of HCl 37% and HNO3 65% and analysed using soil partial digestion to obtain pseudo-total concentration.

3.3 Environmental variables

Environmental variables recorded were: land cover (classified in four classes: 1 tree cover; 2 herbaceous vegetation cover with few trees; 3 herbaceous vegetation cover without trees; 4 other urban green areas - when none of the previous classes were applicable), typology of area (1 big peri-urban park; 2 big urban park; 3 medium-little urban park; 4 other urban green area - when no other classes were applicable; 5 private garden; 6 vacant site), age of the area, vegetation (tree cover in percentage around a 30x30 m buffer), soil depth. The age was assigned related to the year of establishment for samples taken within parks or areas where a ‘foundation’ date was available. For those areas without a foundation year, historical aerial photo (freely available from the Geographical Military Institute) were consulted in order to determine the age.
3.4 Statistical analysis

For each variable, a preliminary exploratory data analysis including calculation of descriptive statistics was performed. We used one-way analysis of variance (ANOVA) followed by Tukey’s post hoc test to compare SOC concentration, C/N, OC stock, available Phosphorous (avP) and pH for each typology of area and land cover type. A principal component analysis (PCA) was performed in order to investigate land cover and typology of use patterns and to reduce correlated variables to a smaller number of uncorrelated factors. Soil and environmental variables used in the PCA were: mean OC stock, mean SOC concentration, mean N, mean C/N, mean pH, mean avP, age of the site, soil depth, coarse sand, fine sand, silt and clay.

In order to test the dependence of organic Carbon stock for the different environmental variables in the surveyed sites we performed mixed effect model. Firstly, the regression assumptions (Osborne and Waters 2002) were tested. LN transformation was performed in order to improve also the linearity of the relationship with the regressors. The variance inflation factor (VIF) test was performed to detect collinearity among regressors (Hsieh et al. 2003). This factor measures the inflation in the variances of the parameter estimates due to collinearities that exist among the regressor (independent) variables. VIF greater than 10 was the criteria used to decide to perform stepwise regression analysis for the selection of the best subset of regressors. The effect of the typology of areas on OC stock were investigated considering typology of areas as fixed effects in the mixed effect model. For the model, typology of areas did not result significant at probability level less than 0.05, consequently this effects were excluded by the successive analyses. Soil and environmental variables tested were: mean C/N, mean pH, mean avP, coarse sand, silt, clay, age of the site, soil depth, and vegetation. The spatial covariance function of residuals was determined iteratively by estimating the partial sill, range and nugget effect parameters, starting from the values of a variogram model based on the residuals of an OLS regression model. The fitting process relied on an iterative procedure aimed at maximizing the log likelihood of the data by restricted maximum likelihood method (REML) (Littell et al. 2006). The fixed effects estimates were obtained as generalized least squares estimates evaluated at the REML
estimate of the covariance parameters. Statistical analyses were performed with STATISTICA and SAS (release 9.4, SAS Institute); the linear models were estimated with the mixed effect model procedure (PROC MIXED); for the spatial model the statement REPEATED was used to specify the R matrix.

4. Results

4.1 Urban soils properties

The characterization of urban soils is presented in Table 2. The average soil pH between the whole study area is moderately acid in the three layers: between 6.44 ± 0.83 and 6.56 ± 0.89. The content of SOC was of 29.23 ± 8.68 g kg\(^{-1}\) for the first layers and lower for II and III layers (16.43 ± 5.21 and 11.70 ± 4.39 respectively). The C stock followed the typical decreasing trend with increase of soil depth with a mean of 3.11 ± 1.16 for the first layer and 1.92 ± 0.59 and 2.56 ± 0.87 respectively for the second and third layer (the latter is thick the double in respect to the others). The level of available P in the first layer is moderately low (5.5 mg kg\(^{-1}\)). Depth of sampling was set at 40 cm, but soil thickness was found lower in some sites, with a minimum depth of 10 cm (mean depth sampled among the study area 35.6 ± 8.22 cm). Age of the sites ranged from 2 years to 330 years.

Results of ANOVA showed that differences between SOC concentration, C/N, C stock, pH and Phosphorus assimilable (avP) occur between some typology of area, but not for land cover. Hereafter we reported the significant results for the typology of area (Tables 3 to 7). Typology of area 1, 2 and 3 corresponded to parks, while the typologies 4, 5 and 6 corresponded to other green urban sites. SOC content and stock generally showed a significant difference between parks and other green urban sites (in particular big peri-urban parks and medium-little urban parks with other urban green area and vacant sites), with parks storing higher quantity of C. Available P did not differed between typologies (except for big peri-urban parks and medium-little urban parks), while pH varied significantly.
between big peri-urban and urban parks and medium-little urban parks, other urban green areas, private gardens and vacant sites).

Table 2. Descriptive statistics of soil data. C stock: organic carbon stored (kg m⁻²); pH: pH in water; SOC: soil organic carbon (g kg⁻¹); N: total nitrogen (g kg⁻¹); C/N: carbon nitrogen ratio; Pass: available phosphorus content (mg kg⁻¹); TXT: cSa = coarse sand content (g kg⁻¹), fSa = fine sand content (g kg⁻¹), Si = silt content (g kg⁻¹), C = clay content (g kg⁻¹); Bd: bulk density (g cm⁻³).

<table>
<thead>
<tr>
<th></th>
<th>n. samples</th>
<th>mean</th>
<th>median</th>
<th>dev.std</th>
<th>min</th>
<th>max</th>
</tr>
</thead>
<tbody>
<tr>
<td>C stock 0-10 cm</td>
<td>83</td>
<td>3.11</td>
<td>2.91</td>
<td>1.16</td>
<td>0.75</td>
<td>6.48</td>
</tr>
<tr>
<td>C stock 10-20 cm</td>
<td>78</td>
<td>1.92</td>
<td>1.84</td>
<td>0.59</td>
<td>0.70</td>
<td>3.36</td>
</tr>
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<td>2.43</td>
<td>0.87</td>
<td>0.81</td>
<td>5.07</td>
</tr>
<tr>
<td>C stock 0-40 cm</td>
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<td>7.00</td>
<td>2.80</td>
<td>1.32</td>
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</tr>
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<td>pH 0-10 cm</td>
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<td>6.44</td>
<td>6.40</td>
<td>0.83</td>
<td>4.70</td>
<td>8.10</td>
</tr>
<tr>
<td>pH 10-20 cm</td>
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<td>6.50</td>
<td>6.50</td>
<td>0.90</td>
<td>4.80</td>
<td>8.20</td>
</tr>
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<td>pH 20-40 cm</td>
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<td>6.56</td>
<td>6.50</td>
<td>0.89</td>
<td>4.90</td>
<td>8.10</td>
</tr>
<tr>
<td>SOC 0-10 cm</td>
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<td>29.72</td>
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<td>8.12</td>
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<tr>
<td>SOC 10-20 cm</td>
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<td>15.26</td>
<td>5.21</td>
<td>6.34</td>
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<tr>
<td>SOC 20-40 cm</td>
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<td>11.70</td>
<td>10.66</td>
<td>4.39</td>
<td>5.48</td>
<td>25.54</td>
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<tr>
<td>N 0-10 cm</td>
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<td>2.75</td>
<td>2.74</td>
<td>0.82</td>
<td>0.75</td>
<td>5.09</td>
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<td>N 10-20 cm</td>
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<td>1.53</td>
<td>0.52</td>
<td>0.53</td>
<td>3.25</td>
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<td>N 20-40 cm</td>
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<td>1.23</td>
<td>1.12</td>
<td>0.41</td>
<td>0.72</td>
<td>2.27</td>
</tr>
<tr>
<td>C/N 0-10 cm</td>
<td>83</td>
<td>10.72</td>
<td>10.51</td>
<td>1.27</td>
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</tr>
<tr>
<td>C/N 10-20 cm</td>
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<td>9.71</td>
<td>1.64</td>
<td></td>
<td></td>
</tr>
<tr>
<td>C/N 20-40 cm</td>
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<td>9.56</td>
<td>9.37</td>
<td>1.91</td>
<td></td>
<td></td>
</tr>
<tr>
<td>avP 0-10 cm</td>
<td>83</td>
<td>20.01</td>
<td>17.03</td>
<td>11.50</td>
<td>5.10</td>
<td>62.60</td>
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<td>338.00</td>
<td>85.61</td>
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<td></td>
</tr>
<tr>
<td>TXT-fSa</td>
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<td>132.83</td>
<td>130.00</td>
<td>31.75</td>
<td></td>
<td></td>
</tr>
<tr>
<td>TXT-Si</td>
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<td>409.63</td>
<td>402.00</td>
<td>82.52</td>
<td></td>
<td></td>
</tr>
<tr>
<td>TXT-C</td>
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<td>119.43</td>
<td>118.00</td>
<td>31.82</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Db 0-10 cm</td>
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<td>1.16</td>
<td>1.15</td>
<td>0.09</td>
<td>0.98</td>
<td>1.49</td>
</tr>
<tr>
<td>Db 10-20 cm</td>
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<td>1.33</td>
<td>0.04</td>
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</tr>
<tr>
<td>Db 20-40 cm</td>
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<td>1.41</td>
<td>1.41</td>
<td>0.05</td>
<td>1.29</td>
<td>1.49</td>
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</table>
Table 3. Tukey’s post hoc test of the mean SOC concentration (%) for typology of areas. Red indicate significance less than 0.05 of the Tukey’s post hoc test.

<table>
<thead>
<tr>
<th>typology of area</th>
<th>1 big peri-urban park</th>
<th>2 big urban park</th>
<th>3 medium-little urban park</th>
<th>4 other urban green area</th>
<th>5 private garden</th>
<th>6 vacant site</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0.989</td>
<td>0.989</td>
<td>0.617</td>
<td>0.040</td>
<td>0.997</td>
<td>0.128</td>
</tr>
</tbody>
</table>

Table 4. Tukey’s post hoc test of the mean C/N for typology of areas. Red indicate significance less than 0.05 of the Tukey’s post hoc test.

<table>
<thead>
<tr>
<th>typology of area</th>
<th>1 big peri-urban park</th>
<th>2 big urban park</th>
<th>3 medium-little urban park</th>
<th>4 other urban green area</th>
<th>5 private garden</th>
<th>6 vacant site</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0.024</td>
<td>0.024</td>
<td>0.599</td>
<td>0.664</td>
<td>0.020</td>
<td>0.425</td>
</tr>
</tbody>
</table>

Table 5. Tukey’s post hoc test of the C stock for typology of areas. Red indicate significance less than 0.05 of the Tukey’s post hoc test.

<table>
<thead>
<tr>
<th>typology of area</th>
<th>1 big peri-urban park</th>
<th>2 big urban park</th>
<th>3 medium-little urban park</th>
<th>4 other urban green area</th>
<th>5 private garden</th>
<th>6 vacant site</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1.000</td>
<td>1.000</td>
<td>0.968</td>
<td>0.002</td>
<td>0.994</td>
<td>0.013</td>
</tr>
</tbody>
</table>

Table 6. Tukey’s post hoc test of the pH for typology of areas. Red indicate significance less than 0.05 of the Tukey’s post hoc test.

<table>
<thead>
<tr>
<th>typology of area</th>
<th>1 big peri-urban park</th>
<th>2 big urban park</th>
<th>3 medium-little urban park</th>
<th>4 other urban green area</th>
<th>5 private garden</th>
<th>6 vacant site</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0.964</td>
<td>0.964</td>
<td>0.011</td>
<td>0.000</td>
<td>0.029</td>
<td>0.001</td>
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</tbody>
</table>
Table 7. Tukey’s post hoc test of the Pass for typology of areas. Red indicate significance less than 0.05 of the Tukey’s post hoc test.

<table>
<thead>
<tr>
<th>typology of area</th>
<th>mean (mg/kg)</th>
<th>dev.std</th>
<th>legal limit (mg/kg)</th>
<th>n. samples exceeding</th>
<th>% samples</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 big peri-urban park</td>
<td>0.274</td>
<td>0.006</td>
<td>0.131</td>
<td>0.667</td>
<td>0.950</td>
</tr>
<tr>
<td>2 big urban park</td>
<td>0.274</td>
<td>0.989</td>
<td>1.000</td>
<td>1.000</td>
<td>0.280</td>
</tr>
<tr>
<td>3 medium-little urban park</td>
<td>0.006</td>
<td>0.989</td>
<td>0.857</td>
<td>1.000</td>
<td>0.065</td>
</tr>
<tr>
<td>4 other urban green area</td>
<td>0.131</td>
<td>1.000</td>
<td>0.857</td>
<td>0.996</td>
<td>0.270</td>
</tr>
<tr>
<td>5 private garden</td>
<td>0.667</td>
<td>1.000</td>
<td>1.000</td>
<td>0.065</td>
<td>0.475</td>
</tr>
<tr>
<td>6 vacant site</td>
<td>0.950</td>
<td>0.280</td>
<td>0.065</td>
<td>0.270</td>
<td>0.475</td>
</tr>
</tbody>
</table>

Heavy metals concentration in the study area exceeded the legal limits for Pb (in almost 50% of the samples) and also for Zn and Cu (Table 8, Figure 3). Concentration of Cr and Ni has never been found to exceed legal limits.

Table 8. Heavy metals concentration and exceed of the legal limits.

<table>
<thead>
<tr>
<th>Metal</th>
<th>mean (mg/kg)</th>
<th>dev.std</th>
<th>legal limit (mg/kg)</th>
<th>n. samples exceeding</th>
<th>% samples</th>
</tr>
</thead>
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<tr>
<td>Cr</td>
<td>60.21</td>
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<td>150</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Cu</td>
<td>54.78</td>
<td>39.61</td>
<td>120</td>
<td>5</td>
<td>6%</td>
</tr>
<tr>
<td>Ni</td>
<td>41.98</td>
<td>9.63</td>
<td>120</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Pb</td>
<td>123.74</td>
<td>92.73</td>
<td>100</td>
<td>40</td>
<td>48%</td>
</tr>
<tr>
<td>Zn</td>
<td>103.56</td>
<td>58.35</td>
<td>150</td>
<td>12</td>
<td>14%</td>
</tr>
</tbody>
</table>

Figure 3. Heavy metals concentration. Error bars indicate 95% confidence interval. Red lines indicate legal limits.
4.2 Carbon stock in urban soils

OC stock in urban soils is highly variable among sampled sites and areas. SOC concentration varied significantly between big peri-urban parks and other green areas (classes 1 and 4) and also between medium-little urban parks and other green areas and vacant sites (classes 3, 4 and 6) (Table 5, Figure 4).

Figure 4. Vertical distributions of the SOC concentration (%) in different typology of areas. I = 0-10 cm depth, II = 10-20 cm depth and III = 20-40 cm depth. Error bars indicate 95% CI.
The first two components of the PCA explained around 55% of variance of the data observed (Figure 5). First component is mainly related to C and pH, while second component to age and textural parameters. The typology of area (Figures 6) showed a low significative clusterization of big peri-urban parks, big urban parks and private gardens (typology 1, 2 and 5). Soils belonging to parks (typology of area 1, 2 and 3) showed a moderate differentiation compared to other area and were characterized by higher SOC and C stock, while soils belonging to non-parks (typology 4, 5 and 6) were characterized by higher pH and content of coarse sand. No evident clusters were found for different land cover types (Figures 7).

Figure 5. Loading plot of the PCA. Cm = mean C stock; Cconcm = mean SOC concentration; Nconcm = mean N concentration; CNm = mean C/N; pH_medio = mean pH; Pass = mean available P; age = age of the site; spess = soil depth; Sg = coarse sand; Sf = fine sand; L = silt; A = clay.
Figure 6. Score plot of the PCA. Different colors defined different classes of typology of area.

Figure 7. Score plot of the PCA. Different colors defined different classes of land cover.
The dependence of organic Carbon stock for the different environmental variables in the surveyed sites was tested using mixed effect model. The results of the variance inflation factor (VIF) test helped us to select the best subset of regressors (Table 9).

Table 9. Collinearity between variables was not detected. veg = vegetation; age = age of the area; depth = soil depth; pHm = mean pH; CNm = mean C/N; Pass = available phosphorus; Sg = coarse sand content; L = silt content; A = clay content.

<table>
<thead>
<tr>
<th>Variable</th>
<th>DF</th>
<th>Estimate</th>
<th>Error standard</th>
<th>Value t</th>
<th>Pr &gt;</th>
<th>Tollerance</th>
<th>VIF</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intercept</td>
<td>1</td>
<td>-1.02537</td>
<td>0.51809</td>
<td>-1.98</td>
<td>0.0516</td>
<td>.</td>
<td>0</td>
</tr>
<tr>
<td>veg</td>
<td>1</td>
<td>-0.00152</td>
<td>0.00108</td>
<td>-1.40</td>
<td>0.1668</td>
<td>0.82105</td>
<td>1.21795</td>
</tr>
<tr>
<td>age</td>
<td>1</td>
<td>0.00090288</td>
<td>0.00045416</td>
<td>1.99</td>
<td>0.0506</td>
<td>0.77431</td>
<td>1.29147</td>
</tr>
<tr>
<td>depth</td>
<td>1</td>
<td>0.02905</td>
<td>0.00617</td>
<td>4.71</td>
<td>&lt;.0001</td>
<td>0.26049</td>
<td>3.83894</td>
</tr>
<tr>
<td>pHm</td>
<td>1</td>
<td>0.03422</td>
<td>0.03041</td>
<td>1.13</td>
<td>0.2641</td>
<td>0.32463</td>
<td>3.08044</td>
</tr>
<tr>
<td>CNm</td>
<td>1</td>
<td>0.06671</td>
<td>0.02706</td>
<td>2.47</td>
<td>0.0160</td>
<td>0.64458</td>
<td>1.55141</td>
</tr>
<tr>
<td>Pass</td>
<td>1</td>
<td>0.00586</td>
<td>0.00267</td>
<td>2.19</td>
<td>0.0318</td>
<td>0.78875</td>
<td>1.26783</td>
</tr>
<tr>
<td>Sg</td>
<td>1</td>
<td>0.00006737</td>
<td>0.00048215</td>
<td>0.14</td>
<td>0.8893</td>
<td>0.40216</td>
<td>2.48658</td>
</tr>
<tr>
<td>L</td>
<td>1</td>
<td>0.00236</td>
<td>0.00049508</td>
<td>4.77</td>
<td>&lt;.0001</td>
<td>0.40597</td>
<td>2.46326</td>
</tr>
<tr>
<td>A</td>
<td>1</td>
<td>-0.00147</td>
<td>0.00088167</td>
<td>-1.67</td>
<td>0.0997</td>
<td>0.83420</td>
<td>1.19876</td>
</tr>
</tbody>
</table>

Spatial correlation of residuals was detected, but the covariance function had the structured component lower than the non-structured one. Results of mixed models showed that the spatial model (Tables 10-11) was not more efficient than the non spatial one (Tables 12-13).

Table 10. Error covariance parameters of the non-spatial model.

<table>
<thead>
<tr>
<th>Cov. Param.</th>
<th>Estimate</th>
<th>Std. err.</th>
<th>Value z</th>
<th>Pr &gt; Z</th>
<th>Alfa</th>
<th>Inferiore</th>
<th>Superiore</th>
</tr>
</thead>
<tbody>
<tr>
<td>Residual</td>
<td>0.06055</td>
<td>0.01002</td>
<td>6.04</td>
<td>&lt;.0001</td>
<td>0.05</td>
<td>0.04486</td>
<td>0.08622</td>
</tr>
</tbody>
</table>
Table 11. Parameter estimates and solutions for fixed effect of the non-spatial model. Veg=vegetation, age = age of the site, depth = soil depth, pHm = mean pH, CNm = mean C/N, avP = mean available P, cSa = coarse sand content, Si = silt content, C = clay content.

| Effect  | Estimate | Std. err. | DF  | T value | Pr > |t| |
|---------|----------|-----------|-----|---------|-------|---|
| Intercept | -1.0254 | 0.5181 | 73 | -1.98 | 0.0516 |
| veg | -0.00152 | 0.001085 | 73 | -1.40 | 0.1668 |
| age | 0.000903 | 0.000454 | 73 | 1.99 | 0.0506 |
| depth | 0.02905 | 0.006169 | 73 | 4.71 | <.0001 |
| pHm | 0.03422 | 0.03041 | 73 | 1.13 | 0.2641 |
| CNm | 0.06671 | 0.02706 | 73 | 2.47 | 0.0160 |
| avP | 0.005856 | 0.002675 | 73 | 2.19 | 0.0318 |
| cSa | 0.000067 | 0.000482 | 73 | 0.14 | 0.8893 |
| Si | 0.002360 | 0.000495 | 73 | 4.77 | <.0001 |
| C | -0.00147 | 0.000882 | 73 | -1.67 | 0.0997 |

Table 12. Error covariance parameters of the spatial model.

<table>
<thead>
<tr>
<th>Param cov</th>
<th>Soggetto</th>
<th>Stima</th>
<th>Errore standard</th>
<th>Valor</th>
<th>Pr &gt; Z</th>
<th>Alfa</th>
<th>Inferior</th>
<th>Superior</th>
</tr>
</thead>
<tbody>
<tr>
<td>Variance</td>
<td>Intercept</td>
<td>0.06431</td>
<td>8.0659</td>
<td>0.01</td>
<td>0.4968</td>
<td>0.05</td>
<td>.</td>
<td>.</td>
</tr>
<tr>
<td>SP(SPH)</td>
<td>Intercept</td>
<td>1000.00</td>
<td>0</td>
<td>.</td>
<td>.</td>
<td>.</td>
<td>.</td>
<td></td>
</tr>
<tr>
<td>Residual</td>
<td>0.06053</td>
<td>0.01028</td>
<td>5.89</td>
<td>&lt;.0001</td>
<td>0.05</td>
<td>0.04453</td>
<td>0.08707</td>
<td></td>
</tr>
</tbody>
</table>

Table 13 Parameter estimates and solutions for fixed effect of the spatial model. Veg=vegetation, age = age of the site, depth = soil depth, pHm = mean pH, CNm = mean C/N, avP = mean available P, cSa = coarse sand content, Si = silt content, C = clay content.

| Effetto  | Stima | Errore standard | DF | Valore t | Pr > |t| |
|----------|-------|-----------------|----|----------|-------|---|
| Intercept | -1.0249 | 0.5786 | 1 | -1.77 | 0.3272 |
| veg | -0.00151 | 0.001091 | 71.8 | -1.39 | 0.1696 |
| age | 0.000902 | 0.000458 | 72.9 | 1.97 | 0.0527 |
| depth | 0.02905 | 0.006202 | 72.8 | 4.68 | <.0001 |
| pHm | 0.03424 | 0.03049 | 72.5 | 1.12 | 0.2651 |
| CNm | 0.06667 | 0.02741 | 73 | 2.43 | 0.0175 |
| avP | 0.005860 | 0.002693 | 72.5 | 2.18 | 0.0328 |
| cSa | 0.000068 | 0.000485 | 72.5 | 0.14 | 0.8895 |
| Si | 0.002361 | 0.000497 | 71.5 | 4.75 | <.0001 |
| C | -0.00147 | 0.000900 | 71.9 | -1.64 | 0.1063 |
5. Discussion

5.1 Urban soils properties

Soils whose properties and functions are profoundly modified and dominated through long-term technical human activity are classified as Antrhosols (IUSS Working Group WRB, 2008). These soils may contain significant amounts of artifacts, or be sealed by technic hard rock and include soils from wastes, pavements with their underlying unconsolidated materials, soils with geomembranes and soils constructed from human-made materials (IUSS Working Group WRB, 2006). Urban soils analyzed in this study were characterized by frequent incorporation of anthropogenic coarse material, like bricks, mortars, concretes, slags or various garbage and the composition generally showed a pronounced heterogeneity among the areas investigated and also among different sites in the same area (i.e. adjacent sites in the same park). Other not characterized or not detected materials presented in the soil samples may be related to industrial waste, processed oil products, ash or sludge and sewage (Lorenz and Lal., 2009; Rossiter, 2007). Besides importing material while constructing urban soils, biogeochemical cycling of C and N may be affected in various way. For example, it can be directly altered by vegetation management with addition of inorganic and organic fertilizers (Lorenz and Lal., 2009) or by presence of garbage that has very heterogeneous properties related to biogeochemical cycling of C and N (Bridges, 1991). However, any generalization of observed effects of the urban environment on biogeochemical C and N cycles remains highly uncertain (Pickett et al., 2001) because of the little ecological information about urban areas. The presence of high amounts of coarse materials that we found resulted in the reduction of the fine earth volume (Lorenz and Kandeler, 2006). Soil bulk density in many sites was found low (i.e. 0.61 and 0.83 g cm\(^{-3}\) in CAVE1 and CAVE4, 0.62 g cm\(^{-3}\) in VENOSTA, 0.74 g cm\(^{-3}\) in CASTELLANZA). The soils with such low values of bulk density were generally characterized by limited soil depth (less than 40 cm) and heavy inclusions (of coarse anthropogenic materials), probably related to recent mixing perturbation that increased the percentage of coarse material. However, soil bulk density for soils with elevated
presence of skeleton (more than 35%) may lose in significance. Soil bulk density found in the urban soils of this study never reached high values.

Industrial waste disposal may be responsible for elevated soil metals concentrations that pose both ecological and human health risks (Davison et al., 2006). High concentration of heavy metals exceeding legal limits were found in the study area. In particular, Pb was found to exceed limits for almost half of the sites investigated (concentration higher than 100 mg kg\(^{-1}\)), and an additional 10% had concentration higher than 90 mg kg\(^{-1}\). Soils with high concentration of Pb belonged indistinctly to parks, vacant sites, private gardens or other green areas. The reasons for the elevated concentration of Pb detected are not easy to determine because of the uncertainties of past polluting deposits. Pb was normally related to traffic as it was previously emitted by cars that used petrol containing this metal. Legislation stopped definitively its sale but this regulation took place only in recent times (2002). In addition to being directly toxic to terrestrial biota (Jim, 1998), heavy metals can reduce metabolism of soil microbes and microfauna, alter soil food webs, and decrease decomposition rates and shifting biogeochemical cycling of urban soils (Pavao-Zuckerman, 2008).

5.2 Carbon stock in the urban soils of Milan

It is known that soil Carbon stock varies according to land use, type of soil and it also depend on kind of vegetation covering soils, soil management practices and past management. Previously, a lack of data has necessitated the assumption that soils in urban areas are so functionally compromised that they are unable to store Carbon (Edmondson et al., 2012). However, as we have found in this study and as literature highlights (Lorenz and Lal, 2015; Tao et al., 2015; Edmondson et al., 2012; Pouyat et al., 2009; Pavao-Zuckerman, 2008; Rawlins, 2008) urban soils may store significant amount of Carbon (and may not).

In this study, we have focused the C stock estimation on topsoil. Generally, soil surface layers account for the main part of the soil carbon stocks. In Lombardy, a previous study (TOKYO project by ERSAF) found for the regional area that on average the 47% of the carbon stored in soils was held in
the first 30 cm. Carbon stock occurring in the topsoil is thus very important, because this is the portion more influenced by external environmental and human factors and therefore the mainly susceptible to mineralization. On the contrary, in deep layers SOC is more stable and less liable to transformation. The concentrations of organic C in soils that developed on anthropogenic parent material resulted to be highly variable (Lorenz and Lal, 2009; Puyat et al., 2006; Scharenbroch et al., 2005). In urban soils of Stuttgart, Germany, Lorenz and Kandeler (2005) found SOC concentrations varied between 0.6 and 26.2 g C kg⁻¹. In Milan urban soils, SOC concentrations (in the layer 0-40 cm) varied between 3.01 and 24.77 g C kg⁻¹. C stock in urban soils of Milan (mean 6.93 kg m⁻²) was comparable with C stock found for croplands in Lombardy (5.7 kg m⁻² in the first 30 cm of soil) and a bit lower than C stock found for forest, pasture and grasslands (from 7.0 and 9.0 kg m⁻² in the first 30 cm of soil) by ERSAF. Average C stock for the whole Lombardy region was determined as 6.88 kg m⁻² for 30 cm depth, and of 5.76 kg m⁻² for the territory of Pianura Padana (ERSAF), where the city of Milan is located. Thus, the investigated urban soils of Milan showed an average C stock lightly higher that those found for the surrounded territory.

Different processes, sometimes not easy to detect and account for, may lead to an increase or to depletion in organic matter in urban soils. Incorporation of garbage rich in organic matter, presence of components of construction waste which contain organic matter (ash, coal, leather), high above- and belowground net primary production (due to urban vegetation) are factors that lead urban soils to store high C amounts (Lorenz and Lal, 2015; Byrne, 2007). Increase in input of water and nutrients also increases the aboveground net primary productivity, soil respiration and total belowground C allocation than in other land uses, suggesting enhanced C cycling rates by urbanization (Kaye et al., 2005; Pataki et al., 2006; Lorenz and Lal, 2009). On the other side, vegetation management practices responsible for the export of material from the vegetated soil (like removal of grass clippings, tree leaves, and other organic debris), mechanical soil removal (that is usually restricted to topsoil and thus to the layer richer in vegetation and humus) and the loss of highly active soil biota (Lorenz and Lal, 2009; Craul, 1999) are responsible to lower SOC storage. Environmental variables that resulted
more influential in explaining variation in C stock in this study were soil depth, silt content, C/N and available P. To a lower extent, also the age of the area resulted significant. High C/N lead to lower mineralization processes and thus to higher OC stock. Urban soil depth, despite being obviously related to the content of C stock, is a parameter of high importance for define soil fertility and other soil functions (such as roots expansion, water retention capacity, contaminants filter, ...).

5.3 Enhancing the accuracy of C stock estimates and constraints posed by urban areas

In order to assess the impact of urban land use on soil organic carbon stocks it is necessary to integrate our knowledge of urban soils by considering the existing variety of land cover type in urban environment and the typical C stored in each land cover type. Studies revealed that assumptions underpinning current regional or national estimates of ecosystem C stocks, as required by Kyoto Protocol signatories, are not robust and are likely to have seriously underestimated the contributions of urban areas (Edmondson et al., 2012). In order to meet the need to define accurate C stock for land use and land cover types for urban soils that will allow our deterministic model to better perform, this study explored the relationships between C stocks and different land cover and land use (in this study defined as typology of area) and the influence of some environmental variables tested. Results confirmed the high variability of urban soils in the C storage that didn’t allowed us to detect in our sampling any significative relationships between a typical C stock and land cover or land use type. Hereafter, we examined some of the constrains when dealing with urban areas that limit our capacity (or possibility) to model C stock for different land use and land cover and which efforts should be addressed in order to enhance our knowledge about urban soils.

Besides of the characteristics of urban soils already stated in the introduction (human-made origins, different and mixed parent material, altered structure and composition, among others) we detected other factor limiting in this study our interpretation of the soil parameters found. All these constrains were relate to the uncertainty of environmental parameters estimation, because of a completely lack of data about urban soils history. Urban and suburban areas often expand into former agricultural,
industrial, extraction or mining areas and the residual soil effects from former human activities complicate our understanding of urban soils (Pavao-Zuckerman 2008). For example, historical land-use changes have been shown to affect current SOC stocks in constructed urban parks (Takahashi et al., 2008). Other studies highlights as historic agriculture doubled the soil C, N, and ion concentrations in soils compared to urban soils with no legacy of agriculture (Lewis et al. 2006) or that biological waste disposal associated with pre-urban human settlements increase topsoil depth (Davidson et al. 2006). However, the effects of land-use history have rarely been assessed as a determining factor for estimating SOC stocks because of the lack of quantitative data available (Schulp and Verburg, 2009; Tao et al 2014).

The effect of the age of the soil on C stock is known to be relevant, but estimation of age for urban soils may be high uncertain. In this study, we estimated the age using the year of establishment (when available) or we used historical photos. However, in both the cases a certain uncertainty was present: in the former case (when a date was available) it referred to the whole area (i.e. a park) and it doesn’t assure that the site of sampling was younger (i.e. for successive mechanical intervention or renewal of the area) or maybe older (in the case of agricultural lands or meadows in large parks); when age was determined by study of aerial photos, the main limitations were the resolution of the data available and the short historical series. Vegetation is another factor influencing C stock. In this study, we have not detected an influent action of vegetation in determining the SOC found. However, different vegetation management practices may altered the vegetation-soil interactions and input of OM into soil. Vegetation management practices are not easy to record, as well as the use of addictive (i.e. fertilizer) and the same land cover type (i.e. forest or herbaceous cover) may be managed differently. Soil depth clearly affect C stocks. In this study, we found soils with a depth of less than 20 cm and in some cases the soil layer was no more than 10 cm. In these cases, the thin layer of soil was found above a thick layer of anthropogenic material (artefacts, bricks, concretes). Low soil depth was generally found in other green urban areas and in vacant sites, but also in some urban parks, without a clear pattern. Another limitation is related to the definition of urban land use and land cover.
classes. In this study, we classified the land cover of the samples sites referring to the cover within a 30x30m area. Land cover was intended to describe the type of vegetation covering the site. Land use was described here as ‘typology of area’, referring to the whole area where the sampled sites were located. These classifications were made based on the knowledge of the study area, because the regional land cover and land use map available (DUSAF) was not specific for urban areas and because a standardized classification of urban soils covers does not exist even some authors has classified them with accuracy (Rall et al., 2015). In this context, land cover of the same class may correspond to areas of different typology (i.e. herbaceous vegetation cover can be found in a large urban park as well as in a vacant site or in a private garden). The difficulty to classify urban land use and land cover types (in relation to soil properties) may further increase our difficulty to model soil C stock.

The complex heterogeneity of urban soils together with the uncertainty of their origins, their history and management may summarize the reasons of the limitation in our capacity to model their characteristics. We might even say that what is missing in the case of urban soils (and probably will in future), is the soil-landscape relationship, that is instead present in agricultural and forest, natural or semi-natural soils, and that is an exceptional support in the preparation of soil maps. In other words, the soil-landscape paradigm (Hudson, 1992), on which is explicitly or implicitly based the soil mapping, does not serve in the case of urban landscapes, for which it is not possible to rationalize the factors and the soil processes, given the absolute variability and unpredictability of the same in an urban environment. These uncertainties may in part be reduced by the implementation of some initiatives, proposed in Italy by the Regional Institute for Agriculture and Forests Services (ERSAF) like a permanent soil monitoring system (to register variations occurred to the relevant soil properties and qualities), a updated and detailed regional soil information system, protocols to certify the changes of organic carbon stock in mineral soils and experimental fields for long-term evaluation of different crop and forest systems (and urban soils) to sequester organic carbon. However, these proposals would address only in part the problem, relating to the changes in time of the benchmark soils. What can enhance mostly our capacity to map urban soils, probably pass through a strong
thickening of soil sampling in urban areas. Furthermore, increased knowledge of spatial relationships of urban soils by comprehensive soil survey programs will allow better predictive modelling of urban ecosystem functions (Lorenz and Lal, 2009).

6. Conclusions

As urban areas dramatically increase globally, more studies on the effects of urbanization on C storage by urban soils are urgently needed. Building or improving soil natural capital is an important aim, contributing to soil resilience, and maintaining balance in the provision of ecosystem services and the European Union has already identified soil ecosystem services as a priority research area in the European Union Soil Thematic Strategy (Robinson et al., 2012).

This study is the first work aimed to investigate the urban soils of Milan and represent an attempt to enhance our knowledge about the potential of C storage of urban systems and, thus, the contribution of urban ecosystem to the provision of ecosystem services. The research presented highlights the constrains that limit our understanding about urban soils and the need for a deeper investigation of C stock capacity in the soils of the region of Milan, that is one of the most urbanized area of Italy, aiming to improve the reliability of regional C stock estimation. Even though quantification of urban soil organic carbon (SOC) and other urban soil properties remains difficult, a better understanding of such fundamental ecosystem functions is crucial to advance towards sustainability. The contribution of urban ecosystems to national C inventories may have implications for land-use change and planning policy, particularly in densely urbanized regions globally (Tao et al., 2015). Regarding cities, Lorenz and Lal (2009) suggested that to cap urban sprawl C and N sequestration should accompany sustainable and dense development within cities. Policy measures to mitigate the negative effects of soil sealing on accumulation of soil organic carbon and other soil ecosystem services are also seen in the replacement of impervious surfaces (such as parking lots, pavements) with surfaces only partially covering the soil, like bricks with gaps or other vegetated solutions (Tao et al., 2014). However, alongside the avoidance of using traditional coverings (paving) and the promotion of
‘green’ spaces instead of ‘grey spaces’ in urban areas, a better knowledge about the effective contribute of these type of land covers in mitigate negative effect of soil sealing should grow (Tao et al., 2014).

References


Public participatory mapping of cultural ecosystem services: match or mismatch between citizen perception and park management in the Parco Nord of Milano

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In preparation for a Special Issue on Sustainability: “Sustainability in an Urbanizing World: The Role of People”
Abstract

Cultural ecosystem services (CES) provided by urban green areas are one of the most important services from the perspective of urban dwellers. It has been illustrated that despite the concept of ecosystem services being new and unfamiliar to many actors in urban land-use planning, the issues contained in the concept have been included in land-use planning principles based on sustainable development. In fact, even if it not explicit, many decisions are made for the purposes of generating benefits to people. Ecosystem services may be underestimated, and consequently threatened, when land-use planning and management decisions are based on inadequate information. Unfortunately, most of the studies aimed to valuate and map CES have not been used for actual decision support in land use planning therefore there is a gap in the literature about its use in practice. The aim of this study were to (i) identify which were the cultural benefits perceived by city park users and map them through participatory mapping (PPGIS), (ii) identify and map the cultural benefits arising from park management (management perception) and (iii) through a comparison analysis explore if the benefits perceived by citizens and park management match or mismatch. The study area was a large peri-urban park in Milan (Parco Nord). Public workshops were organized in order to collect data on people perception of CES analysed with GIS-based techniques. Maps of individual benefits and aggregated maps of benefits richness were produced. The resulting maps showed places that have bundles of different values, and other places with clusters of the same value type. Comparison analysis revealed hotspot and coldspot of CES. Methodological opportunity and needs of the methodology used were discussed in the light of better integration between CES mapping and decision support in land use planning.

1. Introduction

Cultural ecosystem services (CES) provided by urban green areas are one of the most important services from the perspective of urban dwellers (Niemelä et al., 2010). CES provided benefits that are directly and subjectively recognized by people (Andersson et al., 2015). It has been illustrated that despite the concept of ecosystem services being new and unfamiliar to many actors in urban land-use planning, the issues contained in the concept have been included in land-use planning principles
based on sustainable development (Niemelä et al., 2010). In fact, even if it not explicit, many decisions are made for the purposes of generating benefits to people. For example, recreational facilities (like bench or tables) are placed with the intention to generate benefits in terms of social relations or relax in a place, as well as a tree-lined avenue in a park is intended to provide aesthetic benefits or relax. However, ecosystem services may be underestimated, and consequently threatened, when land-use planning and management decisions are based on inadequate information. Unfortunately, most of the studies aimed to map CES have not been used for actual decision support in land use planning and there is a gap in the literature about its use in practice (Brown and Fagerholm, 2014). Therefore, there is a need to demonstrate a pathway from the inception of mapping of cultural ecosystem services through integration and use of the information for land use decision support (Brown and Fagerholm, 2014).

Moreover, a focus on CES may offer a gateway to ES stewardship through civic engagement and public support (Andersson et al. 2014). Research suggests that there is a strong connection between CES, civic engagement and ES stewardship; threatened or reduced positive experiences of nature interactions seem to be important factors in the emergence of civic participation in stewardship of urban ecosystems (e.g. Hunter, 2011). For example, it has been shown that people are able to forcefully organize to protect urban open spaces such as community gardens when these spaces, and the benefits they provide, are threatened (Ernstson and Sörlin, 2009; Saldivar-Tanaka and Krasny, 2004; Schmelzkopf, 2002). CES may thus facilitate community support for ecologically motivated management actions when these have positive effects on valued CES or, alternatively, help anticipate and address conflicts in cases where ecologically important changes are expected to have negative effects on valued CES (Andersson et al. 2014).

Cultural ecosystem services arise from human perception of the ecosystem, rather than from the ecosystem itself (Andersson et al., 2015; Kaplan and Kaplan, 1989). This lead to the fact that assessment of cultural services is different compared to other services because of some features they present, for example the dependence on individual’s value system and the difficulty in using spatial
geographical units for their assessment (La Rosa et al., 2015). From this derives that CES assessment is less quantitative than others (e.g. provisioning services, which can be quantified independently from human’s perception). Moreover, perception of ecosystem services are context dependent (Buchel and Frantzeskaki, 2015). A study of specific green spaces lead us to determine which specific services are relevant to the local context. Measuring services at broad scales is mostly reliant on modelling approaches, which are often limited by the coarse resolution of the input data (Peh et al., 2013). In order to inform local decision-making, there is a growing need to measure ecosystem services at individual sites at a fine spatial grain, as this is the scale at which many land-use decisions are typically made and need to be informed (Peh et al., 2013). In this context, it is important to identify which benefits are recognized by urban citizens in order to evaluate the perceived value and quality of existing urban green spaces (Buchel and Frantzeskaki, 2015) and to support effective management decisions. Mapping cultural services might be informative in order to detect if citizens’ perception of benefits correspond with those of management for the purposes of providing such benefits. This study aimed to: (i) identify which are the cultural benefits perceived by city park users and map them through public participatory mapping (PPGIS), (ii) identify and map the cultural benefits intended to be provided by park management (management perception) and (iii) through a comparison analysis understand if the benefits perceived by citizens and park management match or mismatch. Results were then discussed in regard to the opportunities and needs of the methodology proposed in this study.

2. Materials and Methods

2.1 Study area

Parco Nord (45°53’71”N, 9°20’97”E) is a peri-urban park that extended for 640 hectares in the northern part of the city of Milan (1.25 million inhabitants) about 9 km from the city centre and its
area is comprised in six municipalities (Milan, Bresso, Cusano Milanino, Cormano, Cinisello Balsamo and Sesto San Giovanni) (Figure 1).

![Figure 1. Study area.](image)

Parco Nord was established as Regional park in 1975. It is situated in one of the most densely urbanized area of Europe that is characterized by historic machinery industries (nowadays mostly disappeared as a result of the de-industrialization) and extended construction districts with few remnants of agricultural areas. The chaotic expansion of the urban tissue in this area has led the North periphery of the city of Milan to be connected with the nearby hinterland without a strategic urban design. The idea of establish a park emerged in the late 1960s. The motivation for building the park was to improve the quality of life for the citizens in the area and followed the reorganization of the city by creating a green lung, along with strong demands from citizens. The construction of the park as well as its development were highly supported by local community and the civic engagement was so influential that the park is known today as “the park desired by citizens”. The development of the park is characterized by a ‘work in progress’ process, in which new intervention of forestation and requalification were made progressively in the past years (Figure 2). Despite its recent creation, Parco
Nord cover a fundamental role in the ecological and social requalification of the North of Milan and civic support is still very high with local associations of citizen actively involved in the safeguard of the park.

Figure 2. Requalification process of Parco Nord: the first interventions date to 1983. Green areas represented new intervention of requalification (forestation, land remediation, establishment of meadows). Today, almost 350 hectares of the park are green areas (forests, clearings, rows, bush shrubs, hedges and little water bodies).

2.2 Participatory mapping of CES

Public participation geographic information systems (PPGIS) refers to spatially explicit methods and technologies for collecting and using non-expert spatial information (Brown and Fagerholm, 2014). It is used to inform planning processes with public knowledge by inviting participants to provide
geospatial information about perceived attributes of place (Sieber, 2006). Participatory mapping is a method that can be used to assess and map CES (Brown and Kyttä, 2014; Brown and Reed, 2009). In PPGIS mapping, information is solicited by requesting participants to identify and mark locations on a map about perceived place attributes. A common approach for soliciting and collecting spatial information using PPGIS include participatory workshops. Diversity of approaches to participatory mapping (map attributes, sampling, purpose, technology and location) provide a large number of mapping design options. In order to set our design we made explicit the followings (Brown and Fagerholm, 2014):

<table>
<thead>
<tr>
<th>Choice of map attributes (what is mapped?)</th>
<th>Cultural benefits</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sampling (who does the mapping?)</td>
<td>Park users (citizens, visitors, ...)</td>
</tr>
<tr>
<td>Purpose (reason for mapping?)</td>
<td>Identify which cultural benefits are perceived by citizen, how they are spatially allocated in the park and how important they are</td>
</tr>
<tr>
<td>Technology (how is mapping done?)</td>
<td>Throughout public workshops</td>
</tr>
<tr>
<td>Location (where is mapping done?)</td>
<td>Entire area of Parco Nord (Milano)</td>
</tr>
</tbody>
</table>

In order to collect data about people perception of cultural services we organized public workshops involving citizens during spring and summer 2016. Workshops were organised with the collaboration of the park authorities as well as local cultural association and promoted through public events, internet and official mailing lists. Workshops were held in the main building of the park where usually cultural activities take place (Cascina Centro Parco) and also in some local association’s buildings close to the park. Each workshop was about 2 hours long, divided into two part. First, workshop started with an introduction into and clarification of the ecosystem services framework, cultural benefits were presented and described (Table 1) and the purpose of this study presented (Plieninger et al., 2013). Then, respondents were familiarized with the map of the study area and informed about
the mapping process (Plieninger et al., 2013). In the second part of the workshop we used a four-page questionnaire (Appendix 1) and the map of the park. The questionnaire comprised: 1 - the list of cultural benefits with the codes to use for the mapping; 2 - the list of cultural benefits to which give a score (from 0 to 10); 3 - 13 questions on socio-demographic and site-specific information (about the park) and 4 – a blank page where people were free to add impressions about the exercise. The interviews centered on the following question: “Where in this area do you find or use cultural services? identify on the map the places in the park where you perceived (find or use) cultural benefits” (Peh et al., 2013; Plieninger et al., 2013). During the process of mapping participants marked on the map (using pencils or markers) and link the places marked to an acronym so that it was possible to identify which benefit was located where (Wolf et al., 2015; Fagerholm et al., 2012; Klain and Chan, 2012). Maps and questionnaires were made individually by people but group discussion was allowed and one or more expertise were supervising the exercise and assisting people in the mapping process. This interaction can assist with orientation on the map and may result in more reliable spatial data (Wolf et al., 2015).

2.3 Map of park management

We used a simplified map of the park in order to have a limited number of defined spatial units to elaborate the resulting maps of CES. Cultural benefits arising from park’s management were mapped consulting the official plan of the park (Figure 3), the WebGis page of the park (http://www.parconord.milano.it/vnlib/pmapper-dev/map.phtml) integrated with our knowledge of the park. Presence of some features were univocally intended as intention to provide cultural benefits (i.e. presence of play area for kids were intended to provide recreational, social, mental and physical health benefits; presence of historical monument were intended to provide cultural heritage, aesthetic and sense of place benefits; or the presence of a walk track into a forest stand were intended to provide aesthetic, creative or artistic inspiration, education and ecological knowledge, mental and physical
health and leisure, recreation and eco-tourism). These information were integrated with researcher knowledge of the park (i.e. places or buildings that are important for cultural activities or used for environmental education). This allowed us to have the map of the cultural values perceived by the park management to provide.

2.4 Data analysis

Data collected from respondents were entered into an Excel database and transformed into a GIS. The map used for this purpose was the land use and land cover map of the park simplified. Cultural benefits were attributed to each polygon of the map if one point or one line drawn by people were intersecting that polygon. For cartographic representation, we merged the absolute number of entries by respondents for each polygons and displayed the perceived cultural services on separate maps. In addition, we conducted the mapping of aggregated patterns of cultural ecosystem (Plininger et al., 2013) and for this purpose richness (= the number of different benefits per land unit) was calculated for both the participatory map and the park management map. All the cartographic elaboration were made using ArcGIS 10.2.2.
Figure 3. Land use and land cover map of Parco Nord (not simplified).
Table 1. List of cultural benefits used in this study (from TESSA toolkit - Peh et al., 2013).

<table>
<thead>
<tr>
<th>Cultural benefits (CODE)</th>
<th>Description</th>
<th>Examples</th>
</tr>
</thead>
<tbody>
<tr>
<td>Spiritual and religious (SP)</td>
<td>Benefits derived from specific places, features or species within a natural landscape creating sacred, religious or spiritual inspiration, feelings and values Sites important for spiritual or religious reasons, rituals and ceremonies Religious rules and taboos Links to ancestors, gods or spirit world</td>
<td>• Holy places • Holy springs • Sacred forests • ‘Wish-fulfilling lakes’ • Places where ancestral spirits are believed to reside • Sacred sites used in rainmaking ceremonies • Species considered sacred.</td>
</tr>
<tr>
<td>Cultural heritage (CULT)</td>
<td>Benefits derived from or associated with natural, semi-natural or culturally important landscapes, sites or features, that provide reminders of and retain historic roots Connections to the past providing a sense of continuity and understanding of place ‘Memories’ from ties to landscapes Values associated with physical objects, places, practices, traditions, or languages passed on from generation to generation, linked to landscapes, settings, places or culturally significant species</td>
<td>• Special old trees • Remains of traditional cultivation systems • Historic artefacts e.g. historic records preserved in water bodies and soils • Settings (locations or landscape features) related to traditional story-telling • Historic gardens and landscapes</td>
</tr>
<tr>
<td>Aesthetic (EST)</td>
<td>Benefits derived from scenery, sights, sounds, smells and touch Pleasures associated with appreciation of landscape aesthetics, in particular, scenic beauty Values attributed to sense of open space, wilderness, water features and landforms Benefits associated with the ‘beauty of nature’ including natural, semi-natural and managed landscapes</td>
<td>• Opportunities for aesthetic enjoyment of nature and scenic views • Beautiful trees or flowers • The sound of trees in the wind or birds calling • The smell of fresh air, tree blossom or growing fruits • The feeling of walking through tall grasses</td>
</tr>
<tr>
<td>Inspiration, creative or artistic (ISP)</td>
<td>Benefits derived from nature as a source of inspiration for paintings, sculptures, poetry, music, weaving, architecture, advertising, etc. or as the basis of myths, folklore and national symbols Inspiration characterized as enrichment, experience, solace, enlightenment, fulfilment, renewal, and reflection</td>
<td>• Artistic representations of nature • Use of natural motifs or artefacts in art and folklore • Aboriginal rock art • National emblems inspired by plants and animals • Music inspired by the sound of water babbling in a stream or bird song</td>
</tr>
<tr>
<td>Cultural benefits</td>
<td>Description</td>
<td>Examples</td>
</tr>
<tr>
<td>-------------------------</td>
<td>-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------</td>
<td>---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------</td>
</tr>
</tbody>
</table>
| Sense of place and      | Benefits derived from “sense of place” associated with environmental settings or feature of the natural environment that provides a sense of belonging, relations, or connectedness | • Seeing a familiar landmark or feature - a mountain, savannah, a special tree, an endemic animal or plant  
• Experiencing a sound or smell associated with a particular natural or semi-natural setting or feature  
• Unique features in the landscape that represent ‘home’  
• A fisherman’s sense of self or identity gained from the act of fishing and being a ‘fisherman’  
• Cultural identity associated with the presence of certain habitats or species  
• The opportunity to conduct cultural practices or activities important to sense of identity |
| identity (APP)          | Feeling “at home” creating a sense of fulfilment, meaning, ownership, empowerment and commitment and contributing to the need for protection and affection | Benefits derived from cultural linkages between humans and the environment  
A sense of identity achieved through interactions with nature that give a sense of who and what someone is, within family, community, universe  
A sense of self experienced through interactions with environmental settings and species  
Interactions with nature that shape identity and vice versa |
|                         | Benefits derived from types and quality of social relations and interactions in environmental settings | • A large tree providing shade for community meetings  
• Festivals held to celebrate an ecosystem or landscape feature  
• ‘Greenery’ that leads to greater use of common spaces for face to face social contact  
• Certain plants or animals that have specific roles in social and activities  
• Social relations in fishing communities differ in many respects from those nomadic herding or agricultural societies |
| Social relations/       | Benefits derived from types and quality of social relations and interactions in environmental settings |                                                                                                                                                                                                 |
| community benefits      | Settings, features or species that facilitate positive social interactions between individuals, communities and groups  
Places for social groups to gather  
Opportunities for group hunting or collecting activities which create family or social cohesion and group sharing  
Contributions to wellbeing from social interaction  
Promoting social networks  
Fostering social capital and enhance social wellbeing |                                                                                                                                                                                                 |
| (REL)                   | Contributing to wellbeing from social interaction  
Promoting social networks  
Fostering social capital and enhance social wellbeing |
<table>
<thead>
<tr>
<th>Cultural benefits (CODE)</th>
<th>Description</th>
<th>Examples</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Education and ecological knowledge (ED)</strong></td>
<td>Benefits derived from formal and informal knowledge systems developed by different cultures. Subject matter for education, knowledge and research. Meeting the need for understanding. Opportunities for outdoor learning where observation, experience and experimentation leads to increased ecological knowledge and enhanced connectedness to nature. Positive relationships with nature based on experience and knowledge. Motivating more sustainable ecosystem management. Enhanced knowledge for other disciplines through improved cognitive outcomes, increased enjoyment of education, better behaviour and improved working conditions.</td>
<td>- Ecosystems that provide information for cognitive development. - Increased understanding about species and ecosystem through visits to nature areas. - Direct observation and experience of nature, deepening understanding. - Traditional ecological knowledge gained through interactions with nature. - Traditional knowledge of biodiversity which lies in the memory of local and indigenous communities and is transmitted through daily practices, stories, songs and dance.</td>
</tr>
<tr>
<td><strong>Health - mental and physical (SAL)</strong></td>
<td>Benefits derived from environmental settings that have positive impacts on a wide range of health outcomes. Places where people can undertake physical activity and interact with nature enabling the restoration, maintenance, and/or development of emotional, mental and physical health and well-being. Viewing or being in an environmental setting that contributes to physical, emotional and mental health and wellbeing.</td>
<td>- Contact with nature that improves health by providing a sense of calm and tranquillity. - Positive changes in mood experienced through walking in natural environments. - The presence of attractive nature, that stimulates increased levels of physical activity leading to improved physical health. - Cultural ties to a landscape that strengthen self-esteem. - Viewing nature improves emotional wellbeing.</td>
</tr>
<tr>
<td><strong>Leisure, recreation and eco-tourism (RIC)</strong></td>
<td>Although TESSA considers nature-based recreation in an earlier module, there is a focus on economic valuation which may not be appropriate – for example, if visitors are not charged to use the site. Therefore it may be appropriate to apply this module.</td>
<td>- Hiking. - Bird watching. - Dog walking. - Football fields. - Boating. - Diving. - Viewing iconic/rare species.</td>
</tr>
<tr>
<td><strong>Existence/bequest values (INT)</strong></td>
<td>Some people value knowing that particular natural areas, wild species, or special natural feature exist irrespective of their own use, or the use of others - they just value knowing they exist and/or are protected or preserved for the enjoyment of future generations.</td>
<td>- Value placed on knowing that tigers exist in the wild irrespective of any plans to visit them. - Benefits received from knowing that a species is protected for the next generation to enjoy.</td>
</tr>
</tbody>
</table>
3. Results and Discussion

3.1 Cultural ecosystem services perceived by parks dwellers

*Demographics*

People participating to the study were 31 (sex ratio 19 male:12 female). More of the half of the participants were between 51-70 years-old and age class less represented was that of 20-30 and 41-50 years old (Figure 4).
Figure 4 (previous page). Demographic profiles of respondents based on questionnaires. On the $x$ axis are represented values in percentage, while the number close to the bars represents the absolute number of respondents ('none' when the answer was not given).

Most of the questioned visitors lived close to the park (less than 15 minutes travel) and in the surroundings municipalities (mainly Cinisello Balsamo and Milano) (Figure 4). A large majority of the respondents visited the park at least one or three times a week (Figure 5) and spend there in average 1 hour or half a day. Respondents visited the park mainly for walk, sport, relax (in ‘other’), meet people and for volunteering (Figure 5). Other reasons for visit the park not listed in the questionnaire but added by respondents were: for environmental activities, work, cultural events, naturalistic photography, birdwatching, bike and recreational activities.

Figure 5. Statistics about park visiting based on questionnaires. On the $x$ axis are represented values in percentage, while the number close to the bars represents the absolute number of respondents ('none' when the answer was not given).
Perceived cultural benefits

Cultural benefits scored higher by people were those related to education and ecological knowledge, health (mental and physical), bequest values, aesthetic and cultural heritage (score > 8, where 10 was the maximum) (Table 2). On average, all the benefits (except those related to spiritual and religious values and to artistic inspiration) were scored quite high (> 7.5). The score defined how much people believe a benefit is important to be delivered in an urban park, and do not refer to specific locations inside Parco Nord. This helped us to determine the absolute ‘weight’ of a benefit, and does not resulted biased by the association with some specific location in the park (that may bias the way people weight the benefit). Cultural services were not scattered randomly across a landscape, but rather follow specific patterns in terms of the intensity of their provision (=number of marks for each site) leading to the expression of distinct hotspots and coldspots of cultural services (van Berkel and Verburg, 2012; Willemen et al., 2008) (Figure 6).

Table 2. Cultural benefits as scored by people.

<table>
<thead>
<tr>
<th>Code</th>
<th>Benefit</th>
<th>Mean</th>
<th>dev.std</th>
</tr>
</thead>
<tbody>
<tr>
<td>ED</td>
<td>Education and ecological knowledge</td>
<td>9.42</td>
<td>0.94</td>
</tr>
<tr>
<td>SAL</td>
<td>Health - mental and physical</td>
<td>8.97</td>
<td>1.20</td>
</tr>
<tr>
<td>INT</td>
<td>Existence/bequest values</td>
<td>8.57</td>
<td>1.31</td>
</tr>
<tr>
<td>EST</td>
<td>Aesthetic</td>
<td>8.42</td>
<td>1.26</td>
</tr>
<tr>
<td>CULT</td>
<td>Cultural heritage</td>
<td>8.13</td>
<td>1.16</td>
</tr>
<tr>
<td>APP</td>
<td>Identity and sense of place</td>
<td>7.90</td>
<td>1.49</td>
</tr>
<tr>
<td>RIC</td>
<td>Leisure, recreation and eco-tourism</td>
<td>7.90</td>
<td>1.47</td>
</tr>
<tr>
<td>REL</td>
<td>Social relations / community benefits</td>
<td>7.45</td>
<td>1.68</td>
</tr>
<tr>
<td>SIP</td>
<td>Inspiration, creative or artistic</td>
<td>6.77</td>
<td>1.58</td>
</tr>
<tr>
<td>SP</td>
<td>Spiritual and religious</td>
<td>5.39</td>
<td>2.47</td>
</tr>
</tbody>
</table>

The scores given to benefits highlighted that citizen believed an urban park to be important mainly for its function of education in ecological knowledge. Interestingly, education in ecological knowledge was also one of the benefits most frequently marked on the map (second benefits for the total number of marks, with a hotspot -for the single benefit- in correspondence of the Cascina Centro
Parco, where the main cultural activities normally take place). Ecological knowledge has been defined as ‘accumulated knowledge about nature’ and can be acquired through contact with different natural environments, directly or indirectly as the category includes codes on formal and informal education and aspects of learning from each other about nature (Maraja et al., 2016). Environmental settings are valuable surroundings for outdoor learning where engaging with nature can lead to enhanced connectedness to nature and increased ecological knowledge (Church et al., 2014). The park is also expected to be an important place for mental and physical health, and the positive effects of nature experience on human health and well-being has been extensively documented (Lafortezza et al., 2009; Van den Berg et al., 2007). Health benefits were reported quite frequently by respondents as well (fourth benefit for the total number of marks) and they were mainly placed in correspondence of some natural feature (forest, lake). The environmental settings of the park provided places where people can undertake physical activity and interact with nature. Levels of interaction/engagement of green spaces have been linked with longevity and decreased risk of mental ill-health, as well as long-term health (through the role of vitamin D obtained from sunshine) (Church et al., 2014). Similarly, people who used domestic gardens and local green spaces at least once a month also showed better self-reported health, measured by physical functioning and emotional well-being, compared to those who do not (Church et al., 2014). The ‘green exercise’, defined as any physical activity taking place in the presence of nature, is predicted to lead to positive health outcomes, as well as promoting ecological knowledge, fostering social bonds and influencing behavioural choices. Benefits related to community and social relations were scored among the lower in terms of importance for people, but they were perceived frequently in the area of the park and mainly in correspondence of Cascina Centro Parco and the entrance of via Giancarlo Clerici. Community and social relations included views of a park as a place for social integration, to overcome the loneliness in a city and to strengthen personal social relations (Maraja et al., 2016). Open spaces are normally used as a resting or meeting point, for festivities, as a living or dining space: for people who do not have home gardens or balconies, urban green spaces provide crucial opportunities to leave the apartment and have an
outdoor place to meet (Maraja et al., 2016). Recreation benefits and benefits related to identity and sense of place were ranked equal on average (7.9). **Recreational benefits** were perceived frequently and were mostly associated with presence of facilities (play area, bocce area or walk path). Recreational benefits are mainly identified as the possibility of relaxing or pleasant activities like sport, play, walk or do practical work like gardening. A low perception of benefits related to **identity and sense of place** was common for the whole park. **Cultural heritage** was scored high on average (8.13) and its perception was distributed heterogeneously in the park with some hotspots in correspondence of historical features (i.e. Monumento al Deportato, Villa Manzoni) and to other places with no specific historic or cultural features. Cultural heritage values, identity and sense of place are referred mainly as special or historic features within a landscape that remind us of our collective and individual roots, providing a sense of continuity and understanding of our place in our natural and cultural environment and may thus may be conceptualised as landscape-related “memories” (MA, 2005). In this context, the low perception of benefits related to identity and sense of place may be due to the recent establishment of the park, that even from one side was strongly supported by civic engagement, from the other side it does not yet represent an historical heritage. However, remnants of agricultural landscapes that still exists in the park may link the present landscape to cultural heritage. **Aesthetic benefits** were mainly perceived in correspondence to natural features (forest, lake, meadow, tree row) or architectural features. Aesthetic values addressed a feeling of beauty and other studies have found that they often emerged from diversity in landscapes, from rivers or lakes or from a broad panorama view or they are mostly related to green spaces that seem “natural” and do not show signs of human-built construction (Maraja et al., 2016). Benefits related to **artistic inspiration** were low perceived and low scored. **Spiritual benefits** were low scored and clearly perceived in correspondence of the cemetery of Bruzzano, cemetery of Brusuglio and Monumento al Deportato. People valued among the higher the **bequest value** of a park, that is the value disconnected from direct benefits people may obtain from ecosystems and thus its value is irrespective of the fruition of the park. In this case, the park is valued not only for contributions to
valued experiences, but also simultaneously and sometimes inseparably for its existence, independent of experience (Chan et al., 2012b). This value was also the most frequently perceived in the park. All the respondents defined the perception of this value ‘for the whole park’ without specifying any particular location. The importance of such metaphysical value reflected altruistic, existence motivations values and can be self-oriented (existence value) or other-oriented (e.g., bequest value), and based on virtues, principles, or preferences (Chan et al., 2012b).
3.2 Comparison of maps of CES

Maps of richness of CES (Figure 7) were obtained merging the number of cultural benefits perceived (by people) and intended to be furnished (by park). Maps showed places that have 'bundles' of different values. Resulting hotspots and coldspots of ecosystem services provision is consistent to what found in other studies and are normally related to landscape features, land cover forms or to
specific features of the area (Plieninger et al., 2013; Bryan et al., 2010). Based on the maps obtained, cultural benefits perceived by people resulted to be higher (in richness) than those park management intended to furnish. A part from the number of benefits, some hotspot (in terms of boundles of benefits) matched among the maps: in particular, the area surrounding the entrance of via Giancarlo Clerici, the north areas with forests and extended meadows and the area close to via Giuditta Pasta (north and south).

Figure 7. (A) Map of cultural benefits perceived by people (richness) and (B) Map of cultural benefits perceived by park management (richness).

Coldspot and areas with lower number of perceived benefits by people were in correspondence of the building area of Giancarlo Clerici, the area close to the entrance of via Ruvenzori, the area of Stadio Brera and the area of via Campestre (despite the presence of the cultural area of Oxygen). Higher richness of CES perceive by people was is part due to the fact that some benefits were marked by people ‘for the whole park’, without specify any specific location. This lead each polygon of the park to have not less than 6 benefits. On the contrary, benefits in the map of the park management were
never marked ‘for the whole park’ but considering the presence of specific feature in each polygons. The many positive correlations between assigned cultural benefits to the same places showed that there is considerable overlap between individual services, indicating that people may do not clearly separate one cultural service category from the other (Plieninger et al., 2013). This can be understood - and appreciated - as evidence of the interlinked, holistic nature of cultural ecosystem services (Bieling and Plieninger, 2012; Daniel et al., 2012) and these results further confirm the “bundled” occurrence of cultural services (Raudsepp-Hearne et al., 2010). These results documented also that people find various cultural values in their everyday surroundings and not only in landscapes of outstanding biodiversity, heritage, or scenery (Plieninger et al., 2013) and that they can furnish spatial information about its perception. Aggregation indices of cultural services (i.e. richness) point to different facets of cultural values in landscapes and are useful for identifying priority areas (Bryan et al., 2010).

3.3 Consideration about this methodological approach: opportunities and needs

Perception mapping can shed light into how urban parks are perceived and experienced by citizens. It has been demonstrated that spatially explicit information on cultural ecosystem services that incorporates the differentiated perceptions of the local population provided a rich basis for the development of sustainable land management strategies (Lynam et al., 2007; Brown, 2005). Several studies supported the view that a collaborative, demand-side assessment of cultural services should become part of landscape planning (Termorshuizen and Opdam, 2009), thus incorporating the value that local people attach to landscapes and overcoming the one-sidedness of expert based approaches (Martínez-Harms and Balvanera, 2012). Urban planners and policy makers are increasingly aware of the need to take the citizen (or user) perception aspect of urban nature design and use into account, and there are many different ways in which they attempt to study this subject (Buchel and Frantzeskaki, 2015).
However, cultural services differed in various aspects from other ecosystem services, presenting strong barriers toward their broader incorporation (Chan et al., 2012a,b). Likewise, management plans are not designed to take the intangibility of cultural benefits into account. The mapping of ES using PPGIS is a relatively new field that offers a supplemental approach to expert-driven ecosystem service mapping and modelling (Brown and Fragerholm). Many authors have found that a generally weak relation between map's purposes with the used procedures could explain the restricted incidence of ES on decision-making by limiting the transmission, comparison and synthesis of results (Nahuelhual et al., 2015). Other authors claimed that there is a strong demand for approaches that are able to involve local governance networks and move the ecosystem services research out of the static mapping and evaluation approaches (Opdam, 2013). Because the PPGIS for mapping ecosystem services is relatively recent, it is not surprising that there are few or any examples in the literature describing how mapped ecosystem service data are used for actual decision support (Brown and Fagerholm, 2014).

In this context, the methodology presented in this study move towards a mapping process that, through the comparison analysis, resulted in data directly informative for urban planning. From one side, it provided spatially explicit data about perceived cultural services of the park. By develop a framework of fine-grained of perceivable ecosystem services park users perception was captured (Buchel and Frantzeskaki, 2015). Moreover, it provided information about the matching or mismatching patterns about cultural services provision comparing the users view with the management dimension. In order to be useful at the site scale, the method produced data relevant to decisions affecting that site, that are practical and affordable (in terms of expertise, equipment and time) and that provide results in an accessible form to actors such as policy-makers, planners and land managers (Peh et al., 2013). PPGIS integrated with a comparison analysis may help in offer a practical toolset for efficiently capturing and analyzing stakeholder preferences, allowing managers to make informed decisions and understand tradeoffs (Cox et al., 2014). Moreover, the density of cultural values and the types of values that congregate in different places can help decision-makers anticipate
conflict among user groups (van Riper et al., 2012). Mapping cultural services might be informative also in order to detect if there are conflicts between nature conservation and recreation that might happen in particular around densely populated areas (Niemela et al., 2010). In such cases, urban planners has the task to generate diversified and good-quality local recreational services, alongside nature conservation areas (Arnberger and Brandenburg, 2007).

However the methodology proposed here, that is intended as a preliminary experimental design, still present some limitation. Hereafter we proposed some improvements that we believed should be addressed for better reduce the gap between CES mapping and effective information for decision-making. First, cultural benefits perceived from park management were mapped by experts based on specific features of the plan of the park (as CES are generally related to landscape features or land forms) (Plieninger et al., 2013; Bryan et al., 2010). Considering the holistic and non-tangible nature of CES this mapping process may result in underestimation of CES that the park management intended to provide to the users. In this view, the integration of this mapping (i.e. with parks management interviews) may resulted in a more accurate report of data. Second, in order to explore the opportunities of the data resulted from the maps comparison in inform land-use planning, a discussion with park management would represent a valuable way to link the mapping exercise with the local governance. This would have the double effect to investigate the utility of the data produced with the participatory mapping and to bring the park management closer to the recognition of the cultural services dimension. In this context, Parco Nord may offer a unique opportunity to explore the possible implementation of CES mapping and CES map comparison into planning process thanks to the peculiarity of being an ‘in progress’ park.

4. Conclusions

Cultural ecosystem services are generally overlooked by conventional biophysical and economic assessments and, therefore, they are under-appreciated compared to other more easily quantifiable
ecosystem services (Gee and Burkhard, 2010; Norton et al., 2012). Perception mapping of cultural benefits can signify the importance of existing urban green space from the user’s stand point rather than from planners’ stand points (Peschardtetal.,2012;Bauretal.,2013) and may provide a strong entry point for improving human-nature interactions in cities and help to meet both socially acceptable and ecologically functional sustainability goals (Andersson et al. 2014). Despite remaining methodological challenges, cultural services mapping assessments should be pushed ahead as indispensable elements in the management and protection of landscapes (Plininger et al., 2013) and this study represented an attempt to better integrated CES mapping and land use planning within the urban context.

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Appendix of Chapter V
Four-pages questionnaire presented during the participatory mapping (original questionnaire was in Italian).

**QUESTIONNAIRE - PART 1**

WHERE IN THIS AREA DO YOU PERCEIVED CULTURAL SERVICES?
IDENTIFY AND MARK ON THE MAP THE PLACES IN THE PARK
WHERE YOU PERCEIVED (FIND OR USE) THE FOLLOWING CULTURAL BENEFITS.

*(Remember to always link the places marked on the map with the CODE of the corresponding cultural benefit.)*

<table>
<thead>
<tr>
<th>CODE</th>
<th>Cultural Benefits</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>SP</td>
<td>Spiritual and religious</td>
<td>I value this place for its spiritual or religious values.</td>
</tr>
<tr>
<td>CULT</td>
<td>Cultural Heritage</td>
<td>This place has an historical past (social, natural) that link to my cultural heritage.</td>
</tr>
<tr>
<td>EST</td>
<td>Aesthetic</td>
<td>This place have an aesthetic value (scenary, landscape, sounds or smells).</td>
</tr>
<tr>
<td>ISP</td>
<td>Inspiration, creative or artistic</td>
<td>This place is a source of inspiration (for photography, painting, writing, music and other artistic or creative arts).</td>
</tr>
<tr>
<td>APP</td>
<td>Sense of place and Identity</td>
<td>This place produce a feeling of belonging and identity (to fell 'at home', a feeling of familiarity, to identify yourself in the place and in its characteristics).</td>
</tr>
<tr>
<td>REL</td>
<td>Social relations / community benefits</td>
<td>This place allow social interaction and relationship opportunities (friends, family, and community).</td>
</tr>
<tr>
<td>ED</td>
<td>Education and ecological knowledge</td>
<td>This is a place where ecological knowledge and environmental education can be developed.</td>
</tr>
<tr>
<td>SAL</td>
<td>Health - mental and physical</td>
<td>This place contribute to mental (emotional, serenity) and physical (open-air activities) health.</td>
</tr>
<tr>
<td>RIC</td>
<td>Leisure, recreation and eco-tourism</td>
<td>This place give recreational and/or touristic opportunities (walk, run, dog, play with friends, observe flora and fauna)</td>
</tr>
<tr>
<td>INT</td>
<td>Existence/bequest values</td>
<td>This place has its own intrinsic value, regarding to my utility. I value the fact that this place exists and will exist for future generations.</td>
</tr>
</tbody>
</table>
QUESTIONNAIRE - PART 2

HOW MUCH THIS BENEFITS ARE IMPORTANT FOR YOU?
(think about how much do you think these benefits are important in an urban park).

Give a score to each cultural benefits from 1 to 10, where
1 = minimum (I don’t think it is important at all),
10 = maximum (I think it is of fundamental importance)

<table>
<thead>
<tr>
<th>CODE</th>
<th>Cultural Benefits</th>
<th>Score (min = 1, max = 10)</th>
</tr>
</thead>
<tbody>
<tr>
<td>SP</td>
<td>Spiritual and religious</td>
<td></td>
</tr>
<tr>
<td>CULT</td>
<td>Cultural heritage</td>
<td></td>
</tr>
<tr>
<td>EST</td>
<td>Aesthetic</td>
<td></td>
</tr>
<tr>
<td>ISP</td>
<td>Inspiration, creative or artistic</td>
<td></td>
</tr>
<tr>
<td>APP</td>
<td>Sense of place and Identity</td>
<td></td>
</tr>
<tr>
<td>REL</td>
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<td></td>
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<td></td>
</tr>
<tr>
<td>SAL</td>
<td>Health - mental and physical</td>
<td></td>
</tr>
<tr>
<td>RIC</td>
<td>Leisure, recreation and eco-tourism</td>
<td></td>
</tr>
<tr>
<td>ES</td>
<td>Existence/bequest values</td>
<td></td>
</tr>
</tbody>
</table>
QUESTIONNAIRE - PART 3

- Age:  g 20-30  g 31-40  g 41-50  g 51-60  g 61-70
- Gender:  g M  g F
- Employment:  g student  g retired/unemployed  g part-time worker  g full-time worker
  g other: ______________________________________
- Education:  g primary school  g secondary school  g high school  g university
- Do you have children (less than 14 years old)?  g yes  g no
- Do you have a dog?  g yes  g no
- Where do you live? (indicate the municipality)
  ____________________________________________________________
  in  g apartment  g house with garden  g other
- How far do you distance from the park?
  g less than 15 minutes  g between 15-30 minutes  g between 30-60 minutes  g more than 1 hour
- Do you work close to the park?  g yes  g no
- For what reasons do you visit the park?
  g walk  g children  g sport  g dog  g meet people/friends  g launch-break  g I do volunteer for the park  g other: ____________________________________________________________
- How often do you visit the park?
  g every day (almost)  g at least three times a week  g at least one time a week  g at least one time per month  g only during festivity/events  g other (specify __________________________________________)
- How long does every visit last on average?
  g 1 hour  g half a day  g one day  g other (________________________________________)
GENERAL CONCLUSIONS

Ecosystems, through their normal functioning, provide a range of goods and services important for human well-being, which are collectively called ecosystem services (ES) (Nelson et al., 2009, Raudsepp-Hearne et al., 2010, Rounsevell et al., 2010). Cities, just as any other social-ecological system, depend on ecosystems and their components to sustain long-term conditions for life (Seto et al., 2013). At the same time, urban influence on ecosystems has increased the extent (Pirnat and Hladnik, 2016), intensity (Ode and Fry, 2006) and irreversibility of changes (Kasanko et al., 2006). In the future, an increasing number of people will face ES losses as the urban population and associated human activities increase exponentially (Bolund and Hunhammar, 1999; Douglas, 2012). Because of their importance, a deeper knowledge about ES provisioning in urban areas is strongly advocated.

In this research project, I have focused on the analysis of multiple ecosystem services provided in one of the most extended and densely populated area of Italy, the urban region of Milan, considering urban forests and green areas as the service providers (Ziter, 2016). Ecosystem services in this research were analysed separately, and each study represented an attempt to give new insights into specific issues arising from single ES assessment. The selection of ES generally depends on available data and the regional peculiarities of the observed ecosystem which often results in a subjective choice (Elmqvist et al., 2013; Seppelt et al., 2012). I chose four ES of special interest for the Milan urban context, focusing on regulating, provisioning and cultural services. Quantification of ES can be particularly difficult in areas containing complex land-cover mosaics, and that often represent “novel ecosystems” in terms of their composition (Wu, 2014). In this context, the urban region of Milan represents one of the most extended urban area of Italy, whose land cover composition and land cover changes are complex and where the continuous expansion of the urban area is a process still occurring and that will likely to occur in the next decades. The low urban density accompanied by the expansion
of the urban space that characterized sprawl cities like Milan is known to lead to, among others, loss of green spaces, increased functional land-use divisions, polluting emissions and social segregation. Demand-driven ES approach thus represented a suitable strategy to ameliorate urban living conditions in densely populated metropolitan areas (Burkhard et al., 2012; Douglas, 2012). Urban sprawl causes a consumption of land thereby decreasing both the quality and quantity of available resources and ecosystem functions and processes (Metzger et al., 2006) or increasing the land vulnerability (Salvati et al., 2013; Travisi et al., 2010). The study presented here helped in identifying general trends of urban area sprawl, and also in recognizing which areas suffer from the higher pressures of urbanization and landscape fragmentation, thus help in implementing targeted policies to limit threats to the environment. In this context, balancing urban compaction and maintaining green space is a major challenge for urban planners (COM, 2001). In fact, in some cities that have adopted a compaction strategy, there is a conflict between the conservation of green spaces in a functional urban green infrastructure on the one hand, and exploitation interests for built-up environments on the other (Sandstrom et al., 2006). Although often provided at smaller spatial scales, and being more fragmented or disrupted than in rural or natural environments, urban ecosystem services affect and benefit all levels of Milan’s urban population, as in other metropolitan areas. Human health and well-being, and the amenity values of most cities worldwide, are strongly related to the quantity and quality of urban forests, which can be established on land of all types and quality, and do not need to compromise desires for higher population densities (Meurk et al., 2013). Considering the current and upcoming degree of urbanization, urban parks and forests play a crucial role for the conservation of many species and the maintenance of high biodiversity levels within cities is a challenging goal to undertake. In this research, I explored the habitat provision of urban forests for birds through a multiscale analysis and showed in which way different environmental requirements exist for biodiversity maintenance at different scales in the urban area of Milan and provided suitable data to inform an effective management of green spaces with the aim of support high level of biological diversity. Novel insights about how to better manage green areas within the city would arise from considering
the nature of the matrix that can have a profound effect on habitat use by different species, particularly in highly fragmented urban and rural landscapes (Pirnat and Hladnik, 2016). Green areas fragmentation in Milan appear to be important, and an assessment of the structural and functional connectivity of urban green areas is needed in order to allow the identification of key areas for ensuring biodiversity. Moreover, undertake consistent conservation actions would have the double result to preserve biodiversity and to provide opportunity for residents to experience nature and its diversity, enhancing the cultural benefits people derives from green areas in cities. Besides harvest biodiversity, urban parks and forests undertake other important ecological functions, in particular concerning the regulation of ecosystems. The regulative role of vegetation in urban areas (that contribute to reduce air pollution levels and offset greenhouse gas emissions) has already been recognised by previous studies (McPherson et al., 1998; Nowak et al., 2006). In this research, the potential of plant-associated bacteria for air bioremediation processes in urban areas was investigated.

The management of urban forests to enhance ecosystem service supply can be a cost-effective strategy to meet specific environmental standards or policy targets (Escobedo et al., 2011). The study conducted on *Platanus x acerifolia* leaves in different green areas in Milan represents a preliminary work, which need further investigations (in particular in the quantification of the actual contribution of bacteria in air pollutant removal per unit of leaf weight or leaf area under different environmental conditions, and at the evaluation of the efficiency of different plant-bacteria systems in air quality improvement). However, the contribution of trees in pollution removal through phyllospheric bacterial community represented an interesting field to explore with great potential for the provision of urban regulating services. The regulatory role of urban parks and forests pass also trough the contribution in organic carbon (OC) stock by soils. In Milan, the recent urban expansion has led to the consumption and degradation of the soils regardless to their quality. Trying to tackling with the limitation in the analysis of urban soils this research aimed to shed light on OC storage of urban soils comparing different land use and different typology of area (including urban parks and forests). Urban soils of Milan may contain high quantity of OC as well as be highly depleted, and even the estimation
still suffers of many uncertainties, they appear to be important contributors to regional OC stock. The study conducted in Milan showed how the complex heterogeneity of urban soils together with the uncertainty of their origins, their history and management are important factors that limit our capacity to model soils characteristics. The missing of a soil-landscape relationship in urban areas appear to be one of the major challenge that we need to face to enhance soil mapping and thus quantification of OC stocks at broader scales. As the soil-landscape paradigm (Hudson, 1992) seems to does not serve in the case of urban landscapes, it is not possible to rationalize the factors and the soil processes, given the absolute variability and unpredictability of the same in an urban environment. In view of the increasing conversion of rural to urban soils associated with drastic increases in urbanization, strategies are needed to strengthen C sinks in urban soils and vegetation (Lorenz and Lal, 2009). Some authors suggest that to cap urban sprawl C sequestration should accompany sustainable and dense development within cities (Lorenz and Lal, 2009), that is opposed to the sprawling development that characterized the Milan urban region. However, the ecological benefits of more compact city forms need to be critically assessed in relation to ecosystem services such as OC sequestration in above- and belowground pools (Tratalos et al., 2007). Apart from regulating services, protecting high quality soils in urban and peri-urban areas is valuable also for future option values involving food production or contributing to biodiversity through remnants or green belts (Meurk et al., 2013).

While urban green spaces are frequently acknowledged as important for the enjoyment and health of people (Tzoulas et al., 2007), the assessment of cultural ecosystem services (CES) in urban areas are less frequent than other ES assessment, in part because of methodological constrains (Ziter, 2016). In fact, conventional biophysical and economic assessments often resulted not adequate to capture CES value. Within this project, I assessed CES by mapping people perception and park management perception in the Parco Nord of Milan, giving information on how the park is perceived and experienced by citizens. The study presented a methodology to CES assessment that move towards a mapping process that, through the comparison analysis, resulted in data directly informative for urban planning. Mapping cultural services might be informative to detect if there are conflicts between
nature conservation and recreation that might happen in particular around densely populated areas (Niemela et al., 2010). In such cases, urban planners have the task to generate diversified and good-quality local recreational services, alongside nature conservation areas (Arnberger and Brandenburg, 2007).

To ensure the delivery of urban ES we need heterogeneous, multifunctional and accessible green infrastructure throughout our cities (Gomez-Baggethun et al., 2013). We now understand that it is both cultural and biological diversities that underpin resilience and sustainability (Andersson and Ostlund, 2006; TEEB 2010). This raises the question of how the social and ecological dimensions and their dynamics can be considered in creating a sound analytical framework that informs planning and governance (Haase et al., 2014). For example, urban green spaces can be regarded as a multifunctional system being important for urban biodiversity as well as recreation, coping with storm water and improving the local climate (Mazza and Rydin, 1997), and providing better public health (Grahn, 1994). While ES assessments in urban areas are becoming increasingly common in the literature (Haase et al. 2014), the extent to which we understand the ecology of urban ES provision remains unclear (Ziter, 2016). Several key requirements need to be satisfied if knowledge of ecosystem services is to be effectively translated into operational practice (Elmqqvist et al., 2015). This requires focus on the needs of planners and decision-makers on the one hand and urban dwellers on the other, the combining of ES approaches with sophisticated scenario-based land-use models, the involvement of cross-scale effects, the integration of multiple ES and linkages and the availability of highly diverse approaches for ES assessment (Daily et al., 2009; Elmqqvist et al., 2013).

If from one side urban forests and green areas in cities are essential multifunctional areas, which need to be understood in their role of multiple ES provider, from the other side trade-offs between ES are mainly unknown. Some ES may co-vary positively (more of one means more of another) e.g. enhance carbon storage and hence climate regulation, while other services may co-vary negatively (more of one means less of another) such as when increasing provisioning services may reduce many
regulating services (e.g. provision of agricultural crops may reduce carbon storage in soil, cultural services, etc.) (Elmqvist et al., 2015). The generation of some services may also result in other less desired effects (referred to as disservices) for example, the presence of street trees and peri-urban parks may be highly valued, but the risks of tree falls or social degradation deriving from the same ecosystems have a negative value. Both are products of the way in which the underlying ecosystems are managed, with the result that trade-off decisions have to be made (Elmqvist et al., 2015). Finding ways of assessing how multiple ecosystem services are interconnected and coupled to each other in “bundles” is one of the major research gaps on ecosystem services identified by the MA (Carpenter et al. 2009). Biodiversity is known to play a key role in the ecosystems functioning. However, the quantitative relationship between biodiversity, ecosystem functioning and ecosystem services is still poorly understood (Balvanera et al., 2014; 2016). For example, the valuation of the contribution of biodiversity to regulating services poses particular challenges (Elmqvist et al., 2015). In this view, cultural services that are frequently enjoyed in “bundles” may foster the orientation of ecosystem services management toward multifunctionality, that is a frequently expressed, but rarely achieved desideratum in land-use science and policy (Cowie et al., 2007; Plieninger et al., 2012). As such, the holistic nature of cultural services can help overcome the widespread tendency to design incentive tools for individual ecosystem services in isolation, which often has been accompanied by unintended side-effects on other ecosystem services (Plieninger et al., 2013). Many authors suggest that stronger awareness regarding socio-cultural valuation of CES may be of relevance for nature conservation and sustainable land management (Plieninger et al., 2013) because the direct experience by people lead them to be motivators for owning, managing, and conserving land (Chan et al., 2012a), often being even more important than traditional livestock or timber production (e.g. Bieling, 2004; Plieninger et al., 2012). In order to capture the full potential of cultural services assessments and to enhance planning and management, data would need to be combined with spatially explicit information on biodiversity and other ecosystem services (Raymond et al., 2009). Regarding to this, CES can combine with other ES and are usually interdependent: Chan et al. (2012b) argue that when
disentangling the multi-layered concepts of services, benefits and values, an interlinked view of ecosystem services emerged, in which multiple benefits and values both material and non-material can be produced simultaneously by the same system components (Andersson et al., 2015). The type of the existing trade-offs between ES is important because it have very different implications for the design and management of landscapes and from this derives that knowledge of these relationships is essential to ensure that policy decisions translate into operationally effective and outcomes.

This study, together with similar research dealing with ecosystem services, entered largely uncharted territories, and several challenges that needs to be tackle, still limits the broader integration of different ecosystem services assessment of the complex and highly dynamic socio-ecological system represented by our cities (Plieninger et al., 2013). There is a significant potential in cities to increase benefits from ecosystem services - access to green and blue spaces, recreation opportunities, clean air, quality of environment generally and biodiversity (Dominati, 2013). Milan, like every city, has potential to effectively lower the ecological footprint and progressively lead to improved quality of life for its population. Compared to natural ecosystems, in cities the contraction, degradation and fragmentation of habitat or natural populations, and the intensity or frequency of disturbance, lead the ecosystem to be more vulnerable to being overwhelmed by environmental disasters (Meurk et al., 2013). If we learn about natural processes and their role in sustaining ecosystems - especially in the urban ones we inhabit – and if we look after them and enhance them, we can derive both economic benefit, pleasure, and the full range of ecosystem services from nature in the city (Meurk et al., 2013; TEEB, 2010; MA, 2005). The unfolding story of ecosystem services will revolve around how we sum our human experience and enrich it by making sustainable use of natural processes, and develop an inclusive governance that services that ideal (Meurk et al., 2013).
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SUPPLEMENTARY MATERIALS

In this section are presented additional works dealing with the urban area of Milan I made during the PhD, that were not included in the framework of the research project.
1) Urban forests and biodiversity

Emilio Padoa Schioppa\textsuperscript{a} and Claudia Canedoli\textsuperscript{a}

\textsuperscript{a}Department of Earth and Environmental Sciences, University of Milano-Bicocca, Milan, Italy

Abstract

This is a book chapter into the \textit{Routledge Handbook of Urban Forestry} edited by Francesco Ferrini, Cecil Konijnendijk van den Bosch and Alessio Fini. The chapter aimed to give an overview of the importance of urban forests as suitable habitats to preserve biodiversity in the urban environment. We introduced the concept of biodiversity and the levels in which it can be expressed (genetic, species, habitat and landscape), explained the reasons for which it is important to preserve biodiversity and which flora and fauna can occur in urban forests. The occurrence of different taxa in urban forests such as birds, reptiles and amphibians, mammals, insects and other invertebrates and plants and exotic species were discussed. Further, we highlights how the structure of urban forests can influence biodiversity levels in regard to forests size and forest composition and tree diameter. Case studies are used as examples to illustrate which actions are needed to undertake in order to maintain and preserve biodiversity in urban environment.
2) The soundscape methodology for bird monitoring in urban parks

Giovanni Zambon, Alessandro Bisceglie, Emilio Padoa-Schioppa, Claudia Canedoli

Department of Earth and Environmental Sciences, University of Milano-Bicocca, Milan, Italy

Abstract

The soundscape is a fundamental component of the landscape. It can be defined as every acoustic environment, natural, urban or rural, that can be composed of three fundamental elements: biophonies, geophonies and antrophonies. In natural and semi-natural environments birds contribute most to the soundscape composition, especially during singing activity picks. At the same time they are immersed in the acoustic environment and thus subjected to its influence, to the extent that some of their physiological and ecological aspects can be altered. Soundscape is an important tool because it reflects physic and informative properties of environmental acoustic signals and it contributes to the evaluation of animal diversity and community health status and their state of well-being. The aim of this study is to apply an Acoustic Complexity Index (ACI) to evaluate the spatial and temporal variability of the singing activity of a bird community in an urban park (subjected to intense noise) and to identify potential patterns of sound activity in relation with increasing distances from the main noise sources.

The study has been conducted in a wood lot of approximately 19 hectares, in Parco Nord (Milan). The study area is affected by constant anthropogenic noise, among which traffic noise from A4 highway in the north of the area. We recorded sounds with LCR recordings in 22 sites distributed in a regular grid in the wood lot (at an increasing distances from the main source of noise). Each recording was 4-5 days duration and was repeated for 6 sessions from April to June. Simultaneously to one session, we conducted a campaign of assisted traditional phonometric measures and traditional noise indexes (Leq e L95) were extracted and used for a comparison with ACI values. ACI has been calculated by extrapolating the recordings from 6.30 am to 8.30 am, period in which the birds singing
activity is more intense. The index ACI is an algorithm created to measure the acoustic spectrogram complexity obtained from a linear scale of acoustic intensity and, more precisely, to analyse the soundscape produced by birds (Pieretti et al., 2011; Farina and Morri, 2008).

The measures with traditional sound level meter highlight the negative relationship between vocalization abundance and proximity to a disturbance source. However, from an evaluation of the spatial (on the recording grid) and temporal (during the progress of the season) trend of ACI values, no clear patterns emerged and ACI does not show an univocal response to the increasing of the distance from the main anthropic noise source (the highway). This result can be due to the fact that the study area is too small in size and the highway could be not the major source of disturbance, but multiple and complex source of noise act simultaneously around the wood lot, like the Bresso airport, the Bassini hospital and a school. The study area could be too much immersed in the urban environment, leading responses to noise to be non linear in space and thus hard to find some relevant relations between the ACI index and the distance from noise sources.

From the methodological point of view, LCR recordings have shown some advantages (a very low economic investment, they can work without an operator, with the possibility to obtain a large number of recordings at the same time covering large areas). At the same time they showed some disadvantages that have partially invalidated the quality of data quality (low quality microphone, low spectral sensibility -8kHz- and malfunctioning on the field).

This study experimented how the passive recording of environmental sounds through LCR is a non invasive survey method that allows us to obtain a considerable amount of data without disturbing or altering the birds behaviour, object of the study. Nevertheless from this study we highlighted the necessity to implement some precaution to improve the quality of collected data for future studies. This study also showed the significance of the ACI index in representing and quantifying the presence of biophonies. It can represent a synthetic and efficient tool to explore the sound activity of the bird community, nevertheless its application in urban environment needs further analysis.
PAPERS AND CONFERENCES PROCEEDINGS

Papers

Canedoli, C., Manenti R., Padoa-Schioppa, E. Urban parks as habitat providers for biodiversity: a multi-scale analysis in the metropolitan area of Milan (Italy). Urban Forestry & Urban Greening Submitted

Canedoli, C., Crocco, F. Comolli, R., Padoa-Schioppa, E. Analysis of landscape fragmentation and urban sprawl in the urban region of Milan. Landscape Research Accepted


Papers I co-authored a part of the PhD project, as results of my Master and Bachelor thesis projects and other collaborations


Conferences

Canedoli C., Comolli R., Abu El Khair, Inostroza L., Padoa-Schioppa E. Spatial analysis of land cover land use and urban soil properties in the metropolitan area of Milan 5th International Ecosummit 'Ecological Sustainability: Engineering Change'. Montpellier, August 2016. oral presentation

Padoa-Schioppa E., Canedoli C., Comolli R. Urban parks in green infrastructures and diffuse corridors: biodiversity and ecosystem services. 1° Congresso Nazionale Congiunto SITE - UZI – SIB. Milano, September 2016. oral presentation

Gandolfi I., Canedoli C., Tagliaferri I., Padoa-Schioppa E., Papacchini M., Bestetti G., Franzetti A. Diversity and hydrocarbon-degrading potential of epiphytic microbial communities hosted by different tree species in an urban park in Milan (Italy). 1° Congresso Nazionale Congiunto SITE - UZI – SIB, September 2016. oral presentation


Canedoli C., R. Comolli and E. Padoa-Schioppa, Ecosystem services provided by urban parks in Milan. IALE Europe 2015 Workshop ‘Landscape ecology of urban forests: enhancing ecosystem services’. Ljubljana, June 2015. oral presentation
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