Funded by the Erasmus+ Programme of the European Union

MEDfOR SUMMER EVENT

Master Programme Erasmus Mundus

MEDITERRANEAN FORESTRY AND NATURAL RESOURCES MANAGEMENT

Book of Abstracts - Master Thesis

2015/2018 – 5th Edition.

Pieve Tesino (Tn) – Italy - July 2018

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Intra-specific differences of non-structural carbon investment and DBH variability of Pinus nigra in common garden experiment

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Abstract

Increased non-structural carbohydrates (NSC) storage and decrease growth is considered one of the important responses of trees under different biotic and abiotic stresses, which may either drive by genetically or environmentally. Higher genetic diversity and phenotypic plasticity of Mediterranean black pine (*Pinus nigra*) may possibly have different responses NSC storage and diameter at breast height (DBH). The purpose of this study is to investigate the nonstructural carbohydrates storage (in small branches), as well as DBH variability among 18 provenances and 3 sub-species of *Pinus nigra* in test sites uniform environmental condition. Colorimetric method was used to measure starch and soluble sugar concentration (percentage of dry weight basis) from powdered branch sapwood samples which was collected subsequently process in mid July, 2017. Climatic data of the origin of provenance was collected from KNMI Climate Explorer. In general, percentage of soluble sugar was less than starch and they are highly correlated independent of provenances and sub-species category. Only percentage of starch is significantly different among 18 provenances, however, percentage of total NSC, soluble sugar and starch were significantly different among 3 subspecies level. DBH also vary significantly in both provenance and sub-species level, whereas growth rate (2016-17) was not varying significantly. Climatic variables (mainly T_{max}, T_{min}, $T_{max-min}$ and P_{mean}) on the geographic origin of provenances have influence in NSC accumulation in branch sapwood. Provenances those originating in low T_{max} and $T_{max-min}$ have significantly lower NSC investment in comparison with provenances originating at similar or high T_{max} and $T_{max-min}$, however, P_{mean} have significantly negative relation only with soluble sugar investment, whereas, DBH nearly significant positive correlation only with T_{max}, furthermore, provenances of lower latitudinal origin have significantly higher percentage of soluble sugar and DBH than provenances higher latitudinal origin. Overall study reveals that differences in NSC accumulation (in braches) and DBH of provenances in test plantation site partly regulated by heredity and growth phonology on the basis of their climatic and geographic origin, as well as, their phenotypic response to environmental stress in test site

Key word: Branches, non-structural carbohydrates, Mediterranean, provenances, soluble sugar, starch, climate change.

Factors affecting being a NWFP picker in Spain: a household analysis

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Thesis context

The thesis is embedded into the StarTree project, which is concerned with "Multipurpose trees and non-wood forest products: a challenge and opportunity". The project is a pan-European project to support the sustainable exploitation of forest resources for rural development, which ran from November 2012 - October 2016 and is now completed. One of the work packages addressed the gathering and consumption of NWFP. Mushrooms, asparagus, foliage or berries are rather well-spread but yet, scarce literature reports actual figures on these forest uses. Through a European-wide online survey, data was collected for Spanish households. Such information is crucial for understanding the sustainability levels of these forest uses and provide insights to forest managers and policy-makers. Within the thesis shall be analysed, which consumption habits and which factors of the social and geographical environment affect a household to pick wild forest products.

Suitability of vegetation indices derived from Sentinel-2 imagery to model severity of forest damage caused by snow storms. Case study of the Aleppo pine-dominated forests in the Valencian Community.

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Natural forest disturbances caused by wind or snow storms are reduced to infrequent extreme events in the context of Mediterranean forests. Howere, when such disturbance events occur they can seriously compromise productive, social and protective functions of forest ecosystem. One of such extreme events occured in January 2017 on territory of the Valencian Community (Spain). It resulted in a massive conversion of living forest biomass to dead fuel, an increase in fuel bed depth, and decrease in dead fuel moisture. Such outcomes complicate further fire management activities, which are crucial for the Mediterranean region. Taking into account the severity of damage and its large spatial extent, assessment of post-disturbance damage cannot be done manually.
Goal of our study was to compare the performance of different vegetation indices, which are based on data from Sentinel-2 imagery, to quantify negative effects of intensive wind and snow storms on Aleppo pine forests within the territory of Valencian Community. Particular attention was given to building predictive model for quantifying damage level by using a combination of vegetation indices. Latter will find its use for planning post-disturbance restoration activities of the affected forest areas and infrastructures, panning of fire prevention and extinction actions, prioritization of private forest patches (compartments) for subsidizing forest management activities, updating maps of fuel models, etc.

Relationship between leaf phenology and anatomical traits of xylem in Beech

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European beech (*Fagus sylvatica* L.) is a wide spread, well-known and economically most important temperate tree species due to its availability and the diverse usage of the wood in the Central Europe. During the last decades, the total area of beech forests decreased due to human activity and/ or climatic impacts. Despite its ecological tolerance, therefore, the competitive capacity (advantage) of beech may change due to the increased frequency and severity of hot, dry summer and late frosts, not only at the edge of its distribution range but also in other areas that have been considered optimal for its growth. Knowledge of intra-annual growth patterns of beech may therefore help to predict its future distribution in the context of anticipated climate change scenarios. From previous studies, it reveals that several classical dendroclimatological approach applied to define the associations between climate and wood-anatomical parameters of beech which, limited at xylem anatomical level e.g. monthly climatic data and anatomical properties of the whole ring. In this case, partitioning the whole ring into sectors could be more efficient to understand the sensitivity of beech and their resistance against environmental changes in the recent past. Further, this approach can also contribute to understand the association between leaf phenology and xylem anatomical features of beech.

Consequently, we selected two different phenological types (early and late growing) of beech from the northern Apennine close to the Emilia and Tuscany border. The major objective of our study is to analyze how growth (tree-ring width) and wood-anatomical traits (lumen area, mean hydraulic diameter, minimum cell size and maximum cell size) of selected types of beech respond to climatic variables (minimum and maximum temperatures, precipitation) calculated for different time intervals using dendro-anatomical features. In addition, we are interested to explore any relationship exist or not between leaf phenology and xylem anatomical traits of the selected types of beech.

The use of barcoding sequences for the construction of phylogenetic relationships in the Euphorbiaceae

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Abstract
The use DNA Barcodes has been an effective means for the identification and establishing the phylogenetic relationship of the various plant species. Two DNA barcodes (RbcL and matK) of Euphobiaceae family have been generated from the two study sites i.e. Bukit Duabelas National Park and Harapan Rainforest of Sumatra, Indonesia. Dried leaves specimens of Euphorbiaceae plants were collected from the thirty-two plots distributed in these two sites. The two barcodes are being evaluated for species identification and establish phylogenetic relation. With this study of species identification will help to put conservation efforts in threatened area of Sumatra's forest and contribute to the development of useful barcode information.

Keywords: Barcode, Euphobiaceae, Identification, Sumatra

Feasibility of NIR Spectroscopy in Classifying Pine Nut Species Beech

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Abstract
Edible pine nuts are important non-wood forest products (NWFP) with increasing demand and price. The objective of this study was to assess the potential of NIR spectroscopy and image analysis for the classification of pine nut species. To conduct the study, seeds of pine nuts from Italy, Spain, Russia and China were acquired. Moisture content analysis, NIR spectroscopy and image analysis were carried on the acquired pine nut seeds. For determination the moisture content, samples (20 seeds from each species) were dried at 105 ^{o}C for 48 hours till constant weight was achieved. A Luminar 5030 Acousto-Optic Tunable Filter-Near Infrared (AOTF-NIR) Miniature 'Hand-held' Analyser (Brimrose Corp., Baltimore, USA) was used to perform the spectroscopy measurements while a CM2320nf (Hewlett-Packard, Palo Alto, USA) flat scanner was used to perform the digital image measurement. PLS-DA and iPLSDA models were used to classify the pine nuts and their performance were evaluated in terms of accuracy, specificity and sensitive in calibration, cross-validation and prediction. Moisture content results revealed that P. pinea has a significantly ($P<0.05$) higher (5.36%) moisture content as compared to P. sibirica (3.22%). Using the full spectra, visual inspection was not able to distinguish clear differences to classify the different pine nut species. But with some spectral pre-treatment (SNV, 1st order derivative, Savitzky – Golay algorithm of second degree polynomial, over window 9 and AS) the model #120 of the PLS-DA was found to be the best model in terms of its performance (>95%) in all the calibration, cross-validation and prediction. Using the model # 120 in iPLS-DA, the absorption band1640 – 1658 nm, 1720-1738 nm and 1880-1998 nm were found to be essential for the classification of pine nuts. With the imaging analysis, there was a significant variation in luminance (L*), hue angle (h), chroma (C*), yellowness (b*) and redness (a*), perimeter, major axis, eccentricity but not for minor axis. The PLS-DA and iPLS-DA classification models also performed well with classification rates of >95% except the sensitivity was 93.71% in prediction in the iPLS-DA model. The results found demonstrate the NIR spectroscopy and chemometrics and imaging technique are capable of classifying pine nut species.

Key words: Absorption bands, Chemometrics, CIELab, iPLS-DA NIRS, Pine nut, Pinus pinea L, Pinus sibirica Du Tour, PLS-DA

Next generation sequencing (NGS) and quantitative PCR (qPCR) as tools to detect Fusarium circinatum in different pine species.

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Abstract:
Fusarium circinatum, the causal agent of pine pitch canker (PPC) disease, has been already reported worldwide. In Europe, it is considered a quarantine pest (A2) and in Spain it has been reported causing economic damages in Pinus radiata and other pine species. In order to detect F. circinatum present in samples of different origins, different techniques may be applied such as next generation sequencing (NGS) and quantitative PCR (qPCR). In this aspect, the aim of this study was to evaluate qPCR as a technique to detect F. circinatum in stem's samples of five pine species (P. radiata, P. pinaster, P. nigra, P. sylvestris and P. uncinata) and to compare the results with data obtained in a ITS-based massive sequencing. The NGS analysis was unable to detect F. circinatum in the samples analysed although other Fusarium spp. were detected. Quantitative PCR seemed to be affected by inhibitors present in the samples such as polysaccharides and phenolic compounds which in the case of some samples, was overcome by a nested PCR (conventional PCR followed by a qPCR). With the exception of P. pinaster, F. circinatum was detected in all the other pine species of which samples were recovered. In P. radiata, the fungus was detected not only at the inoculation point (15 cm) but also at 65 and 165 cm height, which brings interesting insights about the movement of the pathogen within the plant's tissues.

Keywords: real-time PCR, massive sequencing, Fusarium circinatum.

Understanding human utilization patterns, impacts on endangered tree species and conservation options in the walnut fruit and nut forests of south-western Kyrgyzstan

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Abstract

The fruit and nut forests of Central Asia harbour a great genetic diversity in wild varieties of many important food trees in the world. However, there has been an alarming decline in the forest area in recent decades, due to unsustainable fuelwood harvesting, over-grazing and habitat loss. As a result, several tree species in the region are threatened with extinction, such as *Pyrus korshinskyi* and *Malus niedzwetzkyana*. The rural communities in the region are largely dependent on fuelwood for their livelihood. Hence, there is an imperative to understand the impact of human use on these threatened tree species and to implement adaptive management to ensure their conservation.

The general objectives of the study are:

1. To understand patterns of fuelwood consumption by rural communities in the walnut forests of Kyrgyzstan and its relationship to demographic and socioeconomic status.
2. To identify whether synergistic relationships exist between fuelwood harvesting, threatened tree species composition and land use changes.
3. To identify measures and management options for the conservation of threatened tree species while integrating sustainable rural development.

A set of five villages representative of the region in terms of use and proximity to natural forest habitats and socioeconomic conditions were selected for the study.

To understand patterns of fuelwood consumption, data was collected through questionnaires and semi-structured interviews on socioeconomic status and fuelwood use of rural households. Some of the parameters assessed are:

- Number of people in the household, average monthly per capita income & livelihood occupation of the members
- Types of fuel used for cooking and heating (wood, fossil fuels, electricity)
- Average amount of fuelwood consumed in a day
- Origin of the fuelwood (forest, plantation, fallen timber)
- Preference of tree species harvested for fuelwood
- Distance of village to wood fuel source
- Time required for firewood collection
- If there is any change in the distance travelled and duration of firewood collection compared to the past.
- Livestock use
- Changes in forest structure

- Threats to the above mentioned threatened tree species

Further, semi- structured interviews will be conducted through focal groups, with key members of the village community to assess:

- Strategies to save energy consumption
- Natural regeneration of forest tree species
- Threats to the fruit and nut forests and solutions
- Climate change impacts on the forests
- Forest management
- Conflicts regarding access and use of firewood and other forest products
- Alternative income generation options

Biophysical and geographical data was collected through field surveys from the sample sites such as; Occurrence and abundance of the above mentioned threatened tree species; Areas showing good natural regeneration of the trees; Diameter distribution of the trees; signs of firewood collection from live trees and the presence of grazing. Comparisons were made between populations that have been fenced and open trees to understand the potential beneficial impacts of fencing.

Data for biomass can be used to understand the impacts of fuelwood harvesting in terms of impacted hectares. The total consumed fuelwood biomass can be estimated from the above mentioned parameters. Quantitative data will be analysed through descriptive statistics for relationships between threatened species abundance, fuelwood consumption and socioeconomic variables. Preferences of tree species harvested and the amount of fuelwood can be used to identify synergisms with species loss and habitat disturbance.

Conservation measures and case studies will be studied to identify suitable mechanisms that can be used to protect the unique and threatened wild fruit tree species in the area. These include sustainable pasture management; protection of tree species from livestock grazing by fencing; energy efficient methods such as improved cooking stoves, thermal insulation of houses and use of solar driers (increasing the value of fruits beyond the usual season); ex- situ conservation through nurseries and by identifying mother trees for seed collection; alternative income generation options.

This research was carried out with support from Fauna & Flora International under the auspices of the Global Trees Campaign.

Estimation of carbon fluxes from eddy covariance data and satellite-derived vegetation indices in a karst grassland (Podgorski Kras, Slovenia)

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Abstract
The Eddy covariance method is a widespread method used for measuring carbon fluxes between the atmosphere and the ecosystem. It provides a high temporal resolution of measurements, but it is restricted to an area around the tower called footprint, and other methods are usually used in combination with eddy covariance data in order to estimate carbon fluxes for larger areas. Spectral vegetation indices derived from increasingly available satellite data can be combined with eddy covariance data to estimate carbon fluxes outside of the tower footprint. Following that approach, the present study attempted to model carbon fluxes for a karst grassland in Slovenia. Three types of model were considered: (1) a linear relationship between NEE or GPP and each vegetation index, (2) a linear relationship between GPP and the product of a vegetation index with PAR, and (3) a simplified LUE model assuming a constant LUE. We compared the performance of several vegetation indices from two sources (Landsat and Spot-vegetation) as predictors of NEE and GPP, based on three accuracy metrics (R^2, RMSE and AIC). Two types of aggregation of flux data were explored, midday average fluxes and daily average fluxes. The VPD was used to separate the growing season in two phases, a greening phase and a dry phase, which were considered separately in the modelling process, in addition to the growing season as a whole. The results showed that NDVI was the best predictor of GPP and NEE during the greening phase, whereas water related vegetation indices, namely LSWI and MNDWI were the best predictors during the dry phase, both for midday and daily aggregates. Model type 1 (linear relationship) was found to be the best in many cases. The best regression equations obtained were used to illustrate the mapping of GPP and NEE for the study area.

Keywords: Eddy covariance, carbon flux, GPP, NEE, Vegetation indices.

Effects of Collembolan (Springtails) on Soil Aggregate formation and Stabilization: A trait-based Approach.

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Abstract
An increasing concern about its physical structure and stability has been evident due to its severe degradation which posing continuous threat to crop yield, environmental quality and sustainability. Soil structure is referred to as the different micro and macro-aggregates which are formed by primary mineral particles being coupled together by the organic matter. Soil aggregation is the key ecological process in soil ecosystems mediated by diverse soil fauna. Having implication on ecological sustainability, soil aggregation has received much attention in recent years, however, the focus was mainly on larger soil animals, while the diverse and highly abundant groups of soil arthropods (e.g., Collembola) have been largely negated. Despite, they are thought to have effects as "driver" of ecosystem process. It has also been evident that, Collembola increases soil aggregation co-occurring with arbuscular mycorrhizal fungi, although they prefer on feeding non-AFM hyphae (saprotrophic fungi). However, being aware of this clear research gaps on how Collembola interacts with saprotrophic fungi and thus influence on soil aggregate formation and stabilization, we aimed to investigate more precisely on how Collembolan trophic interaction with a highly abundant soil borne fungi (*Chaetomium globosum*) and their different functional behaviour influences on soil aggregate formation and stabilization. Collembolan trophic interactions were unrevealed by using a combination of fatty acids (FAs) composition and natural variation in C and N stable isotope ratios. Different functional behaviour of Collembolan (e.g., their ability to reduce fungal growth, fungal spore dispersal capability, ability to transport micrometric particles) were measured through laboratory experimentation. Result indicates that, different Collembola species have shown significant increasing effect on soil aggregation with different trophic and functional behaviours, although significant difference was not found in aggregate stabilization.

Keywords: Soil aggregation, Collembola, Saprotrophic fungi, Phospholipid Fatty Acids, Stable Isotope, Trophic Interaction, Behavioural traits.

Study of the distribution of Terfezia claveryi Chatin soil and roots of different host plants in the Region of Murcia

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Abstract
A distribution map of the *Terfezia claveryi* Chatin potential areas associated with the presence of different Helianthemum species was carried out using a GIS multivariate system (Honrubia et al. 2014). The aim of this project is to scan the roots (mycorrhizas) and the rhizosphere soils (mycelium) of annual and perennial species of Helianthemum in these areas during the T.claveryi fruiting season (February to May), by qPCR technique, in order to evaluate and to discover the T.claveryi distribution in the Region of Murcia, Spain.

Multi-Temporal Analysis of Post-Fire Vegetation Recovery after the 2007 Zaca Fire in California

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Abstract
The Mediterranean region of California is home to a diverse mosaic of chaparral vegetation and conifer and broadleaf woodlands with a unique fire regime. Due to climate change and various anthropogenic factors, these ecosystems are under extreme pressure to regenerate after fire occurrence. Although many species have developed coping strategies such as thick bark or re-sprouting capabilities, much of what depends on completing a life cycle has to do with environmental conditions both pre- and post-fire. Many studies to date have focused on understanding how environmental conditions affect recovery rates by examining spectral vegetation indices through remote sensing and using in situ ground truthing for accuracy assessments. However, these field methods tend to be costly and time-consuming at large scales. This study uses moderate to high resolution satellite imagery to monitor the recovery after the 2007 Zaca Fire in Southern California over a six year time span. Changes in pre-fire and post-fire normalized burn ration (NBR) is determined in order to analyze severity levels and a normalized difference vegetation index (NDVI) will be calculated monthly to examine spatial variability of vegetation regeneration. Further examined in the study area is topography and specie composition in order to determine their influence in recovery dynamics.

Assessing ecosystem functioning on Mediterranean forests in the context of global change: the case study of Mont Ventoux (France)

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Abstract
To assess the global change effects on the functioning of Mediterranean forests ecosystems, we implemented a physiologically multi-layer, process-based model (CASTANEA). The model was firstly evaluated on three Integrated Carbon Observation System (ICOS) sites by comparing measured fluxes by eddy covariance technics and simulations of Gross Primary production, Ecosystem Respiration and Evapotranspiration. A set of simulations have been then conducted on Mont Ventoux in South-eastern France on nine species representative of Mediterranean forest ecosystems dominated by deciduous species (*Fagus sylvatica* L., *Quercus pubescens* Mill.), coniferous species (*Pinus halepensis* Mill., *Pinus nigra* J.F.Arnold., *Pinus sylvestris* L. *Pinus uncinata* Ramond ex DC, *Abies alba* Mill., *Cedrus Atlantica* (Manetti ex Endl.) Carrière, or sclerophyllous evergreen species (*Quercus ilex* L.). The model was first used on past meteorological data using the Safran-gauge-based analysis system for the period 1958-2015. Then, the model has been driven by two regional climate models (RCMs) for the period 2006-2100 following the Representative Concentration Pathway (RCP 4.5 and RCP 8.5) scenarios of the Intergovernmental Panel on Climate Change (IPCC). From 1958 to 2016, the average temperature risen by 1,62 ºC and the rainfall in altitude increased by 48,88 mm. For the future projections under the RCP 4.5 and RCP 8.5 scenarios from CNRM – CM5 the average temperature will rise by 1,28 ºC and 4,52 ºC respectively and the average rainfall will increase 180,48 mm under the RCP 4.5 scenario, but it will drop on average -112,24 mm under the RCP 8.5 scenario. Concerning future predictions under the RCP 4.5 and RCP 8.5 scenarios from HadGEM2 – ES, the temperature on average will increase by 2,85 ºC and 5,79 ºC respectively; likewise, the precipitation on average will drop by -142,84 mm under RCPs 4.5 and by -212,67 mm under RCPs 8.5. The ecosystem always remains carbon sink; however, the sink strength globally decreases for future projections under both RCP scenarios.

Keywords: Forest ecosystem functioning; Global change; Carbon balance; Water balance; Eco-physiological model.

Isolation and characterization of monoterpene synthase genes from*Pinus nigra* subsp *laricio* in Calabria

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Supervisor
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Abstract
The European black pine (*Pinus nigra* J.F.Arnold) is a tertiary relict specie, considered as one of the most economically important native conifers belonging to the Mediterranean region. It is one of the most widespread and polymorphic conifers in Europe with highly fragmented distribution range that extends from North Africa through the northern Mediterranean and eastwards to the Black Sea. Black pine, like every other species in the Mediterranean forest, are been threatened by drastic and rapid climate, among other socio-environmental stress factors. To resist some of these stress factors, such as climate change, disease or insect infestation, plants generally continue to produce and release terpenoids within or outside their system. In black pine, Little is known about the molecular mechanism which they use to do so, first as a result of the absence of genome sequences in conifers and secondly, due to absence of terpene synthase gene sequences specifically in black pine.
In this study, we try to use needles from *P. nigra* subsp *Laricio* to isolate some of the genes responsible for terpene synthase, that is, the enzymes catabolizing the production of diverse terpenoid compounds. As this study is underway, we report the isolation of a full- and partial-length monoterpene synthase (MTPS) genes for Pinus nigra subsp Laricio and the underlying methods by which it was accomplished.
The putative sequences of the genes coding for MTPS in *Pinus* species were identified by a BLAST search in the National Center for Biotechnology Information (NCBI) database. This search allowed us to identify 75 functional MTPS belonging to 26 different *Pinus* species that, on the basis of phylogenetic analysis, can be classified in six distinct groups. The deduced amino acid sequences belonging to the different groups were aligned in order to identify highly conserved regions that were used to design degenerate primers for the isolation by RT-PCR of partial transcripts coding for MTPSs in *P. nigra* subsp. *laricio*. By using this strategy we isolated and sequenced partial transcripts coding for MTPS in Calabrian pine for three of the six identified groups. The three partial transcripts were used as template to isolate full-length MTPS cDNA sequences by 5' and 3' RACE (Rapid Amplification of cDNA Ends) extensions. At the moment we isolated and sequenced a full-length MTPS transcript containing an ORF of 1908 bp coding for a protein of 635 amino acids in length that showed an high level of homology with two functionally characterized MTPS (alpha-pinene synthase) previously isolated in *P. banksiana* and *P. contorta*.
We anticipate that findings from these study provides a solid foundation to further investigate the complex roles of genes responsible for terpene synthesis and their impact on biological processes.

Keywords: Terpene synthase, Primers, Nucleotides, Monoterpenes, Transcript

Forest management promotion for climate change mitigation.

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Abstract

I am working within LIFE CLIMARK project (2017-2021), a Study on Forest Management Promotion for Climate Change Mitigation through Compensation of Climatic Credits.
The project is implementing in the region of Catalonia (Spain).
The main objectives of the CLIMARK project are to contribute to the mitigation of climate change and increase the carbon sink capacity of Mediterranean forests by fostering the mitigating effects of multifunctional forest management through the creation of a climate credit market.
This market as a tool to incentivize multifunctional forest management that focuses on climate change mitigation, which is trying to compensate forest owners for doing treatments in a certain way that increases three ecosystem services (water, biodiversity and carbon stocks).
I am working on biodiversity part, and I want to estimate biodiversity in the forest typologies in the landscape units of this project (6 units) scattered all over Catalonia, to play a role of decision-maker and advice the forest owners on what type of treatments to do in different forest typologies.
In order to characterize the biodiversity in the forest typologies, I did a landscape ecology analysis of the surrounding area to the plots using ARCGIS 10.5 and FRAGSTATS programs.
Moreover, I took some related variables and measurements from Spanish national forest inventory plots (NFI4).
Currently, I am summarizing and linking all this variables from Spanish national forest inventory plots (NFI4) and the results from the landscape ecology analysis together for building a database for the final analysis.
Based on the results, I will estimate some biodiversity indices such as Shannon, Richness, Evenness, then relate the position of the plot to the general structure of the stand.
After that we can know which are the stands that have more biodiversity (can be estimated from the variables of shrubs, trees and dead wood), and this is what I want to know to promote for the forest owners.

Leaf Morphology characteristics of tree species for particulate matter deposition in an industrial city of central Italy

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Abstract

One of the major problems that cities are facing due to the increasing urbanization, is air pollution. Within the context of pollution, we have the particulate matter (PM), which are the solid particles of pollution that can be categorized into different sizes: PM_{10} (coarse), $PM_{2.5}$ (fine) and PM_1 (ultrafine). All are harmful to humans but exceptionally fine and ultrafine that can penetrate deeply into the lungs thus causing serious health problems. Due to this increasing problem, it is urgent to think about how to reduce air pollution and improve people's quality of life. Improving air quality may reduce cases of asthma, rhinitis, heart problems, cancers, and even mental problems.

One of the solutions for this problematic issue lies in the urban green areas. A lot of studies investigated leaves trees capability in PM interception and accumulation, but few researches compared species among them and which characteristics influence differences in PM accumulation. In this study, we compared 12 species commonly used in the urban Mediterranean arborization. The case-study city is an industrial urban area of central Italy, the municipality of Terni, notoriously known as a polluted area, mostly for air fraction. Leaves were gathered in one sampling campaign in 2017, after a dry period. A quali-quantitative analysis was promoted through two different techniques: Sacanning electron microscopy (SEM) and washing and filtering (W/F). The SEM technique coupled with energy-dispersive x-ray (EDX) analysis also allowed to identify PM size and its composition, and to differenciate for each species the main three classes of accumulation: coarse, fine and ultrafine. Secondly peculiar charactersitcs of leaves were observed, recorded and classified: macrostructures – foliage, leaf expansion, texture and leaf type by direct observation and microstructures – roughness, trichomes and stomatal density on leaf surfaces through SEM imaging analysis. Finally, we elaborated a retention index of PM with negative and positive traits to indicate which species, and even more broadly which morphologies are most favorable to enhance PM retention, thus to improve air quality in urban areas.

The species observed as strong PM_{10} accumulators were *Platanus acerifolia*, *Prunus cerasifera*, *Acer saccharinum* that retained relatively 11.96, 11.49 and 9.16µg*cm^{-2}. The same grouping was observed also for $PM_{2.5}$ and PM_1. The study revealed that some morphological micro-macro characteristics such as roughness, trichomes, low grooves dimension, high stomatal density, complex leaf type and non-soft texture acted as positive factors for PM retention. We believe this tool can be useful to help policymakers, urban planners, and landscapers in choosing urban tree species that are more potential to reduce the problem of pollution of their cities.

Key words: air pollution, PM deposition, SEM - EDX, urban trees, leaf morphology

Evaluation of the use of Biochar to remove heavy metals from road runoff.

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Abstract

With the ongoing increasing population of the world, technological advancements in industrialization and urbanization process, release of toxic compounds like heavy metals (Cd, Pb, Zn, Ni, Cr, Cu, As etc.) in the natural resources has become a major concern worldwide. Urban storm water and leachate from the road run off supplies a large amount of polycyclic aromatic hydrocarbons (PAHs), and heavy metals which is received by water bodies. These heavy metals are phytotoxic to the plants and posing a serious threat to natural ecosystem and human health. The soil fauna of urban greens is particularly exposed to these pollutants. Heavy metals contaminated soil may also cause changes in the composition of soil microbial community, adversely affecting soil characteristics. Recently, Biochar has been drawn attention of the researchers for its capacity to absorb metals such as arsenic (As), cadmium (Cd), chromium (Cr), mercury (Hg), and lead (Pb) etc. from soil and water. More accurately, biochar exhibits a countless attraction for heavy metals because of having large surface area, highly-porous structure and various functional groups (e.g., carboxyl, hydroxyl, and phenolic groups).

Considering the world wide position against heavy metals, this piece of research will be conducted to evaluate at lab and mesocosm scale, the use of biochar in green infrastructure designed for urban stormwater treatment: the lab test (column test) is intended to determine the response of the heavy metals removal capacity of biochar to two variables: biochar concentration and time of biochar exposure to the polluted solution whereas mesocosm test to validate in a real scale biofilter as the contribution of the biochar to the stormwater pollutant removal along with green infrastructure.

The experiment includes two phases. In a first phase the capacity of biochar to reduce the bioavailability of heavy metals contained in a solution simulating the composition of road runoff were tested in a column trial. In the second phase the biochar will be tested in mesocosms designed to simulate the green infrastructure for road runoff treatment.

In order to regulate the sorption behaviour and removal capacity of the heavy metals by biochar two types of variables will be considered for column test:, **time of biochar exposure to the polluted solution** and **biochar concentration** in three replications. Columns were prepared mixing the "adsorbent (biochar), soil particles (sand). The biochar of 2000-2800 micron size of beech (*Fagus sylvatica*) was used. The polluted solution contain $(NiCl_2)6$, $CuSO_4$ $5H_2O$, $ZnCl_2$, $(Cd(SO_4))_3$, $K2Cr_2O_7$ and $Pb(NO_3)_2$. For a given biochar concentration the time necessary to reach the equilibrium between biochar and metals was evaluated analyzing the percolate after increasing time intervals (2 h, 6 h, 21 h, 54 h and 165 h). To test the effect of different concentration of biochar on percolate quality, 5 biochar concentrations were used. The toxicity of the percolate obtained from

the column with different biochar concentration was evaluated by phytotoxicity test. The seed germination and root elongation test is one of the simplest methods for assessing phytotoxicity. The species used was for phytotoxicity *Lepidium sativum*.
However, as this experiment is still ongoing, in the column test from the first variable absorption capacity of Biochar reached the equilibrium after 2 hrs. Also the prominent reduction in the concentration of metals solution has been found.

Key words: Urban Storm water, Biochar, Heavy metals, Mesocosm, Column test.