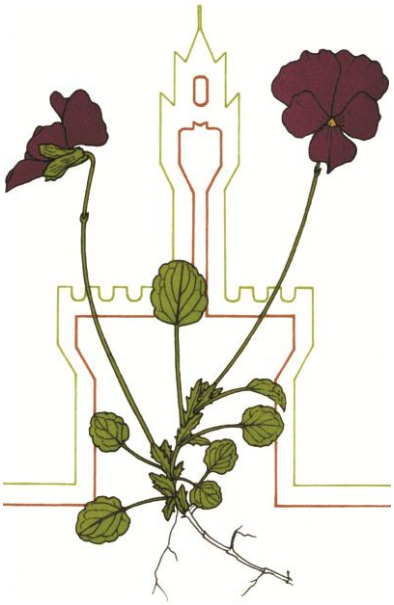

110° Congresso della Società Botanica Italiana onlus

Pavia, 14 - 17 September 2015

ABSTRACTS

KEYNOTE LECTURES, COMMUNICATIONS, POSTERS

II INTERNATIONAL PLANT SCIENCE CONFERENCE (IPSC)

"NOT ONLY FOOD: SUSTAINABLE DEVELOPMENT, AGRO-BIODIVERSITY CONSERVATION & HUMAN WELL BEING"

ISBN 978-88-85915-16-9

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110° Congresso della Società Botanica Italiana

II INTERNATIONAL PLANT SCIENCE CONFERENCE (IPSC)
Pavia, 14 - 17 September 2015

Programme

Monday 14 September 2015

14:00-19:00 Registration (Pavia Botanic Garden, Via S. Epifanio, 14)
14:00-19:00 Scheduled meetings of the Working Groups of the Italian Botanical Society
16:30-19:00 Poster Session (University-main building, Aula del Disegno, Piazza Leonardo da Vinci)
18:00-19:00 Lecture on "Flowers of Lombardy" by the Regional Section of SBI (Pavia Botanic Garden, Via S. Epifanio, 14)

Tuesday 15 September 2014

8:30 Registration (Aula del '400 of the University, Piazza Leonardo da Vinci)
9:30-10:30 Opening Ceremony in the Aula Magna of the University (Piazza Leonardo da Vinci)

General Session – chairperson C. Siniscalco
Aula del '400 of the University, Piazza Leonardo da Vinci

10:40 Introduction to the themes of the Symposia

Keynote lectures – chairperson M. Migliavacca

10:50-12:50

- **M.A. Altieri**, University of Berkeley, California, USA
  Agroecology and the design of biodiverse and climate change resilient small farming systems
- **J. Maschinski**, Kushlan Tropical Science Institute, Fairchild Tropical Botanic Garden, Florida, USA
  Evaluating conservation options for rare and common plant species threatened by climate change
- **J. Doebley**, Genetics Department, University of Wisconsin, USA
  The genetic architecture of maize domestication: low hanging fruit and dark matter

13:00-14:30 Lunch (Cortile delle Magnolie, University area)

Symposium 1 – chairperson G. Lingua
Ecosystem services in agricultural landscapes: the role of plants and fungi
Aula del '400 of the University, Piazza Leonardo da Vinci

14:30-17:35

- **M. Migliavacca**, Max Planck Institute, Jena, Germany
  Climatic and phenological controls on CO_2 and water exchange between biosphere and the atmosphere
- **D. Wipf**, INRA/CNRS/Université de Bourgogne, France
  Study of the biotrophic transportome in the arbuscular mycorrhiza

Communications – chairpersons M. Marignani, A.M. Persiani

- G. Rossi, M.L. Paracchini, C. Bulgheroni, G. Borreani, E. Tabacco, A. Banterle, D. Bertoni, G. Parolo, R. Origgi, **C. De Paola**
  Evaluation of farm ecologic performance in a context of multifunctionality assessment
- **F. Carteni**, S. Falanga Bolognesi, F. Giannino, M. Minacapilli, S. Mazzoleni, G. D'Urso
  Characterization of eco-hydrological processes in different traditional agricultural landscapes of Italy: a process-based modelling approach
- **S. Bagella**, R. Filigheddu
  Biodiversity patterns and related ecosystem services in Mediterranean agro-silvo-pastoral systems
- **S. Tosi**, S. Accossato, M. Rodolfi, M. Faè, E. Lagostina, R. Cella, A.M. Picco
  Agro-residues supplemented with fungi: evidence of its multifunctional properties in agriculture and energy industry
- **F. Guarino**, A. Ciccatelli, C. Nittolo, S. Parrella, S. Castiglione
  Is sunflower a good tool for reclamation of soils contaminated with chromium?
- **D.A. Carbone**, I. Gargano, G. Olivieri, D. Spasiano, N. D'Ambrosio, G. Pinto
  A study of the photosynthetic performances of *Scenedesmus vacuolatus* under different culture conditions
- **A. Ceci**, L. Pierro, C. Riccardi, F. Pinzari, O. Maggi, M. Petrangeli Papini, G.M. Gadd, A.M. Persiani
  β-Hexachlorocyclohexane and fungi: tolerance, biotransformation and phenotypic profiles of the saprotrophic soil fungus *Penicillium griseofulvum*
- **F. Spina**, C. Cordero, T. Schilirò, B. Sgorbini, R. Degan, L. Mantilleri, G. Gilli, C. Bicchi, P. Calza, E. Laurenti, G.C. Varese
  Water contamination by micropollutants and resolutive strategy based on fungal enzymes

18:00-19:30 Poster session

20:00 Welcoming party at Fraschini Theatre (Pavia, City centre)

21:00 Concert offered by ADRAT CNU, University of Pavia. At Fraschini Theatre, Orchestra Sinfonica dell'Emilia Romagna "Arturo Toscanini

Wednesday 16 September 2015

Symposium 2 – chairperson S. Biondi
Plant adaptation and mitigation in the face of climate change
Aula del '400 of the University, Piazza Leonardo da Vinci

09:00-13:00

- **Beat Keller**, University of Zurich, Switzerland
  From the lab to the field: how to improve disease resistance in a changing environment
- **Hans De Boeck**, University of Antwerp, Belgium
  Are more biodiverse systems more stable in the face of climate extremes?
- **Cathie Martin**, John Innes Centre, UK
  The relationship between nutritional value and shelf-life of tomato fruit

Communications – chairpersons G. Falasca, L. Sanità di Toppi

- **L. Selbmann**, C. Cecchini, D. Isola, S. Onofri, C. Coleine, L. Zucconi
  Life at the edge: Antarctic endolithic fungi for monitoring climate change
- **S. Fior**, S. Zoller, A. Widmer
  The genomic architecture of altitudinal adaptation in carnation
- **F. Porro**, M. Gandini, D. Rocchini, M. Tomaselli, G. Rossi
  Assessment of temporal diversity patterns in the vegetation of the northern Apennines

- **V. De Micco**, G. Aronne, A. Balzano, E. Zalloni, P. Cherubini, C. Cufar, G. Battipaglia
  Climate-driven Intra-annual density fluctuations in Mediterranean tree rings: sign of stress and adaptation to environmental changes
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- **V. Carocha**, A.J. Amaral, H. San-Clemente, L. Leal, T. Quilhó, G. Szittya, T. Dalmay, A.T. Freitas, P. Fevereiro, J. Grima-Pettenati, J.A.P. Paiva
  Unraveling post-transcriptional regulation mechanisms of *Eucalyptus* wood formation in response to seasonal and gravitropic stimulus
- **M. Guerrina**, C.N. Macrì, E. Conti, G. Casazza, L. Minuto
  A multidisciplinary approach to study the climate change effect: a case study on *Berardia subacaulis* endemic to SW Alps
- **S. Mazzoleni**, F. Carteni, G. Incerti, M.L. Chiusano, P. Termolino, M. Senatore, F. Giannino, M. Rietkerk, C.E. Vincenot, V. Lanzotti, G. Bonanomi
  Inhibitory effect of extracellular self-DNA: new perspectives in plant-soil negative feedback and species coexistence mechanisms

12:45-13:00 Presentation of the book "*In vitro* embryogenesis in higher plants" (M.A. Germanà, M. Lambardi, Springer Publisher, 2015)

13:00-14:30 Lunch (Cortile delle Magnolie, University area)

Symposium 3 – chairperson A. Chiarucci
Agro-biodiversity and its conservation
Aula del '400 of the University, Piazza Leonardo da Vinci

14:30-17:20

- **L. Maggioni**, Roma, Bioversity International
  Long-term conservation and facilities for an increased utilization of plant genetic resources in Europe: the AEGIS experience
- **J.V. Müller**, Royal Botanic Gardens, Kew, Millennium Seed Bank, UK
  The role of Crop Wild Relatives for future food security: the "Adapting Agriculture to Climate Change" project

Communications – chairpersons A.M. Mercuri, L. Peruzzi

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  A survey of the Italian endemic vascular flora as a source of Crop Wild Relatives
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- **E. Salerni**, C. Perini, E. Bianchetto, S. Mocali, I. De Meo, P. Mori, S. Bruschini, P. Montini, S. Samaden, P. Cantiani
  Selpibiolife: innovative silvicultural treatments to enhance soil biodiversity in artificial black pine stands
- **N.M.G. Ardenghi**, G. Galasso, E. Banfi
  The revenge of American grapes: history and invasiveness of alien *Vitis* (Vitaceae) of crop interest in Italy and Europe
- **M. Sarigu**, G. Bosi, M. Ucchesu, M.C. Loi, G. Venora, O. Grillo, G. Bacchetta
  Image analysis application on waterlogged archaeological *Prunus* remains from a medieval context in Sardinia

- M. Orrù, **O. Grillo**, M. Ucchesu, G. Venora, A. Usai, P.F. Serreli, G. Bacchetta
  Primitive cultivar of *Vitis vinifera* L. during the bronze age in Sardinia
- **C. Giuliani**, G. Flamini, R. Ascrizzi, L. Santagostini, L. Maleci Bini, G. Fico
  The Ghirardi Botanical Garden: a "natural laboratory" for studying the morphological and phytochemical diversity of selected medicinal plants in an integrated ecological framework

17:20-17:35 Presentation of the book "La storia della piante fossili in Italia - Paleobotany in Italy" (Kustatscher E., Roghi G., Bertini A., Miola A. eds, 2014)

17:35 Assembly of the Italian Botanical Society (members only) at Pavia Botanic Garden Via Epifanio 14

21:00 Congress social dinner at Castello Visconteo, City centre (during dinner tasting tipical wines and stories of Pavia by the Association "I Viaggi di Tels")

Thursday 17 September 2015

Symposium 4 – chairperson C. Bicchi
Plants and fungi for human wellbeing
Aula del '400 of the University, Piazza Leonardo da Vinci

9:00-11:35

- **Y.H. Choi**, University of Leiden (NL)
  NMR-based metabolomics as a tool for chemical characterization of medicinal plants
- **L. Conte**, University of Udine
  New areas of olive cultivation and anomalous composition of oils: analytical topics and purity guarantee

Communications – chairpersons V. De Feo, F. Poli

- **E. Ambrosio,** P. Marescotti, M.G. Mariotti, G. Cecchi, M. Brancucci, M. Zotti
  Food traceability: a novel approach for wild edible mushrooms
- **E. Martino,** F. Bracco, M.U. Granata, B. Mannucci, F. Corana, D. Rossi, R. Catoni, L. Gratani
  Fatty acids composition of kernel oil from six different hazel (*Corylus avellana* L.) populations in Italy
- **L. Boggia,** B. Sgorbini, C. Cagliero, G. Pignata, M.L. Colombo, S. Nicola, C. Bicchi, P. Rubiolo
  Total volatile profile of *Artemisia umbelliformis* Lam. as a predictive marker of thujone amount in genepì liqueur
- **M. Daglia,** A. Di Lorenzo, E. Coppo, P. Chavez Carvajal, F. Bracco, G. Zanoni, A. Marchese
  Metabolite profiling and antibacterial activity of *Myrcianthes hallii* (Myrtaceae) used in traditional medicine in Ecuador
- **M. Tacchini,** A. Spagnoletti, A. Grandini, I. Maresca, T. Efferth, A. Guerrini, G. Sacchetti
  Convolvulus pluricaulis, an ayurvedic herbal tool for human and plant well being
- **L. Avesani,** M. Commisso, M. Bianconi, S. Ceoldo, G. Zoccatelli, F. Guzzo
  Potentially neuroactive amines in kiwifruit
- **F. Antognoni,** G. Petroccione, L. Gambuti, S. Centamore, D.L. Taneyo Saa, A. Gianotti, L. Mercolini, R. Mandrioli
  Phenolic acid composition and antioxidant activity in einkorn and white flours, sourdough breads and *in-vitro* digested products

- **R. Venanzoni**, B. Tirillini, E. Bricchi, P. Angelini
  Free fatty acids in *Tuber aestivum - Tuber uncinatum* species complex and its chemotaxonomic significance

11:35-11:50 Presentation of the book "Biologia vegetale, biologia farmaceutica, fitochimica" (A. Bruni ed., Pearson Italia Editore, 2014)

12:00-13:00 Closing Ceremony

13:00-14:30 Lunch (Cortile delle Magnolie, University area)

Special exhibition of "Open Laboratories" from Pavia University researchers, Cortile delle Magnolie

Post Congress, open conference
Aula del '400 (Piazza Leonardo da Vinci)

15:00-18:30 Special Session on "Plant genetic resources and Access Benefit Sharing (ABS) in the framework of the EU Regulation No. 511/2014 and Nagoya International Protocol"/ Risorse fitogenetiche e condivisione dell'accesso ai benefici nell'uso delle piante (ABS), nel contest del Regolamento dell'Unione Europea N° 511/2014 e del Protocollo internazionale di Nagoya". In collaboration with MATTM, MIPAAF, and the Association "Jurists for the Environment Club"

Friday 18 September 2015

Tour to EXPO MILANO 2015 – Milan, Exhibition site
Bus will star at 8.45 from Pavia Railway Station

Special visit to Biodiversity Park area with Prof. Stefano Bocchi and Prof. Ilda Vagge (Milan University) and to some selected pavilions.

Free activity.

Sunday 20 September 2015, Municipality of Pavia and "Orto Botanico Friends Association"

"Piazza Italia Market", "Cluster del gusto": exhibition and market of the best Italian landraces and local products (farmers, research institutions, NGOs, etc.) along Pavia "Decumano" from Piazza della Vittoria (City centre) to Botanic Garden at Piazza S. Epifanio

From **14 to 20 September 2015** three exhibitions will be active in the area of the International Conference:
1) "Secret life of plants", organized by the Botanic Garden Network of Lombardy Via Epifanio 14.
2) "Le carte dei cibi", historical documents regarding food from the University Library of Pavia, organized by Dr. Luisa Erba, Prof. Agnese Visconti and others.
3) "Carte dei Fiori", Biblioteca delle Scienze, Botanic Garden, Via Epifanio 14.

The timetable may be subject to changes

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III. MASCHINSKI J. "Evaluating conservation options for rare and common plant species threatened by climate change"

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IV. MIGLIAVACCA M. "Climatic and phenological controls on CO_2 and water exchange between biosphere and the atmosphere"
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VIII. ZHANG Y., ROBINE M., BUTELLI E., ALSEEKH S., HILL L., FERNIE A., MARTIN C. "The relationship between nutritional value and shelf-life of tomato fruit"

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IX. MAGGIONI L., ENGELS J., LIPMAN E. "Long-term conservation and facilities for an increased utilization of plant genetic resources in Europe: the AEGIS experience"
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Symposium 4. Plants and fungi for human well being

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3. MEZZASALMA V., BRUNI I., AIROLDI C., LOVICU G., FRAU A., RIGOLDI M.P., ARROYO R., CORTIS P., DE MATTIA F., LABRA M. "From grapevines to wines: the "secret" of Sardinian viticulture"

1.3 Multifunctionality

4. BRUSONI M., NEGRI R., GAFFURI F., CAVAGNA B., ROGLEDI M. "Assessment of EU agri-environmental regulations benefits on plant biodiversity in Lombardy"
5. COLEINE C., SELBMANN L., VENTURA S., ONOFRI S., ZUCCONI L. "Fungal biodiversity in arctic biological soil crusts"

6. LANFRANCO L., MIOZZI L., CASARIN V., BRILLI F., ACCOTTO G.P., VAIRA A.M. "Characterization of the tripartite interaction between tomato, arbuscular mycorrhizal fungi and cucumber mosaic virus"
7. LANDINEZ TORRES A.Y, GRANATA M.U., BRUSCHI F., TOSI S. "Assessment of soil quality of differently managed environments by CO_2 and fungal count"
8. MAGGI O., LUNGHINI D., PECORARO L., SABATINI F.M., BLASI C., PERSIANI A.M. "Protecting beech forests and enhancing ecosystem services: the role of fungal biodiversity in Apennine priority habitats"
9. PERSIANI A.M., MAGGI O., PINZARI F. "Are fungal communities decomposing cellulose randomly assembled through dispersal and priority effects?"

1.4 Green economy

10. D'ALEO F., D'ALEO V. "Preliminary study in open raceway for production of *Spirulina* biomass in southern Italy. First experience in Calabria"

1.6 Pollination

11. PIRODDA E., SICLARI P. "Pollination of two tomato cultivars in unheated greenhouses: from pollen transfer to fruit development and bumblebees foraging activity"

1.8 Bioremediation

12. CECCHI G., MARESCOTTI P., DI PIAZZA S., LUCCHETTI G., MARIOTTI M.G., ZOTTI M. "Preliminary geomycological investigations in the Libiola Mine (Liguria, NW Italy)"
13. ISOLA D., ONOFRI S., ZUCCONI L., SELBMANN L. "Isolation of black fungi from contaminated sites: new strains with bioremediation potential"
14. ROCCOTIELLO E., RIGGI A., CECCHI G., BRANQUINHO C., MARIOTTI M.G., ZOTTI M. "Seed germination under nickel stress in a facultative hyperaccumulator"
15. TOMMASI F., D'AQUINO L., DE SIATI F., BRUNO G.L. "Preliminary characterization of fungal endophytes from *Nephrolepis cordifolia* (L.) C.Presl and their role in fern tolerance to inorganic pollutants"

1.9 Cultural rural landscape

16. BERTACCHI A., LOMBARDI T., GIANNINI V., SILVESTRI N., BONARI E. "A case of ecological renaturation in a drained Mediterranean peatland: the case study of the Massaciuccoli Lake basin (Tuscany, It)"
17. CAMPOBENEDETTO C., SABATINI M.E., PAPARELLA S., ARAUJO S., BONADEI M., BRUNI G., CARBONERA D., BALESTRAZZI A. "Bioinformatic and molecular tools to improve bioremediation of polluted air at industrial sites"
18. CIACCIO M., RUSSO R., GIAMMINOLA M.E., DEVIANA M., PALLA F. "Genetics of *Anadenanthera colubrina* var. *cebil* (Fabaceae), a tree from Salta (northwestern Argentina)"
19. LANDINEZ TORRES A.Y., BRACCO F. "Dendroenergetic plants of the Colombian Amazonia: their use and management"

1.10 Green infrastructures

20. D'ARCO M., FERRONI L. "Water use in three *Sedum* species under drought conditions in green roof substrates and their role in green roof performances"
21. FUSARO L., SALVATORI E., MEREU S., MARANDO F., SCASSELLATI E., ABBATE G., MANES F. "Urban and peri-urban forests in the Metropolitan Area of Rome: ecophysiological response of *Quercus ilex* L. in two Green Infratructures, in an Ecosystem Service perspective"
22. SPERANZA M., MAGLIONICO M., FERRONI L., CIPOLLA S.S., D'ARCO M., LAMBERTI A. "Green infrastructures for the urban areas: some results from a one-year monitoring on the experimental green roofs of the University of Bologna"

2. Plants adaptation and mitigation in the face of climate change

23. BARNI E., BAROLIN D., GIUNTOLI F., PETEY M., TEPPA G., BACARO G., SINISCALCO C. "Dynamics of summit floras over the last century in the N-Western Alps"

2.2 Resistance

24. MONDONI A., ABELI T., ORSENIGO S., BERNAREGGI G., BALESTRAZZI A., ROSSI G. "Plastic responses to environmental changes in the longevity of seeds"

2.3 Phenology

25. NOVARA C., FALZOI S., LA MORGIA V., SPANNA F., SINISCALCO C. "Modeling the pollen season start in *Corylus avellana* and *Alnus glutinosa*"
26. SINISCALCO C., PINTALDI E., CREMONESE E., FILIPPA G., CELI L., FREPPAZ M., GALVAGNO M., BARNI E., D'AMICO M., MORRA DI CELLA U. "Microtopography enhances the coexistence of different plant functional types and consequently extends the growing season in subalpine grasslands"

2.4 Reproduction

27. BURATTI F., RESENTINI F., EKIEGLE E., BENEVENT A., ALABADÍ D., BLÁZQUEZ M.A., COLOMBO L., MASIERO S. "TPC15 regulates germination together with DELLAs"
28. GOMEZ-ARIZA J., GALBIATI F., BRAMBILLA V., GORETTI D., MARTIGNAGO D., FORNARA F. "Transcriptional profiling of the shoot apical meristem of rice during transition to reproductive growth"
29. LITTO M., SCOPECE G., FINESCHI S., SCHIESTL F.P., COZZOLINO S. "Effects of herbivory on pollen flow, reproductive success and trophic interactions in *Brassica rapa*"

2.5 Stress adaptation

30. ALOISI I., SCARNATO L., RUIZ K., BIONDI S., DEL DUCA S. "Effect of salinity on seed protein composition of three landraces of *Chenopodium quinoa*"

31. BRANCO D., ALCÂNTARA A., MARQUES DA SILVA J., BERNARDES DA SILVA A., FEVEREIRO P., ARAÚJO S. "Deciphering the regulatory mechanisms behind the improved response to water deficit and recovery of *Medicago truncatula* expressing the *AtTPS1* gene"
32. CERRI M., FERRANTI F., COPPI A., FERRI V., FOGGI B., LASTRUCCI L., VENANZONI R., GIGANTE D., REALE L. "Study of reed-beds decline in different freshwater ecosystems of central Italy: first insights"
33. DELLA ROVERE F., FATTORINI L., PIACENTINI D., RONZAN M., SANITÀ DI TOPPI L., SOFO A., ALTAMURA M.M., FALASCA G. "Auxin accumulation and transport in *Arabidopsis thaliana* (L.) Heynh adventitious roots are modified by cadmium and arsenic"
34. DI FILIPPO A., BRUNETTI M., MAUGERI M., SCHIRONE B., PIOVESAN G. "Tree longevity in temperate forests: using tree-rings to assess the factors affecting the maximum lifespan of broadleaf deciduous trees in different environments"
35. FATTORINI L., PIACENTINI D., BURAN I., ZANELLA L., DELLA ROVERE F., RONZAN M., SANITÀ DI TOPPI L., ALTAMURA M.M., FALASCA G. "Cadmium and arsenic affect adventitious root formation and the definition of the quiescent centre in *Arabidopsis thaliana* (L.) Heynh plantlets"
36. GAMMELLA M., SELVI F., FIOR S., COZZOLINO S. "Local adaptation and gene flow in the serpentine *Dianthus sylvestris*"
37. GIOVANARDI M., PANTALEONI L., VICELLI B., FERRONI L., PANCALDI S. "Dynamics of photosystem II supercomplex in pea plant leaf tissues upon exposure to different incident lights"
38. PERROTTA L., ALBANI D. "Towards the identification of biomarkers of general stress conditions in horticultural species"
39. RONZAN M., ZANELLA L., FATTORINI L., DELLA ROVERE F., CANTAMESSA S., BARBIERI M., SANITA' DI TOPPI L., BERTA G., URGAST D., FELDMANN J., ALTAMURA M.M., FALASCA G. "*Pteris vittata* L. is able to counteract the toxicity induced by cadmium combined with arsenic"
40. RUIZ K-B., SILVA H., MALDONADO J., TORRIGIANI P., BIONDI S. "Morpho-physiological and molecular traits that contribute to exceptional salinity tolerance in *Chenopodium quinoa* Willd."
41. VELOCCIA A., FATTORINI L., DELLA ROVERE F., FALASCA G., ALTAMURA M.M. "Adventitious rooting: what happens between ethylene and auxins?"

2.6 Translocation activities

42. ABELI T., ORSENIGO S., CAUZZI P., GENTILI R., ROSSI G. "The role of population biology and genetics in plant translocation: reinforcement of *Leucojum aestivum* and de-extinction of *Stratiotes aloides*"
43. CARTA A., D'ANTRACCOLI M., BEDINI G., TAMMARO C., PERUZZI L. "Designing plant translocations: *Hypericum elodes* in Migliarino-San Rossore-Massaciuccoli Regional Park as a case study"
44. RUFFINATTO F., ASCOLI D., BERRETTI R., MENCUCCINI M., MOTTA R., NOLA P., PIUSSI P., VACCHIANO G. "Inter individual variability in the influence of cone production on radial growth in Norway spruce (*Picea abies* (L.) H.Karst.)"

2.8 Biology of invasion

45. GENTILI R., CIAPPETTA S., GILARDELLI F., GHIANI A., COLOMBO F., RODIO V., GUARINO M.F., BONINI M., CITTERIO S. "The invasion of *Ambrosia artemisiifolia* L. in Italy: genetic variability, population structure and colonisation routes"
46. PODDA L., SANTO A., PUDDU S., BIAGINI L., BACCHETTA G. "Germination ecophysiology of three invasive Solanaceae species in Sardinia (Italy)"
47. VALENTINI E., LASTRUCCI L., LAZZARO L., MASSI L., DELL'OLMO L., FOGGI B., NUCCIO C. "Preliminary ecological data from a wetland area characterized by massive invasion of *Myriophyllum aquaticum* (Vell.) Verdc."

2.9 Protected natural areas management

48. BERTACCHI A., LOMBARDI T., TOMEI I., LABATE M., FONTANELLI A., PICCHI L. "Vegetation landscape management in the Natural Reserve of Chiarone, Massaciuccoli Lake basin (Tuscany, It)"
49. FERRI V., GIGANTE D. "Effects of environment and different management on morphological traits of a central-Italian population of *Klasea lycopifolia* (Vill.) À.Löve & D.Löve, a near threatened Annex II plant species"
50. GERACI M., ERDFELD D., BAZAN G., SCHICCHI R. "Unmanned aerial vehicle application for mapping individual plants: the case of the Nebrodi beech forest"
51. MURRU V., MARIGNANI M., ACOSTA A.T.R., COGONI A. "Bryophytes in coastal sand dunes and their relationship with main environmental variables."
52. PRISCO I., CARBONI M., JUCKER T., ACOSTA A.T.R. "Winners and losers on Italian coastal dunes in the face of recent global changes: the role of the protected areas network"

3. Agro-biodiversity and its conservation

53. VARESE G.C., PRIGIONE V., PERUGINI J., REALE L. "The role of MIRRI research infrastructure in the conservation and distribution of microbial diversity"

3.1 CWR (Crop Wild Relatives)

54. ANDREELLA M., BIANCO P., CIANCALEONI S., GIACANELLI V., GIARRATANO M.C., LUGERI N., NEGRI V., MORRONI E., PIOTTO B., TARTAGLINI N. "A first approach to the knowledge and conservation of wild crop relatives in Italy"
55. ARDENGHI N.M.G., FOGGI B., MAGGIONI L., ORSENIGO S., CAUZZI P., ROSSI G. "Crop wild relatives of *Festuca* s.l. (Poaceae) in Italy: a national checklist"

3.2 Landraces

56. CANTALUPPI E., LANDONI M., CASSANI E., GIUPPONI L., GIORGI A., PILU R. "Study of an ancient maize from Valcamonica (northern Italy) rich in carotenoids and phlobaphenes"
57. GRILLO O., LO BIANCO M., CREMONINI R., VENORA G. "Seed phenotypic identification of Italian bean landraces (*Phaseolus vulgaris* L.) by biomertric and texture descriptors"
58. LEONI B., DI GIOVINE F., RENNA M., SANTAMARIA P. "The 'Lucera artichoke': a neglected landrace from Capitanata (southern Italy)"

59. ORSENIGO S., ABELI T., VOLPATO M., GENTILI R., GALLA G., CITTERIO S., SGORBATI S., ROSSI G., BARCACCIA G. "Genetic identity of common buckwheat (*Fagopyrum esculentum* Moench) landraces cultivated in Valtellina (Central Alps, Italy)"
60. RENNA M., SIGNORE A., GONNELLA M., SERIO F., SANTAMARIA P. "The Polignano carrot: a multidisciplinary approach for preserving an interesting landrace at risk of genetic erosion"
61. TAZZARI E.R., ABELI T., MONDONI A., CAUZZI P., BODINO S., ARDENGHI N.M.G., VAGGE I., GUZZON F., ORSENIGO S., ROSSI G. "Conservation and characterization of landraces in northern Italy"

3.3 Sustainable crop management

62. ANGELINI P., PROPERZI A., TIRILLINI B., VENANZONI R. "The potential of truffle allelopathy on management of plant-pathogenic fungi in crop fields"
63. BARCELLA M., QUATRALE V., TOSI S., PICCO A.M., SARTORI F. "Bioactive agro-matrix. Effects of its use on vegetative growth, photosynthetic activity and crop production"
64. DONATI L., VALLETTA A., PALOCCI C., CHRONOPOULOU L., BRAMOSANTI M., BALDAN B., PASQUA G. "*Uptake* and internalization of poly(lactic-co-glycolic) acid nanoparticles in *Vitis vinifera* and phytopathogenic fungi (*Botrytis cinerea*; *Aspergillus* sp. pl.)"
65. FEVEREIRO P., ASSUNÇÃO M., SEVERINO R., SANTOS C., BRAZÃO J., SANTOS M., EIRAS-DIAS J. "The (in)compatible story of *Vitis* grafting: the transcription factors route"
66. GRATANI L., GRANATA M.U., SARTORI F., BRACCO F., PUGLIELLI G., CATONI R. "Canopy carbon assimilation rate and related nut yield in *Corylus avellana* orchards under different irrigation systems"
67. PAPARELLA S., GERNA D., LIMONTA M., VACCINO P., ARAUJO S., CAUZZI P., ABELI T., ROSSI G., BENTIVOGLIO A., DONDI D., BUTTAFAVA A., KOCKELKOREN M., VIOLA P., CARBONERA D., BALESTRAZZI A. "Molecular tools to improve seed priming: the 'primtech' approach"
68. RESTUCCIA A., PESCE R., PANDINO G., SCAVO A., MAUROMICALE G. "Allelopathic effects of *Cynara cardunculus* var. *sylvestris* leaf extracts on weed seed germination"
69. SABATINI M.E., PAGANO A., ARAUJO S., VANDENBUSSCHE M., BALESTRAZZI A., CARBONERA D. "Novel molecular hallmarks of seed imbibition"
70. VAGGE I., ORSENIGO S., BALLERINI C., ABELI T., ROSSI G. "Plant Red Lists in Italy: addressing the threats to endemic plants from agriculture"
71. YASEEN FAREED K., PAPARELLA S., TOMA R.S., BALESTRAZZI A., CARBONERA D. "Protoplast large-scale isolation from potato: agronomic and biotechnological applications"

3.6 In/ex situ plant conservation and potential use

72. FUSANI P., AIELLO N., SCARTEZZINI F. "Morphological and agronomic traits of a naturalized population of parsley [*Petroselinum crispum* (Mill.) Nyman] compared with three commercial cultivars."
73. GIOVINO A., SCIORTINO B., SAIA S. "Morphological variability among natural populations of *Chamaerops humilis* L. grown *ex-situ*"
74. IOTTI M., BENUCCI G.M.N., LEONARDI P., AMBROSIO E., PIAZZA G., SEDDAIU S., LEONARDI M., BROTZU R., FRANCESCHINI A., DONNINI D., ZAMBONELLI A., ZOTTI M., TORTA L., SECCHI C., PACIONI G., LANCELLOTTI E. "Ectomycorrhizal fungal diversity in a forest plantation of exotic conifers in Sardinia (Italy)"
75. WIJAYASINGHE M., MONDONI A., ROSSI G., BALESTRAZZI A. "Does seed priming increase the seed longevity of wild alpine plants?"

3.7 Archeobotany

76. BAZAN G., CANFORA L., LO PAPA G., DAZZI C., SCHICCHI R., PINZARI F. "Archeobotanical study of traditional agroecosystems based on SEM-EDX analysis of buried phytoliths"
77. LOMBARDO G., BARBAGALLO M.G., ALEO NERO C., SINEO L., PALLA F. "Ancient DNA extraction from medieval grape seeds"
78. SANTO A., GRILLO O., ORRÙ M., UCCHESU M., BACCHETTA G. "Effect of plant age on seed aspect of native Sardinian grape cultivars"

3.9 Botanic Garden roles

79. ALSAKKAF H., CLAUSER M., FERLI S., GRIGIONI A. "The child gardeners: sowing seeds, harvesting knowlege"
80. LABARDI L., CECCHI L., NEPI C., QUILGHINI G., SELVI F. "The Herbarium of Michele Padula: an inventory of the materials for a "Flora of Casentino" now conserved in the Herbarium Centrale Italicum (Firenze)"
81. MARIOTTI M.G., ZAPPA E. "The *Citrus* collection at Giardini Botanici Hanbury: historical investigation and sustainable conservation"
82. ZAPPA E., CAMPODONICO P.G., MARIOTTI M.G. "Plant list of Villa Roquebrune Garden (Roquebrune-Cap Martin, France)"

4. Plants and fungi for human well being

4.1 Food safety

83. BEDINI S., BERTACCHI A., COSCI F., FLAMINI G., LOMBARDI T., ASCRIZZI R., CONTI B. "*Juniperus oxycedrus* subsp. *macrocarp*a (Sibth. & Sm.) Neirl. (Cupressaceae): a promising resource for integrated post-harvest crop insect pests management"
84. DI SALVATORE M., CARRATÙ G., CARAFA A.M. "Cadmium in tomato berries"
85. MERLA C., ANDREOLI G., GUGLIELMINETTI M.L., ROVIDA E., POZZI C., FABBI M. "Ochratoxigenic moulds in the air of a salami ripening room in Pavia Oltrepò"

4.2 Food security

86. PARREIRA J.R., BOURAADA J., SILVESTRE S., BERNARDES DA SILVA A.B., MARQUES DA SILVA J. , ALMEIDA A.M., FEVEREIRO P., ALTELAAR A.F.M., ARAUJO S.S. "Using proteomics to understand seed development in *Phaseolus vulgaris* L.: developing resources to modulate seed quality traits"

4.3 Medicinal plants and fungi

87. ACQUAVIVA R., TOMASELLO B., MALFA G., GENOVESE C., RAGUSA S., TUNDIS R., MENICHINI F., ARONICA T., DI GIACOMO C. "Effects of *Betula aetnensis* Rafin. (Betulaceae) extract on a human colon cancer cell line"
88. BISIO A., PARRICCHI A., DE TOMMASI N. "A new sesterterpene from *Salvia tingitana* Etl. (Lamiaceae)"
89. BISSO S., CORNARA L., VARNIER O.E., BANFI S., CARUSO E., MCDERMOTT J., MARTINI I., CHIODAROLI L., BRUNI I., CAVALLARI S., VIGANÒ P. "Pharmacognostic and antiviral properties of *Combretum micranthum*"
90. BRACA A., KARKER M., DE TOMMASI N., KSOURI R. "Phytochemical study of *Tamarix africana* Poir. (Tamaricaceae)"
91. FLAMINI G., PARRI F., LEONARDI M., AMBRYSZEWSKA K.E., MELAI B., CIONI P.L., PISTELLI L. " Essential oil composition of *Rosmarinus officinalis* L. from Elba Island (Tuscany, Italy)"
92. GIORGI A., BASSOLI A., BORGONOVO G., PANSERI S., MANZO A., DE PETROCELLIS L., SCHIANO MORIELLO A. "In vitro activity of *Waldheimia glabra* (Decne) Regel extracts on the somatosensory and pain receptor TRPA1"
93. GIROMETTA C., SAVINO E., GUGLIELMINETTI M., LAGOSTINA E., RODOLFI M., ROSSI P., BERNICCHIA A., PERINI C., SALERNI E., PICCO A.M. "The collection of medicinal macrofungi of Pavia University"
94. GIULIANI C., TANI C., DI FALCO P., MALECI BINI L. "Micromorphology and anatomy of bitter melon (*Momordica charantia* L., Cucurbitaceae) fruits and seeds"
95. LEONARDI P., IOTTI M., PULIGA F., PIATTONI F., SALTARELLI R., ZAMBONELLI A. "Ultra-low preservation of the medicinal mushroom *Ganoderma lucidum*"
96. MACCIONI A., MARENGO A., FALCONIERI D., PIRAS A., FARRIS E., MARRAS G., MAXIA A., SANNA C. "Preliminary study on the essential oils of the genus *Teucrium* from Sardinia (Italy)"
97. MANDOLFO A., ANTOGNONI F., POLI F. "Ethnobotanical survey within SCI IT4050001 area (Parco dei Gessi Bolognesi, Calanchi dell'Abbadessa), near Bologna, Italy"
98. MANDRONE M., COQUEIRO A., ANTOGNONI F., POLI F., CHOI Y.-H. "Metabolomics approach for screening collagenase inhibitors from medicinal plants"
99. MARRELLI M., MENICHINI F., NICOLETTI M., TONIOLO C., CONFORTI F. "*Nasturtium officinale* R.Br. subsp. *officinale*: new source of natural inhibitors of pancreatic lipase"
100. MARRELLI M., STATTI G.A., MENICHINI F., CONFORTI F. "Phenolic content and *in vitro* anti-inflammatory activity of *Echinophora tenuifolia* L."
101. MENGHINI L., LEPORINI L., RECINELLA L., ORLANDO G., FERRANTE C., SHOHREH R., CHIAVAROLI A., VACCA M., BRUNETTI L. "Chamomile extracts as potential treatment for inflammatory bowel diseases"
102. MICELI N., FILOCAMO A., RAGUSA S., PATERNITI MASTRAZZO G., DE ROSE R.F., MAGGISANO V., MELCHINI A., TAVIANO M.F. "Biological properties of the polar extracts from leaves and flowers of *Isatis tinctoria* L. (Brassicaceae) growing in Sicily"
103. RAGUSA S., RUSSO R., ADORNETTO A., CHIAPPINI C., VARANO G.P., BERLIOCCHI L., BAGETTA G., CORASANITI M.T. "*Citrus bergamia* Risso & Poiteau (Rutaceae) essential oil exploited in studies for human health applications"
104. SANNA C., BALLERO M., TRAMONTANO E., MAXIA A., MACCIONI A., MARENGO A., TAGLIALATELA-SCAFATI O., CORONA A., ESPOSITO F. "*Onopordum illyricum* L.: new anti-Hiv-1 agents from an edible Mediterranean plant"
105. SANTAGOSTINI L., FLAMINI G., ASCRIZZI R., BOTTONI M., GIULIANI C., FICO G. "Discovering hop in Italy"
106. TUNDIS R., PERUZZI L., BONESI M., RAGUSA S., MENICHINI F., LOIZZO M.R. "*Cryptotaenia thomasii* (Ten.) DC. (Apiaceae) n-hexane extract as a source of potential anticholinesterase inhibitors"
107. VITALINI S., MADEO M., IRITI M., AVATO P., COCUZZA C.E., ARGENTIERI M.P. "Phenolic profile and biological activity of *Achillea moschata* Wulfen from Italian and Swiss Alps"

4.4 Functional food

108. BRUNI I., MAGONI C., PALMIOLI A., REGONESI M.E., LABRA M. "Vegetable waste for medical and cosmetics device: the coffee plant example"
109. DE ANGELIS G., VALLETTA A., PASQUALETTI V., MULINACCI N., DE GARA L., INNOCENTI M., PASQUA G. "Plant antioxidants in the protection against radiation damage"
110. LOIZZO M.R., BONESI M., MALFA G., ACQUAVIVA R., RAGUSA S., MENICHINI F., TUNDIS R. "Radical scavenging and hypoglycaemic potential effects of edible flowers extract from *Borago officinalis* L. (Boraginaceae)"
111. SCARNATO L., ALOISI I., MONTANARI C., LANCIOTTI R., DEL DUCA S. "Gluten-free flour doughs: texture improvement by protein cross-links occurring via microbial transglutaminase from *Streptoverticillium mobaraense*"

4.5 Local and traditional food and their properties

112. GUERRINI A., CIRACÌ L., MARESCA I., MAIETTI A., TEDESCHI P., POLI F., LORENZI B., CONFORTI F., SACCHETTI G., BELLAU ABDI M.L.,.GRANDINI A. "*In vitro* study on new drugs and food supplements from plants used in Sahrawi (Western Sahara) culinary and medicinal tradition"
113. REALE L., FICHERA C., FERRI V., CERRI M., FERRANTI F. "Morpho-histological characterization and nutritional properties of the prickly pear (*Opuntia ficus-indica* (L.) Mill)."
114. SCHICCHI R., DI NOTO G., GERACI A. "Native taxa of Sicilian flora utilized as vegetables"

5. Free Topics

115. ASTEGIANO D., GIROMETTA C., GONTHIER P., GIORDANO L., SILLO F., SAVINO E., PICCO A.M. "*Perenniporia fraxinea* population: a case of study in an urban park at Pavia (Italy)"
116. ASTUTI G., ADAMÉC L., PERUZZI L. "Are shoot features of actual diagnostic value in European bladderworts (*Utricularia* L., Lentibulariaceae)?"
117. BACARO G., ROCCHINI D., DIEKMANN M., GASPARINI P., GIORIA M., MACCHERINI S., MARCANTONIO M., AMICI V., LANDI S., TORRI D., CASTELLO M., ALTOBELLI A., CHIARUCCI A. "Shape matters in sampling plant diversity: evidence from the field"
118. BARBERIS G., SIRTORI M., GAMBA P.E., MARIOTTI M.G. "Burned areas detection and fire weather. A case study"
119. BONARI G., ANGIOLINI C. "Vascular plants in a Mediterranean pine coastal forests: which driver prevails?"
120. BONINI I., VELLINI S. "National catalog form BNB: examples of application in the Herbarium Universitatis Senensis (Siena)"

121. CALEVO J., GIOVANNINI A., DE BENEDETTI L., BRAGLIA L., CORNARA L., PECCENINI S. "Morphological characterization, molecular analysis and chromosome counting for the identification of *Serapias* hybrids (Orchidaceae)"
122. CARÉ R., FEVEREIRO P. "Explaining plant biotechnology to people"
123. CECCARELLI M., SARRI V., FERRI V., LUCENTINI L., MANELI F., REALE L., GIGANTE D. "Macromorphological traits and molecular analysis in central-Italian populations of *Quercus* Gr. *pubescens*"
124. CORTIS P., NIMIS P.L., COGONI A., AMBUS A., PUTZOLU M.S., MARTELLOS S. "Digital key for the identification of Italian orchids"
125. D'ANTRACCOLI M., ROMA-MARZIO F., CANALE A., PERUZZI L. "First data on the pollination ecology of *Campanula medium* L. (Campanulaceae), a species of ornamental and conservation interest"
126. DEL MONDO A., PINTO G., CINIGLIA C., PORCELLINI A. "Determining the composition of biofilms living on stone monuments"
127. DI GRISTINA E., DOMINA G., RAIMONDO F.M., SCAFIDI F. "Taxonomic remarks on *Isatis tinctoria* (Brassicaceae) from Pollino National Park (Basilicata, Italy)"
128. DI PIAZZA S., ZOTTI M., PAVARINO M., PERRANDO M., VIGNOLA S., ZAPPATORE S. "A powerful tool for a safe collection of mushrooms"
129. DOMINA G., CAMPISI P., SCAFIDI F., JURY S.L. "The discovery of plant biodiversity by children through the animated movies: Alice in Wonderland"
130. DOMINA G., GALLEA R., RAIMONDO F.M. "Implementation of an online information system for the "Flora critica d'Italia"
131. FOSSATI F., BERTOLI L. "The fountain of St John the Evangelist's abbey in Parma: diagnostic analysis for restoration purposes"
132. GIOVANNINI A., CALEVO J., NARDOTTO C. "*In vitro* germination of tropical epiphyte orchids *Cattleya bowringiana* Veitch and *Epidendrum nocturnum* Jacq."
133. GIROMETTA C., ZEFFIRO A., DONDI D., ROCCHINI G., MALAGODI M., NIELSEN E., SAVINO E. "Growth of the wood decay fungus *Perenniporia meridionalis* on *Medicago sativa*"
134. IIRITI G., CADDEO A., ORRÙ G. "Environmental education activities and guided tours at the Botanical Garden of Cagliari"
135. LATINI M., IBERITE M., ROSARI I., ABBATE G. "Analysis of woody flora diversity in two sectors of central Italy characterized by different environmental features"
136. LOMBARDI T., PALESTINI V., BERTACCHI A. "Floristic and vegetational characterization of degraded areas in the province of Pisa the case of a dismissed quarry and the neighboring territory (Municipality of Vecchiano)"
137. MANDAGLIO M., PISTARINO A., MUCCIARELLI M. "Seed dormancy and germination in *Campanula martinii* F. Fen., A. Pistarino, Peruzzi & Cellin. and *C. bertolae* Colla (Campanulaceae)"
138. MANFREDI P., SALVI R., CASSINARI C., ANDREIS C., MAROCCO A., TREVISAN M. "Preliminary comparison of the development of spontaneous vegetation between degraded soils and reconstituted ones (Piacenza, Italy)"
139. MANIGA A., ROBERT L., PERROTTA L., ALBANI D. "Visualization of E2F-dependent transcriptional activation in *Arabidopsis thaliana* plants"
140. MANNINO A.M., VAGLICA V., ODDO E. "Interspecific variation in total phenolic content in temperate brown algae"
141. MARCUCCI R., LICANDRO G. "The Bottari-Chiereghin (XVIII-XIX century) herbarium of the Natural History Museum of Venice, Santa Croce, 1730, 30135 Venice (Italy)"
142. MONTANARI G., VARALDO L., MARCHETTI C., GUIDO M.A., MAGNANI L.G. "Flowery vaults: studies on the flora of genoese frescoes of the 17th century"
143. MURRU V., SANTO A., PIAZZA C., HUGOT L., BACCHETTA G. "Seed germination requirements and effects of sodium chloride and potassium nitrate in three Tyrrhenian coastal species of the *Silene mollissima* aggregate (Caryophyllaceae)"
144. NEGRI S., ZENONI S., PEZZOTTI M., TORNIELLI G.B., GUZZO F. "A metabolomic approach to evaluate the effects of environment on the Garganega grape metabolome during ripening"
145. NOLA P., CASTAGNERI D., CARRER M., ASSINI S., BRACCO F. "A comparison between pedunculate oak and black locust vessels in tree rings: it is all a matter of size"
146. PERUZZI L., BAGELLA S., FILIGHEDDU R., PIERINI B., SINI M., ROMA-MARZIO F., CAPARELLI K.F., BONARI G., GESTRI G., DOLCI D., CARIA M.C., MARROSU M., D'ANTRACCOLI M., BEDINI G. "Wikiplantbase: a collaborative platform for floristic data. First steps towards Italian Regional Floras online"
147. PEZZOLESI L., VANUCCI S., GUERRINI F., GUIDI F., PISTOCCHI R. "Influence of abiotic and biotic factors on the production of toxins in the dinoflagellate *Ostreopsis* CF. *ovata*"
148. PICHIERRI S., PEZZOLESI L., ACCORONI S., GUERRINI F., PISTOCCHI R., TOTTI C. "Growth inhibition of *Ostreopsis* CF. *ovata* culture by toxic algal-derived polyunsaturated aldehydes"
149. POLI MARCHESE E., TURRISI R.E. "New floristic data of Mount Etna"
150. POPONESSI S., ALEFFI M., MANELI F., VENANZONI R., GIGANTE D. "Seasonal variability of bryophytic vs. vascular species in the vegetation of submediterranean temporary pools in central Italy"
151. RINALDI R., SANTANGELO A., STRUMIA S., CRISTAUDO A., FINESCHI S., CAFASSO D. "Genetic diversity and connectivity in relict populations of *Platanus orientalis* at the edge of its distribution"
152. ROMA-MARZIO F., BEDINI G., PERUZZI L. "Updating the woody flora of Tuscany: the first comprehensive survey, two centuries after Savi's work"
153. RONCHINI M., BRUNO L., ZILIO M., CHIAPPETTA A., FIOR S., BINELLI G., BITONTI M.B., GEROLA P. "Sub- or neo-functionalization following gene duplication: inference from gene evolution analysis"
154. RUGGIERO F., ORLANDINI S., NATALI F., CECCHI L., BALDACCI S., MAIO S., SARNO G., CERRAI S., SILVI P., BERGER U., PRENTOVIC M., ANNESI MAESANO I., MOUSTAFA A., THIBAUDON M., MONNIER S., OLIVER G., BEDINI G. "AIS LIFE – Aerobiological Information System and allergic respiratory disease management - LIFE13ENV/IT/001107"
155. SPADARO V., MAZZOLA P., RAIMONDO F.M. "Diversity of *Smyrnium perfoliatum* (Apiaceae) in Sicily"
156. TOMEI P.E., TRIMARCHI S., CAMANGI F., BEDINI G., VANGELISTI R., VIEGI L. "A map of ethnobotanical knowledge in Tuscany"
157. TRAVAGLINI A., FANELLI G., FRATARCANGELI C., LEFOSSE C., BRIGHETTI M.A. "Citizen science at Rome: an example in phenologial activity"
158. TUDELA M., WIJAYASINGHE M., ROSSI G., MONDONI A. "Patterns of seed germination within alpine grasslands of different provenance and habitats"

159. VASHEKA O., CRESCENTE M.F., PUGLIELLI G., VARONE L., CATONI R., GRATANI L. “Morphological and anatomical differences in two fern species growing under common light environments”
160. ZOTTI M., DI PIAZZA S., CECCHI G., BELLINI E., MARIOTTI M.G., VENTURA F. “Human mycodiversity in various stages from death”

KEYNOTE LECTURES

AGROECOLOGY AND THE DESIGN OF BIODIVERSE AND CLIMATE CHANGE RESILIENT SMALL FARMING SYSTEMS

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Adaptation is considered a key factor that will shape the future severity of climate change impacts on food production. Fortunately in the developing world, many traditional farming systems still persist and constitute repositories of a wealth of agroecological principles and measures that if effectively disseminated, can help thousands of small farmers become more resilient to climatic extremes. Understanding the agroecological features that underlie the resilience of traditional agroecosystems is an urgent matter, as they can serve as the foundation for the design of adapted agricultural systems. Many of the agroecological strategies used by traditional farmers that reduce vulnerabilities to climate variability include: crop diversification, maintaining local genetic diversity, animal integration, soil organic management, water conservation and harvesting, etc. Observations of agricultural performance after extreme climatic events (hurricanes and droughts) in the last two decades have revealed that resiliency to climate disasters is closely linked to farms with increased levels of biodiversity in the form of polycultures, agroforestry systems, etc. Field surveys and results reported in the literature suggest that agroecosystems are more resilient when inserted in a complex landscape matrix, featuring adapted local germplasm deployed in diversified cropping systems managed with soils rich in organic matter and water conservation-harvesting techniques. The identification of systems that have withstood climatic events recently or in the past and understanding the agroecological features of such systems that allowed them to resist and/or recover from extreme events is of increased urgency, as the derived resiliency principles and practices that underlie successful farms can be disseminated to thousands of farmers via campesino a campesino networks to scale up agroecological practices that enhance the resiliency of agroecosystems. Even bio-diverse agro-ecosystems may be threatened in the long run by climate change if they are not undergoing a constant adaptation – or even transformation – process. Therefore adapting local agrobiodiversity managed with agroecological practices will be required on a continual basis to confront future climatic conditions.

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THE GENETIC ARCHITECTURE OF MAIZE DOMESTICATION: LOW HANGING FRUIT AND DARK MATTER

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The domestication of maize from a wild Mexican grass called teosinte occurred about 9,000 years ago and resulted in dramatic changes in plant morphology. The genetic changes that underlie maize domestication have been investigated using quantitative trait locus (QTL) mapping, QTL cloning, genome-wide selection scans, and genome-wide scans for altered gene expression. QTL analyses suggest that some morphological traits are governed by relatively large numbers of genes (30 or more), but that other traits have relatively simple inheritance involving a single QTL of large effect plus a few smaller effect QTL. We have identified and characterized QTL (genes) with large effects on some domestication traits. First, *teosinte branched* (*tb1*) is largely responsible for the difference between the long branches of teosinte versus the short branches of maize. Second, *teosinte glume architecture* (*tga1*) is largely responsible for the formation of a casing that surrounds teosinte seeds but is lacking in maize. Third, *grassy tillers* (*gt1*) contributes to differences between having many small ears like teosinte or a few large ears like maize. While QTL studies enabled the identification and characterization of a few domestication genes for morphological traits, genomic scans have identified hundreds of genes that show evidence for selection during domestication or differential expression between maize and teosinte, suggesting that selection during domestication may have targeted a broad array of genes controlling unknown traits.

EVALUATING CONSERVATION OPTIONS FOR RARE AND COMMON PLANT SPECIES THREATENED BY CLIMATE CHANGE

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Increasing threats of climate change pose great challenges for conserving plant biodiversity. Particularly critical are those species whose existing habitat may not be able to support them in the next century due to changing climate, changed management regimes, offsite events, or other factors (Fig. 1 (1,2)). Using examples of endangered plants I present a conceptual model contrasting generally accepted vs. controversial conservation options (Fig. 3 (3)). Further I show how the model can be used as a decision-making tool for land managers and policy makers. The best decisions for conserving rare and common species threatened by climate change will depend upon species-specific attributes, the conditions of the existing habitat, the availability and condition of recipient habitat, as well as ethical, practical, political, genetic, and ecological considerations. If considering assisted colonization, it is important for conservationists to consider these important components.

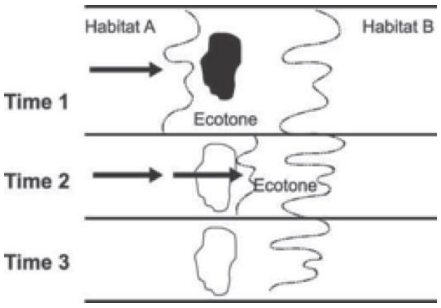

Fig. 1 Changing conditions in Habitat A, due to changes in climate, management or natural succession may eliminate the suitability of habitat for the species growing in the shaded area. (1)

Fig. 2. Key Tree Cactus (Pilosocereus robinii) is an example of an endangered plant whose habitat is gravely threatened by climate change. (2)

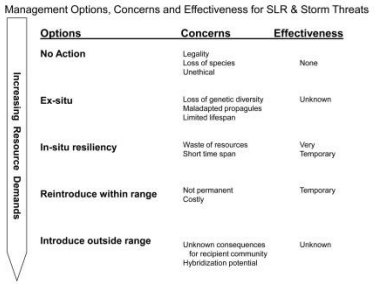

Fig. 3. Management options, concerns and effectiveness for species threatened with sea level rise (SLR) and storm threats (3)

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CLIMATIC AND PHENOLOGICAL CONTROLS ON CO_2 AND WATER EXCHANGE BETWEEN BIOSPHERE AND THE ATMOSPHERE

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The main causes of the year to year variability of CO_2 and water exchange between the biosphere and the atmosphere are still unknown. Climate and phenology are often reported as main driver of the interannual variability of carbon uptake, together with disturbances.
Phenology, the timing of recurring life cycle events, controls numerous land surface feedbacks to the climate system through the regulation of exchanges of carbon, water and energy between the biosphere and atmosphere. Beside foliar phenology, physiological phenology (i.e. seasonal changes in physiological processes) is a relevant aspect of the development of the land surface.
Traditionally, plant phenology is observed in the field through field campaigns. In the last 20 years the development of earth observation systems improved the temporal and spatial description of land surface phenology. Moreover, at site level, the development of methodologies to track the seasonal variation of canopy greenness through the use of commercial digital cameras provided new data to extract phenological events and for the improvement of phenology models and process understanding. Moreover, the development of the carbon and water fluxes observation network allows for monitoring the physiological phenology, opening interesting research perspectives.
However, the major effort was focused on temperate, boreal forest, and grasslands, while water limited and tropical ecosystems are largely ignored, except by few studies.
In this contribution I will focus on a review of the relationship between phenology, year to year variability of biosphere-atmosphere CO_2 exchange, emphasizing then current research questions and gaps.
I will discuss the uncertainty associated with the current formulations of phenology models in land surface scheme and their impacts on future scenario of land-atmosphere interactions.

STUDY OF THE BIOTROPHIC TRANSPORTOME IN THE ARBUSCULAR MYCORRHIZA

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Understanding mechanisms underlying high nutrients use efficiency and carbon allocation in a context of mycorrhizal interactions is critical for sound management of croplands taking care of ecosystem services rendered by mycorrhizal fungi. Transport processes across the polarised membrane interfaces are of major importance in the functioning of the established mycorrhizal association as the symbiotic relation is based on a 'fair-trade' between fungus and host plant. Uptake and exchanges of nutrient and/or metabolites, at biotrophic interfaces are controlled by membrane transporters and their regulation patterns are essential in determining the outcome of plant fungal interactions and in adapting to changes in soil nutrient quantity and/or quality. The talk will present the current state of art with a special focus on S and C transports.

ARE MORE BIODIVERSE SYSTEMS MORE STABLE IN THE FACE OF CLIMATE EXTREMES?

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Research on the relationship between biodiversity and stability of ecosystems has a long history (1, 2), with evidence from theoretical and empirical studies suggesting that high species diversity within ecosystems tends to increase plant community stability (3, 4). Such community-level stability is usually attributed to differences in sensitivity to fluctuations in environmental factors, with more diverse communities having a wider range of sensitivities: even though species responses vary, community functioning is more stable under a range of conditions due to species asynchrony and compensatory responses. The majority of studies on diversity-stability relationships in plant communities focus on year-to-year stability and/or moderate fluctuations in environmental conditions. We here assess whether the biodiversity-stability concept applies also to more extreme and sudden disturbance events such as severe droughts and heat waves.

When scanning literature for studies that simultaneously considered climate extremes and species richness (or biodiversity-stability relationships), we found that conclusions varied. Some studies were supportive of the 'diversity-stability hypothesis', while others came to opposite conclusions or found no or inconclusive effects. This indicates that the biodiversity-stability concept may not be readily applicable to extreme event situations, for which we propose several reasons.

The first relates to an "extreme" having many definitions. As some ecosystems may be responsive over a long stress gradient whereas other ecosystems may be resistant up to a certain threshold (after which resistance quickly erodes), differences in resistance and therefore stability between such systems would very much depend on what is considered "extreme". Secondly, some studies consider only resistance, while others also consider resilience. Because biodiversity may affect both characteristics differently (5), the inclusion of resilience or not could alter conclusions regarding biodiversity-stability. Thirdly, biodiversity loss is often non-random *in natura*, this is usually not the case in manipulative studies on diversity-ecosystem functioning. This may lead to traits modulating responses to extreme events not being equally represented at the same biodiversity level between studies, as a direct consequence of how these studies were conceived. In turn, this can influence the outcome of diversity-stability studies. Finally, diversity-stability relationships may not only be mediated by mechanisms of community assembly (driving patterns of trait abundance at different diversity levels), but also by changes in diversity per se. For example, increased productivity in more diverse communities, an often observed phenomenon, could adversely influence plants during drought, when the higher leaf area in more species rich systems could accelerate drought stress (6).

In conclusion, other mechanisms may be important in extreme event studies than merely those described in traditional diversity-stability literature. Better comparison between studies could be achieved by addressing (i) specifics in the experimental design and extreme event applied that can influence the relative importance of resilience vs. resistance, (ii) community characteristics that change with diversity and directly affect the response to the disturbance, and (iii) the root-cause for why diversity differs between communities featured in the study and its effects on community composition. Such an approach would allow for better mechanistic understanding of biodiversity-stability effects, thus avoiding making "right predictions for the wrong reasons" (7) and increasing the confidence with which results could be extrapolated.

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2) S.J. McNaughton (1977) Diversity and stability of ecological communities: a comment on the role of empiricism in ecology. Am Naturalist, 111, 515-525.
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FROM THE LAB TO THE FIELD: HOW TO IMPROVE DISEASE RESISTANCE IN A CHANGING ENVIRONMENT

BEAT KELLER[1], SEVERINE HURNI[1], DANIEL STIRNWEIS[1], SALIM BOURRAS[1], KAITLIN MCNALLY[1], JAVIER SANCHEZ-MARTIN[1], SUSANNE BRUNNER[2], SIMON KRATTINGER[1]
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The environment and the specific plant and pathogen genotypes present in a particular agro-ecosystem shape the outcome of plant-pathogen interactions. The introduction of new genotypes from breeding programmes, the evolution of the pathogen as well as a changing environment form the background for the efforts to keep plant pathogens under control to avoid economic damage. Thus, there is a constant need for the development of new resistance sources for resistance breeding. We specifically work on fungal disease resistance genes of wheat, their diversity and evolution, and most importantly on strategies to use existing resources more efficiently, but also to explore new ways of creating new diversity and functional specificities.
In a first set of projects and approaches we are developing and testing strategies for a better understanding and use of naturally occurring resistance genes. The wheat *Lr34* is one of the globally most important durable, quantitative resistance genes against rust diseases in wheat. It encodes an ABC transporter protein, making it a highly unusual resistance factor (1). We are studying its function in heterologous systems such as barley and rice. In addition, the powdery mildew resistance gene *Pm3*, which occurs in 17 functional alleles is used to evaluate the use of major genes in improved ways, e.g by making mixtures of lines with individual *Pm3* alleles, the so called multilines, or by combining different alleles in the same genotype by pyramidization (2,3). These approaches are based on the use of transgenic wheat lines which are used for basic studies on gene function (4) as well as for field testing. Field experiments with genetically modified wheat are conducted on a specific research site established by the Swiss government near Zurich (see Figure 1).
Second, we study if modifications of resistance genes can result in improved function. We found that a broad *Pm3* spectrum range correlates with a fast and intense hypersensitive response (HR) in a Nicotiana transient-expression system and this activity can be attributed to two particular amino acids. The combined substitution of these amino acids in narrow-spectrum PM3 proteins enhances their capacity to induce an HR in *Nicotiana benthamiana*, and we demonstrate that these substitutions also enlarge the resistance spectrum of the *Pm3f* allele in wheat. These results highlight the importance of an optimized 'molecular switch' for the conversion of initial pathogen perception into resistance-protein activation, and we describe a possible approach to extend the effectiveness of resistance genes via minimal targeted modifications.

Fig. 1. Field site for experimental research on genetically modified plants in Switzerland. Transgenic wheat plots are shown. More information is available on www.protectedsite.ch.

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THE RELATIONSHIP BETWEEN NUTRITIONAL VALUE AND SHELF-LIFE OF TOMATO FRUIT

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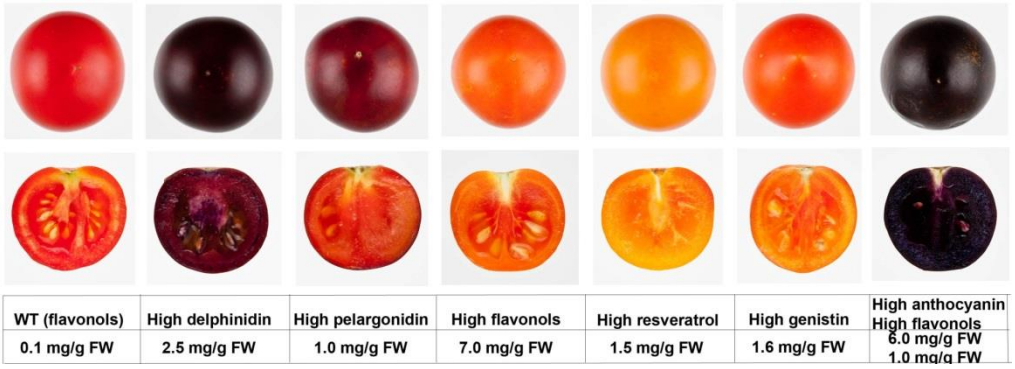
Fruit and vegetables are important sources of many phytonutrients that promote health. Because these compounds are not essential for life, they have been disregarded by many nutritional and biofortification programmes. However, their inclusion in the diet is essential to meeting the objectives of food and nutritional security as defined by the FAO. Phenylpropanoids are derived from phenylalanine and comprise an important class of plant secondary metabolites that include specialized bioactives with medicinal properties and important phytonutrients, that promote human health. By engineering phenylpropanoid metabolism we have been able to enrich tomato fruit with different phenylpropanoid compounds allowing us to assess the relative effects of these compounds on cardiovascular disease in preclinical studies.
The shelf-life of tomato (Solanum lycopersicum) fruit is determined by the processes of over-ripening and susceptibility to pathogens. Post-harvest shelf life is one of the most important traits for commercially grown tomatoes. We compared the shelf life of tomato fruit that accumulate different flavonoids (a subclass of phenylpropanoids) and found that delayed over-ripening is associated with increased total antioxidant capacity caused by the accumulation of flavonoids in the fruit. However, reduced susceptibility to Botrytis cinerea, a major post-harvest fungal pathogen of tomato, is conferred by specific flavonoids only. There is an association between flavonoid structure, selective scavenging ability for different free radicals and reduced susceptibility to B. cinerea. These studies provide mechanistic insight into how flavonoids influence shelf life of tomato, information which could be used to improve the shelf life of tomato, and potentially of other soft fruit. It also links development of nutritionally enriched fruit, to increased post-harvest shelf life, addressing positively the grand challenges of food and nutritional security and sustainability of food production.



Examples of tomatoes enriched with different polyphenolics

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LONG-TERM CONSERVATION AND FACILITIES FOR AN INCREASED UTILIZATION OF PLANT GENETIC RESOURCES IN EUROPE: THE AEGIS EXPERIENCE

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The genetic diversity that is conserved in germplasm collections maintained by genebanks is the base material providing breeders and farmers with the possibility to face current and future objectives and challenges. These include yield increase, resistance to pest and diseases and tolerance to abiotic stresses, improved nutritional value and other qualities demanded by consumers, sustainable agriculture as well as the need for adaptation of cultivated crops to climate change.
The International Treaty on Plant Genetic Resources for Food and Agriculture (ITPGRFA) (1), which entered into force in June 2004, has created a Multilateral System (MLS) for access and benefit sharing of plant genetic resources. It should be noted that the Treaty regulates the exchange of germplasm belonging to a limited list of crops, largely consisting of 35 major food crops and temperate grasses and forages (Annex I of the Treaty).
At present, PGRFA in Europe are conserved in some 600 genebanks or collections, scattered over more than 40 European countries. Considering that genetic diversity for most crops crosses national borders and that conservation and effective use in plant breeding programmes are significantly facilitated when the larger part of the genepool is covered, a closer collaboration between all genebanks has always been a legitimate goal, shared and acknowledged by national programmes across Europe. However, the level of coordination and actual collaboration on conservation activities throughout Europe is still rather low. The European Cooperative Programme for Plant Genetic Resources (ECPGR) (2) aims at facilitating the long-term conservation on a cooperative basis and the increased utilization of PGRFA in Europe. Within this framework, it was decided in 2004 to work towards the establishment of an effective, efficient and rational European conservation system, with an initial focus on existing *ex situ* genebank collections in European countries. The goal is to create A European Genebank Integrated System (AEGIS) (3, 4) with the establishment of a decentralized European Collection operating within the legal framework of the MLS. AEGIS aims at conserving for the long-term the genetically unique and important germplasm accessions in Europe and making them available for breeding and research. Member countries are in the process of selecting individual accessions as European Accessions, for which they are prepared to assume long-term conservation responsibilities according to agreed technical standards, to ensure the safety duplication, and to make the European Accessions available along with the pertinent documentation, in accordance with the principles defined in the Standard Material Transfer Agreement (SMTA) of the International Treaty.
This SMTA will not only be used for the distribution of genetic resources of species included in Annex I; the same terms and conditions will also apply to the transfer of non-Annex I crops, thereby extending the multilateral system in Europe to cover all the relevant crops. In May 2015, 34 countries were members of AEGIS and the European Collection included over 15 000 accessions from five countries. The European Collection is expected to grow, so that breeders and other users will have easy access under equal terms to well characterized and well maintained samples collectively held by any of the Associate Member institutions in Europe, as part of the European Collection.
Information management is an important component of conservation activities. In the case of AEGIS, passport data of the European Accessions are provided online by the European Internet Search Catalogue EURISCO (5), which will soon also include characterization and evaluation data.

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2) http://www.ecpgr.cgiar.org
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THE ROLE OF CROP WILD RELATIVES FOR FUTURE FOOD SECURITY: THE "ADAPTING AGRICULTURE TO CLIMATE CHANGE" PROJECT

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It is estimated that a 70% increase in crop yield would be needed to ensure adequate food security for the rising global population, which is predicted to stabilise at 10 billion by 2050. At current levels of food production, there will simply not be enough food of sufficient nutritional quality available for all people at all times. In addition to that, climate change has severe implications for global food security. Many pests and diseases are predicted to thrive under the projected scenarios and this is likely to be a problem for crop plants in particular. Adapting Agriculture to Climate Change is one of the most urgent challenges of our time. The genetic diversity of our crop plants has been substantially reduced during the process of domestication and breeding. Since the 1900s, some 75 percent of plant genetic diversity has been lost as farmers worldwide have left their multiple local varieties - instead of locally adapted landraces, genetically uniform, high yielding varieties are cultivated. No country is today self-sufficient with respect to the Genetic Resources for Food and Agriculture. Crop wild relatives (CWR) are the richest source of untapped diversity available to improve the world's crops. This was already recognised in the 1920s, when Nikolai Vavilov collected and researched CWR widely and identified what are called the Vavilov Centres, regions such as the Fertile Crescent and the Andes, where the world's major crops were first domesticated and the greatest diversity of their wild relatives is still found. CWR have proven use in providing resistance to pest and diseases, greater yields and are increasingly being used to provide solutions to abiotic stresses. In addition, CWR can be used for testing for example biological control and herbicides. They are morphologically diverse and are a treasure trove of genetic diversity compared to cultivated plants, most of which have been through domestication bottlenecks and subsequent artificial selection reducing their diversity, whereas CWR have been evolving through natural selection to a broad range of factors. They are part of the breeders' toolbox to adapt crops to climate change and help to feed the growing population. Breeding is a lengthy process of controlled hybridisation, evaluation for desired traits and selection of promising plants. It takes about ten years to produce a new variety and even longer for perennial crops.
The "Adapting Agriculture to Climate Change" project (1) is a ten year, global initiative to collect, protect and prepare CWR. Led by the Royal Botanic Gardens, Kew and the Global Crop Diversity Trust, the project focuses on the CWR of 29 major crops, all listed in Annex 1 of the International Treaty on Plant Genetic Resources for Food and Agriculture (ITPGRFA). In the first phase of the project, the project partners analysed 80 different data sources with 4 million individual data records to contribute to a global analysis of CWR. The results of this gap analysis showed that about 50% of all CWR are not adequately preserved in gene banks, and they allowed us to prioritise about 30 countries worldwide with significant CWR diversity and gaps in currently available CWR holdings, with the aim to collect those CWR in their natural habitats. The collection of a total of 6,000 CWR accessions, led by the Royal Botanic Gardens, Kew's Millennium Seed Bank, started in 2013 and will continue until 2017. The collected CWR accessions are stored and processed at the gene banks in country and at the Millennium Seed Bank, where the material will be conserved in long-term storage but also passed on to specialist pre-breeders to be evaluated, charaterised and included in breeding programmes. All material can be requested for agricultural reserach under a SMTA of the ITPGRFA.

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NMR-BASED METABOLOMICS AS A TOOL FOR CHEMICAL CHARACTERIZATION OF MEDICINAL PLANTS

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Natural products, particularly medicinal plants are the most important resource of all kinds of bioactive compounds. Over 50% of new drugs are still being originated from natural products (1). Their huge diversity has served a wide range of source materials to life sciences. However, in other side, the chemical complexity of natural products is also an obstacle to properly use them as resource of bioactive compounds. In a single organism more than 30,000 chemicals are present in which most of bioactive compounds are in low level and vary by many factors e.g. genetic variation and environmental condition (seasonal and geographical).
In the past, to develop a bioactive compound from natural resource, bioactivity-guided fractionation has been often applied to by the isolation of the compounds one by one and testing them in a certain biological activity system. However, this approach has several limitations, long time for the isolation and lost of bioactivity during separation procedures. Therefore, in these days, a comprehensive profiling technique, metabolomics have been extensively employed for the development of bioactive compounds from medicinal plants (2, 3).
Metabolomics has developed rapidly as an important tool for analyzing the total of all metabolites in an organism, thus allowing measuring differences between different accessions, between the organisms subject to different external conditions, etc. Of the analytical platforms used in metabolomics, NMR is one of the most popular methods for metabolomics due to its high robustness of data, broad coverage of metabolites and easiness of quantitation. When NMR-based metabolomics applied to medicinal plants many reports showed that large number of bioactive compounds could be identified from the mixture in combination with chemometric methods, which showed that NMR-based metabolomics could be a promising bioactive metabolites screening methods for natural products (4).
In this presentation, advantages of NMR-based metabolomics as a bioactivity screening system for medicinal plants will be shown as well as practical aspects of the methods with examples of several medicinal plants (5, 6, 7).

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NEW AREAS OF OLIVE CULTIVATION AND ANOMALOUS COMPOSITION OF OILS: ANALYTICAL TOPICS AND PURITY GUARANTEE

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Extra virgin olive oil is one of the most valuable edible oil, being the cornerstone of the Mediterranean diet, depending on this, it underwent to a number of frauds through centuries.
Faked oils had been described since the XXIV century b.C. so that rules had been established to defend its purity and quality through centuries.
Nowadays, rules are established by EU as Regulation that, is well known, are mandatory laws for all the EU countries, nevertheless, the worldwide market requests rules for a widest scenario, so that the International Olive Council was enforced to provide to this (2).
Rules established to defend purity of edible olive oils take into account chemical compounds which presence and relative abundance in the oil are closely bonded to the botanical family of the plant from which oil was extracted.
Within fatty acids, limits had been established for selected acids useful to highlight admixtures with different oils (e.g. linolenic for soybean, lignoceric for groundnut), as well as had been done for sterols (e.g. brassicasterol for rapeseed, Δ-7-stigmastenol for *Compositae* oils –namely sunflower and safflower).
Of course, these standards were established on the basis of couple of decades of chemical analysis on the composition of oil of the area were its production was carried out.
It is important to bear in mind that virgin olive oils (extra virgin and virgin) are obtained directly by mechanical extraction from olive fruits, and that none technological process can be applies neither before extraction to fruits of after extraction to oil but washing and filtering, that's to say that the oil "is born" on the tree and by the tree, not by man (so different from wine).
Such approach worked very well for a number of years, then something changed.
At the very beginning, the olive oil production was limited within the Mediterranean basin (EU and not EU areas), with North and South America and Australia and New Zealand as importers but later, these countries began to produce oils.
Criteria adopted to choose which cultivar of olive introduce in these new areas probably ra with agronomical considerations as resistance to cold or to dryness, or time elapsing before a suitable amount of fruits are produced, resistance to diseases and parasites and so on.
The quality and composition of oil had never be considered, or, at least, never been considered within the new environmental conditions that the olive tree will meet.
Even at the earlier years of oil production, not adequate attention was probably paid to this topic, but when a suitable amount of oil was produced to be offered on the international market, a number of parameters not fitting standard were highlighted and often these parameters were among those suitable to check for faked oils.
The challenge was not to expel from the market the production or a significant part of the production of some countries and in the meantime not to lower the hurdle against faked oil.
In other words, once the production began and heavy financial investments were done, food chemists were asked to "square the circle".
A number of strategies had been proposed and some solution was found, even if new problems arise year after year.
This lecture will give details on these while the ultimate question probably is: "Is possible to establish rigid rules for natural product?"

1) Regulation (EEC) 2568/91, July, 11th, 1991 and further amendments EU Official Journal L/248, September, 5th, 1991
2) International Olive Council Trade standard applying to olive oils and olive-pomace oils, COI/T.15/NC No 3/ Rev. 9, June 2015

COMMUNICATIONS

EVALUATION OF FARM ECOLOGIC PERFORMANCE IN A CONTEXT OF MULTIFUNCTIONALITY ASSESSMENT

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In the framework of the SOSTARE project (Analysis of farm technical efficiency and impacts on environmental and economic sustainability), a diagnostic model has been developed for an integrated assessment of sustainability and efficiency at the farm level (1, 2). The model is based on the use of indicators which are aggregated in a stepwise fashion to provide the user with an immediate valuation of a farm's performance from an agronomic, economic and ecologic point of view. The diagnosis is made on the basis of a synthetic indicator for each of the three above mentioned aspects of sustainability, which are decomposed into twelve sub-dimensions and hence into the respective indicators. The user can then assess the general performance of the farm and explore in detail any perceived weaknesses in farm management and investigate the impact of changes that might improve efficiency. The question of whether on-farm diversification can lead to an increased farm income is addressed, within the context of ecological sustainability. It is thus possible to measure to what extent environmental protection can provide new opportunities for income to farm managers.

The indicators selected to describe the ecological state of the farm have the aim of providing information on the active role that farmers play in maintaining biodiversity and guaranteeing the ecosystem service flow to society (3). Measures of quantity, quality and the spatial organisation of natural and semi-natural vegetation were identified, in order to provide a balanced assessment from both a naturalistic and a functional perspective. Analysis of the impact of farming practices on ecosystems and landscape is carried out through a survey of the natural and semi-natural vegetation elements of the farm holding. These include hedges, ditches, grasslands, wood-lots and forests, whose natural value is expressed based on their size, length, and presence of native species, Red List species according to the Habitat Directive, and species protected at regional level. From a landscape ecology perspective the natural and semi-natural elements are quantified with respect to their capability of blocking alien species and easing gene flow between communities of native species (4). From the economic point of view the focus is on farm income, in order to assess whether a farm can remain on the market, independently of agricultural subsidies. Agronomic farm management is analysed in a similar way, with the interactions between the various elements of agricultural management (i.e. crop choice, rotations selection, management of resources and inputs) being modeled by the diagnostic system. Furthermore, an integrated analysis of agronomic and environmental aspects, that includes nutrient cycle, management of soil fertility, consumption of non-renewable energy, use of water resources, pesticide management, landscape quality, has been carried out in order to achieve a sound valuation of the sustainability and efficiency of agricultural management systems. The selected indicators describing these aspects are easily derived from data requiring a minimum effort in terms of surveying.

The sample is based on a survey of 70 farms. The sampled farms have been selected in order to represent the panorama of farm typologies that are characteristic of the agriculture of Parco del Ticino and Lombardy Region in the widest sense, and include multi-functional and non-multi-functional farms, low-input, organic, conventional management typologies, subdivided into production of rice, cereals, livestock (meat and milk).

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CHARACTERIZATION OF ECO-HYDROLOGICAL PROCESSES IN DIFFERENT TRADITIONAL AGRICULTURAL LANDSCAPES OF ITALY: A PROCESS-BASED MODELLING APPROACH

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One of the aims of the PRIN 2011 project is the development of an integrated methodology based on the use of simulation models of eco-hydrological processes integrated with Earth Observation (EO) data, in order to characterize major agro-forestry systems of traditional agricultural landscapes. The methodology will output eco-hydrological "indicators" for the analysed landscapes, assessing their impact on the eco-hydrological multifunctionality of rural areas.
The simulation model is formulated as a set of ordinary differential equations representing the daily dynamics of three groups of state variables, i) soil water, ii) plant and iii) soil microbial activity (Fig. 1), whose outputs will be used to derive the eco-hydrological indicators. The first submodel is formulated as a multilayer bucket model (based on (1)) simulating the water content in the soil. The second submodel represents the growth dynamics of cultivated plants explicitly taking into account several physiological processes involved in carbon metabolism (photosynthate production, respiration, reserve dynamics, allocation of assimilates and growth) and the transitions between the main phenological phases. The third submodel simulates the decomposition dynamics of soil organic matter (based on (2, 3)), reproducing the amount of CO_2 produced by microbial activity.
At regional scale, the Normalized Difference Vegetation Index (NDVI) has been widely used to estimate changes in plant greenness and spatial and temporal variations in: (a) the onset of photosynthesis, (b) the peak photosynthetic activity, and (c) the senescence, mortality or removal of vegetation. At the local scale, the analysis of EO data at medium to high spatial resolution (6.5 - 30 m) has been used in conjunction with simulation models of water balance in the Soil-Plant-Atmosphere continuum for estimating water requirements and detecting stress conditions (4).
Preliminary simulation results reproducing the dynamics of an olive tree field in Castelvetrano (Sicily, Italy) have been compared with remote sensing data for the period 2003-2014 (NDVI composites from MODIS available from Terra and Aqua platforms) and micro-meteorological data (eddy covariance) for the midseason phenological periods of 2009 and 2010. The results show good potential for the application of the model over long periods and further work is ongoing to calibrate and then validate the model for different case studies over the Italian territory.

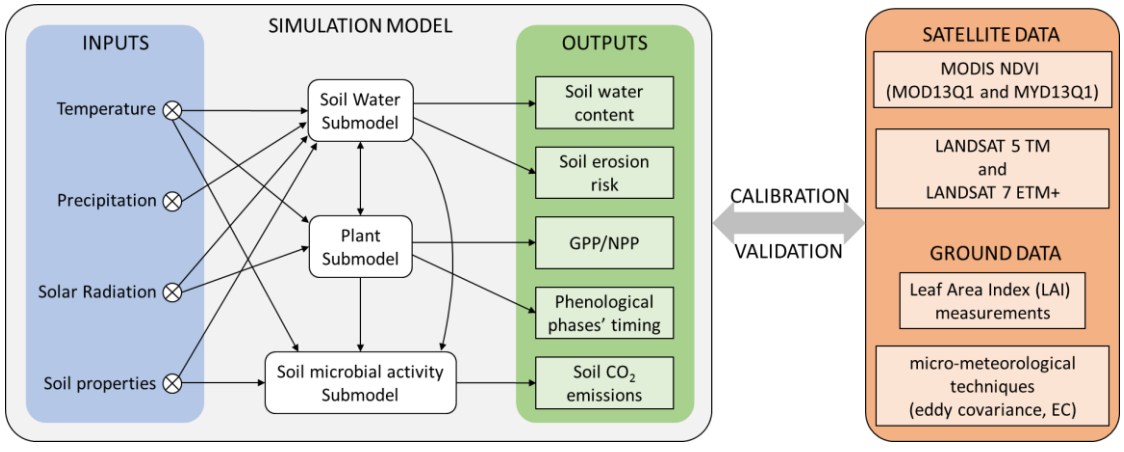

Fig. 1. Schematic representation of the model structure

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BIODIVERSITY PATTERNS AND RELATED ECOSYSTEM SERVICES IN MEDITERRANEAN AGRO-SILVO-PASTORAL SYSTEMS

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Agro-silvo-pastoral systems are recognized as crucial for maintaining the viability of rural areas in Europe and have particular significance for resource and nature conservation. However they are endangered by the dramatic changes towards intensification and abandonment that are occurring in the agricultural and agroforestry systems of many areas in the world, which in the long run can result in a general change of biodiversity and consequently of functionality. In the last decades the European agricultural and environmental policies have started to recognize productive, environmental and societal services of these systems, but the formulation of effective management proposals to balance human activities and biodiversity conservation would greatly benefit of more detailed information on the patterns of diversity and on the related ecosystem services.
Mediterranean agro-silvo-pastoral systems are complex mosaics shaped over the long-term by human activities based on multiple-use-oriented management characterized by different levels of management practice intensity. Each element of the mosaic is related to a cosmos of biota, potentially inter-linked, exhibiting different levels of diversity and providing different types and levels of ecosystem services.
The objective of this paper was to compare the diversity patterns of different biota under a gradient of land use disturbances in a long-term observatory representative of Mediterranean agro-silvo-pastoral systems under uniform environmental conditions. In this long-term observatory a number of multidisciplinary teams are analysing the relationships between land use, biodiversity and ecosystem services in the framework of different research projects.

AGRO-RESIDUES SUPPLEMENTED WITH FUNGI: EVIDENCE OF ITS MULTIFUNCTIONAL PROPERTIES IN AGRICULTURE AND ENERGY INDUSTRY

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Agro-residues such as straw, corn stover, oil cakes, waste wood are rich in lignocellulosic materials and can be converted into biogas and used for heat, steam or electricity generation or left on the soil as biofertilizer. Although these material are particularly interesting being abundantly available and relatively cheap they often remain unused and not fully exploited. Most of the researches are now focused to improve physical, chemical and biothechnological tratments of agro-residues to enhance and valorize their value (1). Integration of agro-residues with selected viable microorganisms can significantly contribute to upgrade these materials in a sustainable way. The Mycology Laboratory of Pavia University has also been involved in isolating and studying fungal strains with activity as biological control agents and powerful organic matter decomposers. Recently, isolates belonging to the Trichoderma genus have been investigated for their antagonistic activity towards important plant fungal pathogens such as Fusarium spp.. The same isolates show powerful capacity to attack recalcitrant compounds such as aliphatic and aromatic hydrocarbons and also degrade lignocellulose-rich biomass.
In vitro experiments were carried out to verify the multifunction activity of these isolates and most of the positive results were obtained with of Trichoderma asperellum. Hydrolytic enzymes, such as xylanase and cellulose, were detected in the secretome of this fungus growing on agro-residues such as rice straw. The obtained results underlined that species of Trichoderma can play important role in enhancing the depolymerisation of lignocellulose into simple sugars that represents the initial step for fermentation processes in the domain of green chemistries.
Thanks to the strong decomposition activity of some Trichoderma strains and to their well-known nature of plant growth promoters, significantly positive results were also obtained in field experiments. Treatment of corn crop with an agro-residue represented by vegetable oil integrated with selected fungal strains of Trichoderma asperellum, T. hartianum, and T. atroviride, had a significant positive effect on vegetation growth and production (2).
The results obtained with agro-residues supplemented with selected viable microorganisms seem to highlight their strong potential as innovative and sustainable tools both in agriculture and in industry.

Acknowledgment:
work supported by “Agromatrici Bioattive” Project, MIUR-Lombardia (ID 30176165).

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IS SUNFLOWER GOOD TOOL FOR RECLAMATION OF SOILS CONTAMINATED WITH CHROMIUM?

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Chromium (Cr) is a metal commonly introduced into the environment through anthropogenic activity and it causes serious contamination of soils, sediments and waters[1]. Principal oxidation states of Cr are: Cr^{3+} and Cr^{6+}, both elemental forms are very stable and even extremely toxic. Chromium is mainly released into the environment by industrial activities of leather tanning. Traditional physico-chemical methods for the remediation of heavy metals (HMs, including Cr) polluted soils result expensive and not environment-friendly. In the last decade phytoremediation has gained interest as a "green" technology that employ plants for reclamation of different polluted matrices. Several plants have proven to be able to clean up contaminated soils. In case soils are polluted by HMs, the primary constraint to their reclamation is due to the limited HM bioavailability, however it can be improved adding chelating agents (*e.g.*, EDTA, EDDS, etc.). Sunflower (*Helianthuus annus* L.) is considered an effective HM phytoremediation plant species[2]. The sunflower Pretor variety was used to evaluate the possibility to reclaim an abandoned agricultural soil collected near the basin of the Solofrana river (Salerno-Italy), where illegal and uncontrolled discharge of waste waters of leather tanning plants caused a large release of Cr into the river waters. The soil collected from a flooding area of Solofrana river was highly contaminated with Cr (P soil - 650 □g g^{-1} and 90 □g g^{-1} of Cr and Cu, respectively) as already described by Adamo *et al*.[3].To improve the efficacy of the phytoremediation process, as stated above, EDTA and/or EDDS were added, (final concentration 5 mmol kg^{-1} soil D.W.), to the polluted and unpolluted soils (NP - 95 □g g^{-1} D.W. and 230 □g g^{-1} D.W of Cr and Cu, respectively). At the end of the experimentation, sunflower plants were harvested, separated into roots, stems, leaves and inflorescences, and metal content (Cr, Fe, Zn, Mn, Cu) was determined using ICP-OES instrument after acidic treatment digestion. Pot filling soils were characterized for total and available metal content. The Bioavailable fraction was low for all HMs in both soils; Cr bioavailable content, in particular, was detected only in P soil and resulted very low (0.01% of the total). Translocation (TF) and Bioaccumulation (BAF) factors were also calculated. Sunflower plants didn't show any sign of stress, or growth inhibition either on P or NP soils, even in the presence of chelants. In all theses, HMs reached the highest concentrations in roots, confirming the data present in literature[4]. On NP soil, Zn and Cu root concentrations were significantly increased after EDDS addition, respect to those of P soils. Whilst for Fe and Mn the same situation was observed after addition of chelant mixture. Chromium content, on the contrary, was high in the sunflower roots after EDTA addition. On P soil, on the other hand, chelants mixture improved the accumulation of the HMs in the roots and the TF values of Cr, Fe and Mn were higher when EDTA was added alone, while it was the contrary in the case of Cu and Zn in NP soil. The BAF values of Cr, on P soil, were higher when chelants were added as mixture. For all remaining HMs, the higher BAF values were detected after the EDTA addition. In all cases, the TF values were very low (<1.0), as well as the BAFs, revealing that, in our experimental conditions, sunflowers were not able to accumulate or translocate HMs and especially in the case of Cr. Although present in P soil at high total concentration, Cr was not up-taken from sunflowers because strongly associated to the soil matrices, even when chelants were added to both soils. Unfortunately, Pretor variety, in our experimental condition, was not able to remove Cr, and the other HMs, when grown on the Solofrana soil, even in the presence of chelants, because of an extremely low bioavailabilty of Cr. Although, sunflower is considered, in literature, an effective phytoremediation plant and an HM hyperaccumulator, however, in our case, it didn't meet the expectations, even using it in combination with efficient chelants. Hence, it is good practice to proceed with pilot experiments before employing this biotechnology on a large scale, thus reducing the risk to incur unpleasant situations, and therefore considering erroneously phytoremediation an ineffective methodology for reclamation of HM contaminated soils.

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A STUDY OF THE PHOTOSYNTHETIC PERFORMANCES OF *SCENEDESMUS VACUOLATUS* UNDER DIFFERENT CULTURE CONDITIONS

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Closed photobioreactors allow microalgal growth at very high rates, leading to massive productions of microalgal biomass under controlled conditions (1). In photobioreactors, light availability is one of the most critical factor determining microalgal productivity. Light should be provided at the appropriate intensity and wavelength, as low light levels can be growth-limiting, while excessive light intensity may lead to photoinhibition of the photosynthetic process, and therefore a decrease of the growth rate (2). Photosynthesis and photoinhibition occur preferentially in cells near the culture surface, while mutual shading of algal cells causes steep gradients of light intensity within the culture. To describe this behavior, Gargano et al. (3) recently measured on cultures of different algal strains three kinetic parameters on which is based the model proposed by Eilers and Peeters: photoadaptation, photoinhibition and flashing light effect (4).
In this study, we have monitored the photosynthetic performances of *Scenedesmus vacuolatus* (strain ACUF 053) under batch, fedbatch, and semicontinous mode of growth. This species has been chosen because it is considered one of the best candidate for biofuel production coupled with phyco-remediation. Cultures were grown in vertical cylindrical bubble column photobioreactor (CBC) bubbled in air or in CO_2 enriched air stream (CO_2 concentration about 2%v/v). Tests were carried out setting the irradiances for 24 h at 240 $\mu E\ m^{-2}\ s^{-1}$ using white light sources (Philips Master, TL-D 90 de luxe; 36 W/940). The aim of this tests was to integrate these new set of data into the model recently proposed (3).
The photosynthetic activity of *S. vacuolatus* cultures was analyzed by measuring gas exchange and photochemical process. The gas exchange was measured with a Hansatech oxygraph at different light intensities. Maximal gross photosynthetic rates (P_{max}) were obtained from the fit of the curves to the equation provided by Henley (5). The photochemical process was studied by pulse amplitude modulated fluorimetry (PAM), using a Hansatech Fluorometer. The analysis is based on the measurement of the fluorescence parameters in response to saturating light in dark- or light-adapted specimens. The algae were exposed at different light intensity and four parameters were considered: Fv/Fm, F0, FPSII and NPQ. Finally the photochemical data were processed with Mathlab according to Gargano et al.(3).
In the batch phase the increase of algal concentration is linked to a progressive decrease of photochemical activity, probably due to nutrient depletion. In the fedbatch phase, medium was added to the cultures twice a week and a stable improvement of the photochemical activity was observed. During the semi-continuous phase, 30% of culture volume was weekly replaced with fresh medium containing 3N (three times concentrated) $NaNO_3$. During the first 24 hours after the dilution the photochemical activity result very high, while progressively decreased in subsequent days.
The analyses of photosynthetic activity show that the rate of photosynthesis is driven by CO_2 availability under batch, fedbatch, and semicontinous mode of growth. Moreover, the gas exchanges indicate that the photosynthetic activity progressively increases from batch to semicontinous cultures. Photochemical data confirm that the best photochemical activity takes place during fedbatch and semi-continuous phases, and that at irradiances lower than 1000 $\mu E\ m^{-2}\ s^{-1}$ the photochemical process is controlled by the photons capture, while at higher irradiance photoinhibition competes with photochemical quenching.

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β-HEXACHLOROCYCLOHEXANE AND FUNGI: TOLERANCE, BIOTRANSFORMATION AND PHENOTYPIC PROFILES OF THE SAPROTROPHIC SOIL FUNGUS *PENICILLIUM GRISEOFULVUM*

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β-hexachlorocyclohexane (HCH) is a persistent organic pollutant (POP) of global concern with potentially toxic effects on humans and ecosystems. Fungal tolerance and biotransformation of toxic substances hold considerable promise for environmental bioremediation as many fungi can tolerate extreme environmental conditions and possess efficient extracellular degradative enzymes with relatively low specificity. In this work, we have investigated the potential of a saprotrophic soil fungus, *Penicillium griseofulvum* Dierckx, isolated from soil with high concentrations of isomers of hexachlorocyclohexane, to biotransform β-HCH, the most recalcitrant isomer to microbial activity. The growth kinetics of the fungus were characterized after growth in stirred liquid Czapek-Dox medium in the presence of 1 mg L^{-1} β-HCH and in stressful nutritional conditions at different sucrose concentrations in the medium (0 and 5 g L^{-1}). Phenotype Microarray™ technique was used to study the effects of β-HCH and its solvent, toluene, on fungal metabolism. Phenotypic profiles for *P. griseofulvum* suggested the selective activation of certain metabolic pathways as a response to oxidative stress due to the presence of the xenobiotic. β-HCH biodegradation was confirmed by gas chromatographic analysis of isomer concentration and by the formation of benzoic acid derivatives as dead-end products. A minimum value of 18.6% for the β-HCH residual concentration was observed. These findings have important environmental implications for possible *in situ* or *ex situ* bioremediation of hexachlorocyclohexane.

WATER CONTAMINATION BY MICROPOLLUTANTS AND RESOLUTIVE STRATEGY BASED ON FUNGAL ENZYMES

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Since global water consumption of fresh water doubles every 20 years, clean and safe water is a primary need and challenge. Aside from traditional pollutants, new emerging xenobiotics (plasticizers, pesticides, herbicides, personal and pharmaceutical care products) called Endocrine Disrupting Chemicals (EDCs) spread and persist in fresh waters. They are able to interact with human estrogenic receptors, and alter the levels of hormones that control growth, reproduction, and development. Interfering with the endocrine system, they cause serious damages to all the exposed organisms, not only to human beings, undermining the integrity of whole ecosystem (Snyder et al. 2003). Coupling human agricultural practices and industrial wastes to the ineffectiveness of conventional treatment methods, these compounds tend to accumulate in aquifers, and eventually use for irrigation, getting multiple ways of re-entry into the life cycle and ultimately perturbing human health and environment.

A possible approach to arrest this risky trend is at least to consistently remove micropollutants from surface and civil waters and avoid its constant accumulation and diffusion. Wastewater treatment plant are deeply deficient, claiming for additional tertiary methods specifically focused and design to be active towards EDCs. A biological approach opens new intriguing scenarios, being fungal enzymes able to degrade a wide spectrum of xenobiotics operating at various pH, temperatures and ionic strength. Indeed the present study investigated the potential of *Trametes pubescens* laccases to be part of an advance bio-oxidation approach applicable in the municipal water treatment plant of the Torino district (Italy) that collects also waters coming from the up-stream reservoir of the Padan plan. It treats around 42,000 m^3/day and serves four towns of the metropolitan area and almost 250000 inhabitants.

The actuality of the problem has been confirmed by finding up to 20 molecules at high concentration (ng/l or even µg/l), including the well-known bisphenol A, ketoprofen, 4-n-nonylphenol, alachlor. A great seasonal variability of the chemical composition in term both of quality and quantity of detected compounds was observed, stating once again how human activities have reflection on water purity. The most abundant chemicals were pesticides, plasticizers and drugs, reaching concentration much higher (µg/l) than those recognized to be effective towards living organisms (ng/l).

Laccases degraded several EDCs, belonging to different chemical classes and uses. The proper enzymatic concentration, able to carry out extensive and fast conversion reactions, was set at 100 U/l: within 24 h, high process yields (50-96% reduction) were obtained for most of the detected compounds. More in detail, the initial pollutants concentration (from 120 to 15,700 ng/l) was considerably lessened by the enzymatic treatment and the amount of five compounds decreased even below 100 ng/l.

Besides, chemical data were coupled with a precise toxicological risk assessment, evaluating the biological interferences ascribable to the samples before and after the enzymatic treatment: a significant abatement of the ecotoxicity and the estrogenic activity was demonstrated by different bioassays (the plant *Lepidium sativum*, the algae *Pseudokirchneriella subcapitata* and the human breast cancer cell line). A clear correlation between the compounds (and each metabolite) presence and toxicity is not possible in such complex sample where synergic effects could occur; however, some experiments will be performed to define the most dangerous micropollutants, in order to define scientific records which could be useful for the definition of the upcoming legislative resolution.

Further studies are in progress to enhance the stability and the efficiency of the laccase-mediated system. Laccase immobilization is a primary challenge because it would allow recycling the enzymatic biocatalyst with limited costs and manageable techniques. Beads of functionalized silica are now under study: optimization of the enzymatic recovery and stability has to be optimized as well as the catalytic efficiency of the immobilized laccase towards micropollutants of interest.

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LIFE AT THE EDGE: ANTARCTIC ENDOLITHIC FUNGI FOR MONITORING CLIMATE CHANGE

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Polar ecosystems are very sensitive to Climate Change; over the past 50 years some Antarctic and sub-Antarctic regions have experienced some of the most rapid increases in mean air temperatures on Earth [1]. The Antarctic endolithic communities, living at the edge of their biological potential, are particularly sensitive to any external variation [2]. Environmental pressure and isolation promote adaptive radiation and speciation and these communities host plenty of peculiar endemic genera and species [3, 4, 5]. The establishment of more permissive conditions and subsequent possible introduction of competitive alloctonous species may cause the extinction of some highly adapted autoctonous components of the communities [6]. For these reasons it is of utmost importance to investigate the amplitude and preserve this threatened and mostly still unknown biodiversity before any climatic variation leads to possible extinction. Moreover, a deeper understanding of responses of these communities in terms of distribution, species richness and biodiversity variation under different environmental stresses may allow identifying changes and give clues to monitoring or predict the effect of any future variation due to Climate Change.
Recently it was proved that typical cryptoendolithic colonization was exclusive of sandstone pushing lithobionts towards harsher conditions at higher altitudes or inner sites of the Antarctic continent [7]. Fungal biodiversity variation in relation to environmental parameters of altitude and sea distance was studied using DGGE approach in a selection of 72 rock samples, representative of a much wider sampling. Results revealed the presence of few dominant species indicating a high degree of organization and specialization but high vulnerability of the community to external changes; this means that a long recovery time might be required after intense perturbing events. Rock porosity seems to influence richness of biodiversity and in a certain extent a proper porosity allows colonization towards high altitude and sea distances; yet, when airspaces reduces, amplitude of biodiversity is mostly dependent to altitude and sea distance. Sandstone, as the most porous rock, allows a more efficient colonization of endoliths and influences the richness of biodiversity.
Rock inhabiting fungi were isolated from the same rock samples; a multilocus phylogeny, along with some recurrent endemic species in these Antarctic niches, revealed the presence of 3 new genera and 7 new species that are being described.

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THE GENOMIC ARCHITECTURE OF ALTITUDINAL ADAPTATION IN CARNATION

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The Alpine region is characterized by a great variety of habitats that differ in both their biotic and abiotic components; one factor that is of primary importance in shaping the differing environments is certainly altitude, as this in turns affects a number of key climatic variables, e.g. temperature, precipitations, solar radiation. Solid scientific evidence for altitudinal adaptation in plants is presented in an extensive body of ecological literature, and insights on the genetic mechanisms underlying the divergence of ecotypes occurring at different elevations are recently emerging in a number of systems. However, evolutionary genomic studies that investigate the architecture of altitudinal adaptation are still missing. In this work, we explore the genomic architecture of altitudinal adaptation in two species of carnation, *Dianthus sylvestris* and *D. carthusianorum* (Caryophyllaceae), from a sampling of populations occurring at contrasting elevations in the Alps. Three pairs of high- and low-elevation populations representative of each species were collected in the region of Valais (Switzerland). Phenotypic measurements performed on representatives of these populations reveal significant differences in a number of key traits that characterize ecotypes from contrasting elevation, and which we hypothesize being indicative of adaptation to different environments. The search for genomic signature of selection was performed using complementary analyses of next-generation sequencing data. A draft sequence of the genomes of both species was produced by assembling Illumina data with a multi-step pipeline implementing assessment of reliability of the scaffolds. For both species, the assemblies captured more than 90% of the exome in ~20,000 scaffolds representing ~60% of the total estimated genome size. Population genomic data were obtained following two parallel strategies: i) whole-genome resequencing of 20 individuals per population using a Pool-seq approach and ii) ddRAD sequencing at the individual level of the same samples used in the Pool-seq approach. This strategy enabled to thoroughly scan the genome to detect regions of selection using the Pool-seq dataset, and validate the allele frequencies obtained in the latter using the information from individual RAD markers. We identified ~20 candidate regions under selection within each of the two species. Annotated genes in these regions appear to be consistent with expectations on altitudinal adaptation, including e.g. flowering time, seed germination, light response, photomorphogenesis, oxidative stress response and pathogen response. Whilst selection appears to target similar functions in both species, the genes involved differ in all cases, with one exception. Within-population statistics show candidate regions to undergo variable selection regimes, with differences between populations from contrasting elevation that appear to be gene-dependent. These results provide insights to interpret the role of antagonistic pleiotropy and conditional neutrality in the process of altitudinal adaptation, and they reveal whether the same genetic architecture underlies the response to the environment in the two species. Overall, we infer that divergent selection drives ecotype formation at contrasting elevation, and loci under selection appear to potentially underlie the establishment of reproductive barriers. These are interpreted as potential key components of the early stages of speciation between altitudinal ecotypes in carnation.

ASSESSMENT OF TEMPORAL DIVERSITY PATTERNS OF VEGETATION IN THE NORTHERN APENNINES

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The ongoing process of global warming represents one of the major threat for biodiversity of the high mountain ecosystems, and its impact on plant can already be tracked in the upward migration of species (1), the spreading of thermophilous and ubiquitous species, and, at the same time, the extinction of rare and specialized one (2). The resulting loss of heterogeneity and variability of the natural alpine environments, thus, could lead to a "biotic homogenization", i.e. a loss of biodiversity in the summits vegetation (3).
The present work aims to investigate and quantify the changes in alpine vegetation, in terms of species richness and species diversity, occurred along a temporal gradient in the alpine environment of the northern Apennines (Italy), in order to detect and assess the eventual impacts of climate change on plant biodiversity of these areas.
In this study, changes in species richness, α-diversity and β-diversity of four summits along an elevation gradient, at different investigation scale levels (overall study region, single summit, and section of summit) have been analysed. Hence the changes in diversity and the degree of biotic homogenization have been evaluated relying on the plant species datasets collected within two separate surveys carried out with a time gap of seven years (2001-2008), following the standardized sampling design described by the guideline established by the "Global Observation Research Initiative in Alpine Environments" (GLORIA), an initiative towards an international research network to assess climate change impacts on mountain environments (4).
The analyses on species richness, performed by means of Mixed ANOVAs and Paired t-tests, and the analysis of α- diversity, performed by means of Rényi entropy profiles and Shannon diversity index, display the same patterns of variations, with a significant increase in species number during the period 2001-2008, especially in the lower summits and at lower summit sections level, which are becoming more similar in terms of species richness and diversity to the highest ones, comporting a flattening of the differences due to the altitudinal gradient. On the contrary β-diversity, expressed by the Sørensen dissimilarity index (sensu Baselga 2010 (5)), remained almost unchanged after seven years, showing only slightly but interesting differences at the summit section scale level.
These results can be read as signals of an ongoing process of biotic homogenization of the plant communities of the study region, induced by climate change and by the related upward migration of species characteristic of lower elevations, as testified by the fact that the greatest variations occurred within the lower summits and the lower sections.

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CLIMATE-DRIVEN INTRA-ANNUAL DENSITY FLUCTUATIONS IN MEDITERRANEAN TREE RINGS: SIGN OF STRESS AND ADAPTATION TO ENVIRONMENTAL CHANGES

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Plant growth is mainly driven by climate and any change in environmental conditions can affect ecosystem productivity. On-going climate changes in Mediterranean ecosystems will likely trigger longer periods of drought and an increase in intensity and frequency of extreme events (1). Such changes may, in turn, have severe impact on woody plant growth performance thus affecting ecosystem structure and carbon balance. To cope with the typical Mediterranean double stress due to dry summers and cold winters, Mediterranean tree and shrub species show specific wood traits allowing either high conductivity or high safety against embolism when environmental conditions are respectively favourable or limiting (2). Under such stressful conditions, specific patterns of cambial activity are responsible for the formation of intra-annual density fluctuations (IADFs) in tree rings, also known as false rings, double rings or missing rings (3).
The analysis of the relations between wood traits and environmental parameters in tree-ring chronologies (*retrospective analysis*) combined with the monitoring of xylogenesis (*current analysis*) is a useful approach to reach a comprehensive understanding of wood response to climate fluctuations, in order to make hypotheses on future climate-driven alterations in wood growth dynamics (4, 5, 6).
Herein, we summarise the results of some investigations based on both types of analyses. More specifically, the *retrospective analysis* was aimed to the characterization of IADFs (e.g. frequency and position of density fluctuations in tree rings) and was conducted on tree-ring chronologies of Mediterranean shrub (*Arbutus unedo* L. and *Erica arborea* L.) and tree species (*Quercus ilex* L. and *Pinus* spp.). We applied a multidisciplinary approach based on the combination of dendrochronological methods (for tree-ring identification through cross-dating), quantitative wood anatomy (to measure efficiency/safety of water conduction) and stable isotope measurements (to evaluate physiological behaviour). Correlations between anatomical, eco-physiological, isotopic and environmental variables (rains and temperature) were calculated.

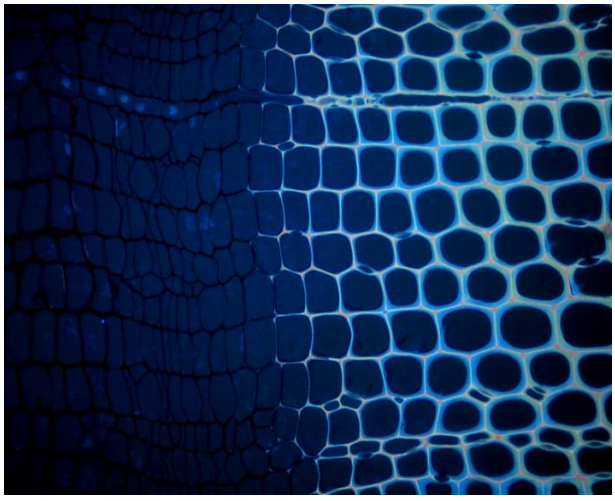

Fig. 1. Microscopy view of a cross section of *Pinus halepensis* wood: from cambium (left) towards differentiating cells (right).

The *current analysis* of xylogenesis was aimed to identify the periods of IADFs formation. It was conducted through the analysis of weekly sampled microcores in *A. unedo* and *Pinus* spp., in order to analyse the phenology of cambial activity to follow the processes of cell division, enlargement and differentiation.
Results showed that the type and frequency of IADFs in tree rings is species- and site-dependent. Moreover, major climatic drivers in IADFs formation can be different in the various type of IADFs and in different species.
The overall investigation allowed to formulate hypotheses on IADFs formation in the analysed species and possible implications on their adaptive capability in the sight of climate changes.

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INVESTIGATION ON THE EFFECTS OF TIO_2 NANOPARTICLES AND BULK COUNTERPART IN *VICIA FABA* L.

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TiO_2 nanoparticles (NPs) are among the top five NPs used in consumer products (toothpastes, sunscreens, cosmetics, food products, medicines and pharmaceuticals, agriculture and environmental cleanup products) for their high stability, anticorrosive properties, redox selectivity, low production costs and wide spectrum of applications (1). NPs have been recently included among the emerging contaminants by USEPA (2) since living organisms may be affected by their exposition to NPs released into aquatic, terrestrial and atmosphere environments. The experimental design of the present work was to study the model system *Vicia faba* L. var. *minor* and the potential effects of the same concentration (50 mg/l) of a commercial source of TiO_2 NPs <100nm (tetragonal crystals, from Sigma-Aldrich) (S), of a NP laboratory sample <10nm (spherical shape, kindly provided by PlasmaTech, Pisa) (P) (3), and of the corresponding bulk material (B) recently classified as possibly carcinogenic to humans by the International Agency for Research on Cancer (4). The above materials were applied to *V. faba* seeds considering different endpoints such as germination/root elongation, root meristem mitotic activity, possible anomalies and/or disturbances during cell cycle and root ultrastructure in function of the shape and size of NPs. In addition oxidative stress and antioxidant response were evaluated by biochemical approach and *in situ* histochemical techniques. Germination percentage did not show significant differences among control and treated materials, but roots from bulk-treated seeds were significantly shorter in comparison to the control and to nanoparticle-treated materials after 72 of imbibition. Concerning cytological analysis, the aberration index only evidenced a significant increase in samples treated with S and B materials.
Bulk TiO_2 induced an oxidative stress in terms of both hydrogen peroxide and TBA-reactive material, higher than in control and in NPs-treated seedlings. Defense system following this treatment seemed to rely mainly on low molecular weight antioxidants, at the expense of the reduced forms of these molecules.
NPs exposure caused a lower oxidative stress with a high reducing power of glutathione in P treated seedlings and by a high POD activity in S material.
Histological evaluation associated to *in situ* detection of oxidative stress was effected by different probes for hydrogen peroxide, reactive nitrogen species, lipid peroxidation and peroxidase activity. In root cross sections differences in staining intensity and localization of the signals were observed mainly in S and B treated roots, to evidence a stressful effect of these materials, while, following the P treatment, the staining pattern was less different from the control.
Under electron microscope, the cytoplasm in control root cells was rich in well structured organelles, particularly rough endoplasmic reticulum and dictyosomes. The cells in root treated with B material showed the most disorganized cytoplasm, with swollen cisternae of smooth endoplasmic reticulum, not well organized mitochondria and organelles often not well recognizable. When exposed to nano-scaled TiO_2, the ultrastructural appearance of cells was less affected after treatment with P than with S NPs.
These results suggest that TiO_2 NPs may exert different actions with different levels of toxicity, depending on their size and their shape and that the bulk counterpart, in our experimental condition, seems to provoke the major adverse effects in *V. faba* root.

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FAD8, *LIP* AND *OSMOTIN* ARE COLD-ACCLIMATION GENES IN *OLEA EUROPAEA* L.

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Olive tree is an evergreen species of economic value lacking winter dormancy and showing low tolerance to frost. This low tolerance limits its cultivation in cold regions, where, by contrast, oil quality is improved by an enrichment in unsaturated fatty acids, i.e., linoleic (C18:2) and linolenic (C18:3) acids, produced by the activity of specific fatty acid desaturases (FADs). Cold-resistant genotypes have been empirically selected from centuries however the genetic network controlling cold-tolerance in olive tree is still unknown. Transient changes in cytosolic calcium are involved in sensing the cold stress and in activating cold acclimation in numerous plants, including olive tree (1-2). A role for the PR-5 protein Osmotin (OeOSM) in olive tree cold acclimation has been suggested, possibly as transfer protein to the cell wall of cutinsomes, containing unsaturated fatty acid-derived compounds (3, 4). The expression of genes coding for specific FADs, i.e. OeFAD2.2 and OeFAD7, necessary for C18:2 and C18:3 production, respectively, has been demonstrated to be positively related to olive tree drupe cold-response. However, the expression of both genes normally occurs during oil biogenesis, and increases under cold-stress independently of the acclimation capabilities of the genotypes, suggesting that these genes are unrelated with cold acclimation (2). By contrast, in *Arabidopsis thaliana* and other plants, *FAD8,* isoform of *FAD7,* is specifically activated by cold (5). Moreover, *FAD8* expression in maize is activated concomitantly with the beta-Zip LIP transcription factor *mlip19* (6), and members of *LIP19*-family are activated by calcium transients (7). The research aim was to identify transcripts of an *OeLIP* gene in leaves and drupes of two genotypes, one incapable and the other capable of cold acclimation, by cold-stresses applied before, during and after the possible natural/artificial acquisition of cold acclimation, investigating, in parallel, the changes in *OeOSM* and *OeFAD8* expression, the immuno-localization of OeOSM, and by detecting the levels of C18:3-compounds deriving by OeFAD8 activity. Preliminarly, leaves and drupes belonging to cv. Moraiolo and cv. Canino were exposed to cold shocks of different duration and intensity at the same developmental stages, and the cold response evaluated in their protoplasts in terms of presence/absence of cytosolic calcium transients, for determining their differences in cold acclimation under all cold conditions. Both genotypes were cold sensitive at the beginning of the oil biogenesis in the drupe (WAF 10), however Canino was able to acquire artificial acclimation at this WAF. Only cv. Moraiolo showed calcium rises, i.e. remained cold-sensitive at the end of oil biogenesis (WAF 19) in leaves and drupes, and continued to be sensitive even in full winter (WAF 26), showing its total inability to cold acclimate, differently from the other genotype.
An *OeLIP* gene was isolated and characterized, and its changes monitored by q-PCR in both leaves and drupes under the same cold-shocks applied for monitoring cytosolic calcium changes. *OeLIP* was activated by cold-induced calcium signalling. Its expression increased with cold, but became stable only in Canino. *OeFAD8* transcription was also induced by calcium signalling, and changed in parallel with that of *OeLIP*. The production of C18:3, and related compounds, by OeFAD8 activity, increased in Canino in concomitance with acclimation. Also *OeOSM* was activated by calcium signalling, and its transcripts were high and stable during acclimation acquisition and maintenance by Canino drupes and leaves. Moreover, both organs showed increased cutinisation of the outer cell walls of epicarp and adaxial epidermis, respectively, with this event strongly increasing in Canino, and positively coupling with an increased immunolocalization of OeOSM in the cuticle. All together, results demonstrate that *OeFAD8, OeLIP* and *OeOSM* jointly control cold-acclimation in *Olea europaea* drupes and leaves**.**

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UNRAVELING POST-TRANSCRIPTIONAL REGULATION MECHANISMS OF *EUCALYPTUS* WOOD FORMATION IN RESPONSE TO SEASONAL AND GRAVITROPIC STIMULUS

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Eucalyptus grandis and *E. globulus* are among the leading sources of wood biomass used worldwide for pulpwood and timber. *Eucalyptus globulus* is a prevalent species in Portugal occupying about one third of the total forest area. Also, Portugal is one of the world's largest producers of high quality pulp and paper, mainly using *E. globulus* wood, with exports accounting for about 5% of GDP. In the context of rapid climate changes and limited land availability, the ability to drive wood quality would represent a huge contribution for the Portuguese economy since the use of more adequate and productive genotypes will optimize the exploitation of available spaces releasing pressure from agricultural areas.

Wood complex anatomical, chemical and physical properties are determined upon composite ontogenetic processes, requiring the spatiotemporal transcriptional and post-transcriptional regulation of a large number of genes. Wood is essentially formed by xylem dead cell walls, the remnants of a highly controlled process of successive cycles of cell formation as a result of the vascular cambium activity and a programed cell death mechanisms. In the long lived trees, the cell wall biosynthesis (xylogenesis) is influenced by variation inherent to seasonal cycles and the exposure to a plethora of external stimulus. The ability to partially reprogram these complex mechanisms provides an important adaptability to some external stimulus. A remarkable example is the Angiosperms' response to gravitropic stimulus, leading to formation of tension wood, an excellent model to study xylogenesis.

In plants, the post-transcriptional regulation by microRNAs (miRNAs) has been recognized to play crucial roles in diverse biological processes including vascular cambium differentiation. MiRNAs are small (20–24 nt) non-coding RNAs that control a vast array of developmental and stress-related biological processes by down-regulating miRNA translation either by mRNA cleavage or by translational repression (1). In contrast to recent progresses made in understanding the transcriptional regulation of wood formation, our knowledge of the role of miRNA-mediated post-transcriptional regulation of these processes is still very limited.

Using two *Eucalyptus* xylogenesis models of study (seasonal variation and tension wood), 748 miRNAs have been identified by cross-validating miRNA prediction from data generated by the deep sequencing (sRNA-Seq) of 15 *E.globulus* smallRNA libraries with in silico miRNA prediction of candidates identified over the *E. grandis* genome sequence by using the CRAVELA framework (www.cravela.org). This is the largest, high-confidence *Eucalyptus* miRNA dataset reported so far – among which 82% have never been reported before. Large scale validation by degradome sequencing analysis of a control (not bent) xylem library allowed us to identify 123 target genes for 98 miRNAs including 71 newly identified ones. The functional classification of the target genes for these newly identified miRNAs identified genes related to the secondary cell wall such as those involved in cellulose and heteroxylan biosynthesis and deposition, as well as genes related to the availability of cinnamate, p-coumaroyl CoA, and S-adenosylmethione for the lignin biosynthetic pathway. This fundamental knowledge of the post-transcriptional regulation of xylogenesis and cell wall biosynthesis will enable to develop new and more efficient strategies to improve wood biomass for various species of trees.

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A MULTIDISCIPLINARY APPROACH TO STUDY THE CLIMATE CHANGE EFFECT: A CASE STUDY ON *BERARDIA SUBACAULIS* ENDEMIC TO SW ALPS

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Future climate change may lead to a substantial loss of biodiversity (1). Significant impact of global warming is already discernible in animal and plant populations that, for instance, have shifted their distributions according to temperature variation (2, 3). The SW Alps have been identified as the region with the highest diversity of plant species, specifically of endemics, in the entire Alpine mountain chain (4). Endemic species are exposed at an increased extinction risk by global warming; in fact, they are likely more dispersal-limited and less genetically variable, thus less able to rapidly adapt to climate change than species with broader distribution (5).
In plant species, global warming can lead to changes in: i) distributional range; ii) reproductive biology and plant-insect interactions; and iii) interactions with other plant species. In order to produce reliable estimates of the risk and to draw up proactive strategies, for any species under consideration it is fundamental to investigate several aspects such as life history, ecology and genetics (6).
The present study is aimed at using *Berardia subacaulis*, an ancient species that has already experienced the past climatic oscillations, as a model to investigate how future global warming might affect the survival of a palaeoendemic species restricted to the SW Alps. In particular, a multidisciplinary approach was carried out, examining the following aspects: phylogeography, species distribution modelling, reproductive biology and ecology.
Using genotyping by sequencing and the projections of the species distribution models in the past, the phylogeographic history of *B. subacaulis* was inferred. The results suggest the *in situ* survival of the species during the Ice Age and the persistence of an old genetic structure throughout several Quaternary climatic cycles. In order to predict how the distributional range of *B. subacaulis* may change in response to future climate change, the broad temporal spectrum of range shifts was investigated. Species distribution models forecast high loss of potential suitable habitat, but the forecasted contraction of the distributional range is similar to the potential contraction that the species seems to have already experienced during the last interglacial. For these reasons, future changes in distributional range as response to an increase of temperature seems to not represent a threat for the survival of *B. subacaulis*.
The study of reproductive biology of the species pointed out that the flowers are protandrous, favouring cross-fertilization, but self-fertilization is allowed. The flowers are pollinated by a wide array of insects, but the visits are scarce. *B. subacaulis* probably takes advantage of its self-pollination and self-compatibility, which assures reproduction also in case of low pollinators service and in harsh high altitude conditions. *B. subacaulis* seems to be less exposed to changes in the interaction plant-insects which are detrimental to plant reproductive success of obligate outcrossers.
The study on the microhabitat preference of *B. subacaulis* reveals that this species is stress-tolerant and avoids competition occupying a very specialized habitat, characterized by high level of physical and hydric stress. Changes in interactions between species due to altitudinal shifts may increase the accessibility of these types of habitats from other stress-tolerant species. This might represent a possible threat for the survival of *B. subacaulis*.
The present study supports the importance of investigating demography, life history, ecology and genetics in conservation biology. Using this multidisciplinary approach it is possible to better understand the possible effects of climate global change on the biodiversity.

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INHIBITORY EFFECT OF EXTRACELLULAR SELF-DNA: NEW PERSPECTVES IN PLANT-SOIL NEGATIVE FEEDBACK AND SPECIES COEXISTENCE MECHANISMS

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Plant-soil feedback (PSF) is considered an important factor shaping the structure and diversity of plant communities (1). In particular, negative PSFs play a fundamental role in soil sickness (2) and evidence of autotoxicity resulting from litter decomposition is wide spread in agriculture (3). Theoretical modelling work showed how autotoxicity could explain gradients of species diversity (4) and spatial organization of vegetation (5), (6). In this context, autotoxicity has been related to the release of inhibitors by litter decomposition and their removal by water.

Mazzoleni et al. (7) tested the hypothesis that extracellular DNA is responsible for litter autotoxicity. Such hypothesis, formulated on the base on theoretical reasoning, was thoroughly investigated by integrating ecological, phytochemical, and biomolecular studies. In particular, metabolomic characterization was performed on a large set of plant litter materials at different decomposition stages by nuclear magnetic resonance (13C NMR). Finally, the effect of purified DNA on a range of organisms from microbes to higher plants were tested (7, 8).

Results showed a general occurrence of species-specific litter toxicity. Data indicate that extracellular self-DNA accumulates in litter during decomposition and that there is a strong relationship between this process and the inhibition of root growth. The reported discovery reveals a new unexpected function of extracellular DNA at ecosystem level. Moreover, tests using purified extracellular self-DNA demonstrated the general occurrence of inhibition in plants and various organisms including bacteria, fungi, algae, protozoa, and insects. Such findings provide strong evidence for the involvement of DNA in autotoxicity. This discovery, while bearing general implications for life sciences and suggesting possible applications in agriculture understand the problems related to soil sickness, also opens a new scenario of ecological research on plant succession and species coexistence mechanisms in relation to litter decomposition cycles.

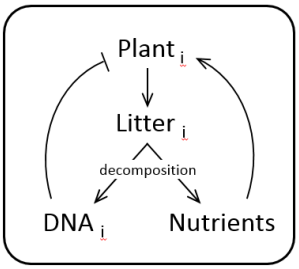

Fig. 1. Schematic representation of the plant-soil feedbacks by litter decomposition.

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A SURVEY OF THE ITALIAN ENDEMIC VASCULAR FLORA AS A SOURCE OF CROP WILD RELATIVES

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Recent studies highlight the prime importance of CWRs in the framework of global food safety and biodiversity conservation. In these contexts, a number of studies are being carried out within EU-funded projects like PGR Secure (http://www.pgrsecure.org/) and Adapting Agriculture to Climate Change (http://www.cwrdiversity.org/).
In Italy, using an inclusive definition of CWR based both on gene pool (1, 2) and taxon group (1) concepts, a recent study recognized 7032 CWR species (3). However, no data is available as regards the ex situ collections of these taxa, as urged in (4).
To this end, we are reporting a survey of the Italian endemic vascular flora as a source of CWRs, based on a recently published and continuously updated list (5), checked against both gene pool and taxon group concepts. Within Italian CWRs, endemic species should be given high priority as regards their study and conservation, as they are not found outside the country.
Of the 1410 endemic species and subspecies, 1227 (i.e. about 87%) are CWRs based on the broad definition proposed by Maxted et al. (1). Of this impressive amount of CWRs, 95 are those related strictly to human and animal food, a further restrictive criterium used in (6).
Approximately 20% of endemic CWRs are conserved in ex situ collections in germplasm banks (7), but this percentage is attained due mainly to few genera with at least one accession for each species (*Allium*, *Brassica*, *Crocus*, *Ribes*), highlighting the need for further efforts to secure this important source of genetic load.
Our analysis show a wide gap in basic biosystematic knowledge such as chromosome number and ploidy level, which is completely unknown for over 40% of Italian endemics (8). This information is crucial since differences in chromosome number and ploidy level may cause difficulties in the practical use of the Gene Pool concept. Furthermore, despite the deeper understanding of dormancy-breaking and germination requirements for some groups of endemic CWRs (e.g. 9,10,11), much more effort is required to understand their reproductive biology. Clearly, further research is needed to fill these gaps and to compose a clearer picture of this important resource, to evaluate their potential use and ultimately their management and conservation.

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PRE-BREEDING ECOLOGICAL CHARACTERIZATION OF GERMINATION IN *AEGILOPS* (POACEAE)

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The genus *Aegilops* L., nom. cons., represents the secondary gene pool of wheat and some *Aegilops* species played a key role in wheat evolution and domestication (1). Wheat Wild Relatives have been used in wheat breeding programs and they are considered as potential sources of useful traits for modern wheat improvement, especially in a future scenario of climate change (2, 3). A better understanding of the ecology of *Aegilops* species is therefore important to improve their use in breeding programs and strengthen their *in/ex situ* conservation. This work was undertaken as part of the initiative "Adapting Agriculture to Climate Change: Collecting, Protecting and Preparing Crop Wild Relatives" which is managed by the Global Crop Diversity Trust with the Millennium Seed Bank of the Royal Botanic Gardens.

In the first steps of this project, we focused our research on germination of *Aegilops* spp. under different temperatures (15°C, 20°C, 25°C) and different levels of osmotic stress (0 MPa, -0.8 MPa, -1MPa, -1.3MPa,-1.5MPa, -1.7MPa) to imitate drought stress, since seed germination is a key event in the life cycle of plants and is mainly driven by temperature and moisture availability in the soil. Seeds of two different species of *Aegilops* (*Ae. geniculate* Roth and *Ae. neglecta* Req. ex Bert.) were collected in the field according to ENSCONET Seed Collecting Manual (4) from different populations along a latitudinal gradient and germination was tested on fresh seeds (5). Germination of *Aegilops* was compared to different accessions of *Triticum aestivum* L. and *T. durum* Desf., including a drought intolerant cultivar, used as a reference of the germination performance. The experiment followed a full factorial design.

Our results show that seeds of *Ae. geniculata* and *Ae. neglecta* tolerate high degrees of osmotic stress. Northern populations of *Ae. geniculata* showed a more negative effect of osmotic potential on germination than southern ones. Moreover, southern populations of *Ae. geniculata* performed as well as different accessions of *Triticum aestivum* and *T. durum* used as a reference. Additionally, the effect of osmotic potentials differed among seeds from different positions on the spike, but only in the populations from northern Italy. These results suggest that seeds of populations from different latitudes vary in their resistance to drought. Our results also underline the importance to apply a population approach in the pre-breeding characterization of Crop Wild Relatives, considering the different ecological features of different populations of the same species that often experience a wide range of abiotic conditions across their distribution.

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TESTING THE POTENTIAL OF VINEYARDS TO CONSERVE PLANT SPECIES OF DRY CALCAREOUS GRASSLANDS

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Agriculture is among the main causes of biodiversity loss worldwide (1) and there is increasing awareness that future biodiversity conservation will largely depend on the capability of cultivated areas to provide suitable habitats for species of conservation concern (2, 3). The increasing development of vineyards in Mediterranean areas is a major driver of loss of dry grasslands (4) that are targeted for biodiversity conservation by the EU according to Natura 2000 policies. It is therefore crucial to evaluate the potential of vineyards to provide surrogate habitat for the restoration and conservation of plant communities of dry grasslands. This study was carried out in one of the economically most important winemaking districts of Italy (Conegliano-Valdobbiadene DOCG, Veneto) characterized by a hilly landscape with steep slope vineyards (Fig. 1, 2). In this region dry grasslands and their associated plant communities are severely declining due to agriculture intensification, being replaced by vineyards even on the steepest slopes. Forty vineyards on steep slopes and 20 remnants of dry calcareous grasslands were selected along a landscape gradient. We tested the effect of cover of semi-natural habitats in the landscape and herbicide frequency and their interaction on plant species richness, as well as on floristic and functional similarity between vineyards and arid grasslands. In spite of the beneficial effect of semi-natural habitats in the landscape to plant richness of steep slope vineyards, our results indicate that vineyard areas are not fully effective for the conservation of the species of dry grasslands. Local management impacted plant diversity, also interacting with landscape composition. In particular, the use of herbicides was detrimental for plant diversity and seems to offset the positive effect of semi-natural habitats in the surrounding landscape. Herbicides were mainly composed by glyphosate whose negative effects on plant diversity are related to the pauperization of the seedbank and the selection of a pool of glyphosate-resistant species that tend to dominate the community (5). Results concerning the compositional and functional overlap between vineyards and dry grasslands suggest a scarce permeability between these two habitats and the low occurrence in vineyards of plants associated with dry grasslands corroborates this view. The effectiveness of vineyard hilly landscapes for biodiversity conservation should be improved by (a) decreasing the current management intensity for weed control, (b) strictly conserving the remnants of dry grasslands (Fig. 3) that are irreplaceable refugia for a target community of conservation concern, and (c) enlarging existing edge areas between vineyard and forests that would increase suitable habitat area for specialists of dry grasslands.

Fig. 1. The hilly landscape of the Conegliano-Valdobbiadene DOCG

Fig. 2. Steep slope vineyard

Fig. 3. Remnant of dry grassland

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SELPIBIOLIFE: INNOVATIVE SILVICULTURAL TREATMENTS TO ENHANCE SOIL BIODIVERSITY IN ARTIFICIAL BLACK PINE STANDS

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The world's forests play an important role in maintaining fundamental ecological processes, such as water regulation and carbon storage, as well as in providing livelihoods and supporting economic growth. As the home of two-thirds of all plants and animals living on land, forests are the most biodiverse terrestrial ecosystem. In Italy the forest management has been based mainly in obtaining feature improvements productive and protective. The focus on "sustainability" of forest management has never occupied the soil biodiversity. The soil plays an important role in forest ecosystems: the functionality of the soil is closely related to the functionality of the root apparatus, the dynamics of the succession forest and is home to micro- and mesofauna, fungi and plant diversity.
Forest canopy is the active interface between 90% of terrestrial biomass and atmosphere determining the incidence of solar radiation and its transmission to the soil. Therefore canopy has a decisive central role in the ecosystem dynamics, affecting photosynthesis, air and soil temperature, evapotranspiration.
SelpiBioLife is a multidisciplinary project financed by the European Union under the category Biodiversity (LIFE13 BIO/IT/000282), for innovative or demonstration projects that consider biodiversity issues within the LIFE+ Nature and Biodiversity strand. The main goal of the project is to demonstrate the positive effects of an innovative silvicultural treatment in young black pine forests. The treatments applied in the stands improves growth rates and stands stability, as recognized for other vegetations, and enhance the level of biodiversity of the various biotic soil components (flora, fungi, bacteria, mesofauna, nematods and microarthropods), according with the EU Biodiversity Strategy to 2020 and the European Atlas of Soil Biodiversity.
In particular, the project aims are to evaluate the effects of selective thinning compared to traditional thinning (selecting trees from below leaving well-spaced, highest-quality trees) and to none management in two areas localized in Pratomagno and on Mount Amiata (Tuscany) where in the past silvicultural treatments were not carried out. The goal is to demonstrate that the selective thinning not only improves the growth rate of the trees and the stability of the stands but increases the overall biodiversity.

THE REVENGE OF AMERICAN GRAPES: HISTORY AND INVASIVENESS OF ALIEN *VITIS* (VITACEAE) OF CROP INTEREST IN ITALY AND EUROPE

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If today we can taste a glass of Tuscany's Chianti, Pantelleria's Passito or Oltrepò Pavese's Bonarda, the fruits of 'Italia' grape or Panettone's raisins, we owe this privilege not only to an ancient Italian agricultural tradition, but also to American grapes. Although these high-quality products are obtained from Euro-Mediterranean *Vitis vinifera* L., a number of *Vitis* species from across the Atlantic Ocean played a key role in preserving one of the most ancient crop production originated in the Mediterranean basin.
After the introduction of 'Isabella' (a hybrid between the American *V. labrusca* L. and *V. vinifera*) in Europe at the beginning of the 19th century (7), new American hosts unintentionally reached the old contintent: in 1845, the first cases of powdery mildew [*Uncinula necator* (Schwein.) Burrill] were recorded, soon followed by phylloxera [*Daktulosphaira vitifoliae* (Fitch, 1855)], and downy mildew [*Plasmopara viticola* (Berk. & M.A.Curtis) Berl. & De Toni], catastrophic diseases which, within less than 50 years, changed forever the European viticulture scenario (4).
Agronomical and botanical knowledge led to the identification of the *Vitis* species from North America as the solution to the problem, being the only taxa resistant to the compatriot diseases; their root system proved to be immune from phylloxera, while the foliage resistant to powdery and downy mildew. Two different strategies were adopted, employing both "pure" American species and American-American (mostly involving *V. berlandieri* Planch., *V. riparia* Michx., and *V. rupestris* Scheele) and Euro-American hybrids: 1) production of wine directly obtained from the hybrids (known as "direct producers"); 2) grafting of *V. vinifera* cultivars ("vitigni") on hybridogenic rootstocks. In the course of the decades, the latter alternative prevailed on the former (at least in Europe), although wines from American- and Euro-American crossings, such as Clinton (*V. labrusca* × *V. riparia*) and Bacò (*V. riparia* × *V. vinifera*), met a relevant success until the 1950's in countries like Italy and France (4).
Although the first cases of spontaneously growing American *Vitis* taxa in Europe date back at least to the 1890's (1), botanists began to become acquainted with this phenomenon only 120 years later (see e.g., 5, 6). Thanks to subsequent studies (e.g., 2, 3), American species and artificial hybrids have been demonstrated to escape, spread and form self-perpetuating populations, often behaving as noxious invasive in both anthropogenic and natural environments (such as alluvial forests and mediterranean maquis) of most southern Europe (1). The reproductive and ecological autonomy, along with an environmental impact usually greater in hybrids than in parental species, led the authors to formally describe three new nothospecies, widely employed as rootstocks and naturalized/invasive over Italy and Europe: *V.* ×*instabilis* Ardenghi, Galasso, Banfi & Lastrucci (= *V. riparia* × *V. rupestris*), *V.* ×*koberi* Ardenghi, Galasso, Banfi & Lastrucci (= *V. berlandieri* × *V. riparia*), and *V.* ×*ruggerii* Ardenghi, Galasso, Banfi & Lastrucci (= *V. berlandieri* × *V. rupestris*) (1). This marked a separation of the taxonomic investigation from the misleading ampelographic approach which commonly involved the previous botanical contributions (1).
Currently, studies on systematics and distribution of Euro-American hybrids and seed reproduction of the taxa occurring in Italy (the latter in collaboration with the Seed Ecology Laboratory of the Pavia University) are being carried out by the authors. The results may serve as basic tools to enable the possible request for the inclusion of the invasive taxa (*V. riparia*, *V.* ×*instabilis*, and *V.* ×*koberi*) in the Union list of invasive alien species recently introduced by the EU Regulation No. 1143/2014.

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IMAGE ANALYSIS APPLICATION ON WATERLOGGED ARCHAEOLOGICAL *PRUNUS* REMAINS FROM A MEDIEVAL CONTEXT IN SARDINIA

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Fruit remains such as cherries, plums, sloes and peaches are frequently recovered from archaeological waterlogged contexts. During an archaeological excavation in the city centre of Sassari (Italy), in 2007, a medieval well, dated at 1330-1360 AD, was discovered. The sediment appeared very rich in waterlogged plant remains and a consistent presence of *Prunus* endocarps, attributed to different species, was recorded (1, 2). Thanks to the exceptional state of preservation of the remains, the morphological and morphometric analysis was performed. The discrimination among *Prunus* species is routinely based on visual evaluations of some traditional morphological characters of the endocarps. Currently, computer vision and image analysis techniques represent a more accurate, reliable and repeatable method to distinguish wild species from cultivated ones (3, 4). Digital images, acquired with a flatbed scanner, were processed and analysed using the image analysis software KS-400 (Carl Zeiss Vision, Germany). A macro, specifically developed to measure endocarps of the *Prunus* L. genus, allowed measuring 33 morpho-colorimetric features and 80 elliptic Fourier descriptors (EFDs).

In this work, the results obtained from the comparison between archaeological endocarps and the modern samples of *Prunus* collected in Sardinia, are presented. A clear statistical discrimination among 17 *P. domestica*, 111 *P. domestica* subsp. *insistitia* and 130 *P. spinosa* archaeological endocarps, was achieved. Moreover, from the comparison with the modern endocarps samples, the medieval ones were correctly identified with an overall percentage of 98.4 %.

The LDA applied an archaeological *Prunus* endocarps preserved in waterlogged contexts have allowed us to investigate the taxonomic level of *Prunus* species present in the medieval period in Sardinia.

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PRIMITIVE CULTIVAR OF *VITIS VINIFERA* L. FROM THE BRONZE AGE OF SARDINIA

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A recent discovery of waterlogged grape pips (1), recovered from three wells dating back to Middle and Late Bronze Age (ca. 1350–1150 BC) in the archaeological site of Sa Osa (Cabras-Oristano, Sardinia), allowed investigating the domestication process of *V. vinifera* and verifying the possibility that primitive cultivars might have already existed in Sardinia during the Bronze Age.

A morphological comparison of archaeological seeds and modern wild and cultivated Sardinian grape pips was performed to determine the similarities between them.

Modern seed materials were collected from 13 wild populations (*V. sylvestris*) and 37 traditional cultivars (*V. vinifera*) grown in Central-West and South Sardinia. In addition, two Italian and French cultivars and 25 wild plants, grown by the Agricultural Research Agency of Sardinia (AGRIS) for five years through propagation by cuttings, were also sampled.

Digital images were acquired using a flatbed scanner and then processed and analysed using a macro specially developed, with the image analysis software KS-400 (Carl Zeiss Vision, Germany), to characterise wild seeds (2, 3, 4). A total of 98 morphometric features were measured on 98,338 grape pips. The recorded data were statistically analysed, applying the stepwise Linear Discriminant Analysis method (SPSS Inc. 2006), to compare the modern cultivars with the archaeological seeds, which were considered as unidentified specimens.

The results showed that the archaeological seeds from the Middle Bronze Age have intermediate morphological traits between modern wild and cultivated grape pips of Sardinia. In contrast, the analyses performed on the archaeological seeds from the Late Bronze Age showed a high degree of similarity with the modern cultivars. These results provide the first evidence of primitive cultivated *V. vinifera* in Sardinia during the Late Bronze Age (AMS radiocarbon dated at 1286–1115 cal. years BC, 2σ interval). This evidence may support the hypothesis that Sardinia could have been a secondary domestication centre of the grapevine, due to the presence of ancient cultivars that still exhibit the phenotypic characteristics of wild grapes.

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THE GHIRARDI BOTANICAL GARDEN: A "NATURAL LABORATORY" FOR STUDYING THE MORPHOLOGICAL AND PHYTOCHEMICAL DIVERSITY OF SELECTED MEDICINAL PLANTS IN AN INTEGRATED ECOLOGICAL FRAMEWORK

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The Botanical Garden "G.E. Ghirardi" (BGG, Toscolano Maderno - Brescia) of the Department of Pharmaceutical Sciences of the Milan University has a long tradition in the cultivation, study and preservation of the genetic resources of officinal species, in accordance with the priority tasks of the Convention on Biodiversity.

Recently a dedicated research project started, integrating multidisciplinary analyses on target officinal species preserved at BGG, with the principal aim of characterizing these "plants" as interacting components of their own ecosystem. Indeed, the project was planned combining a dual perception: phytocentric, through the study of the biotic mutualistic interactions, mediated by the emission of secondary metabolites, and anthropocentric, referring to the potential importance to humans in the therapeutic, food and cosmetic sectors.

The work program includes: *(i*) the census of the plant heritage preserved at BGG; *(ii)* the selection of target-species for the morphological characterization of the secretory tissues and for the analysis of phytochemical profiles; *(iii)* the analysis of the biological activities of the secondary metabolites; *(iv)* case-studies on their ecological and functional roles.

In this perspective, BGG will represent a "natural laboratory" for studying the morphological and phytochemical diversity of officinal plants in an integrated ecological framework, aiming at assessing their importance for potential future exploitation by humans.

As preliminary activities of the project, we addressed our interest towards a better understanding of the micro-morphology and anatomy of the secreting structures, and on the characterization of VOC and essential oil profile of target-species belonging to the Lamiaceae family: *Ballota acetabulosa* Benth., *Ballota rupestris* Vis., *Lavandula angustifolia* Mill., *Lavandula dentata* L., *Salvia greggii* Grey, *Scutellaria sieberi* Benth., *Scutellaria altissima* L. and *Scutellaria brevibracteata* Stapf.

We report the results of the preliminary micromorphological observations on *a)* the structure and distribution of glandular tissues, *b)* the histochemical nature of the secreted substances, *c)* the mode of production and release of the secretory products. In addition, the VOC fingerprint and the characterization of essential oil produced in the glandular tissues are presented.

FOOD TRACEABILITY: A NOVEL APPROACH FOR WILD EDIBLE MUSHROOMS

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The concept of traceability is used to define the origin, quality and itinerary of food from raw material to finish products, with the purpose to guarantee product safety in terms of regulations and risks to consumer health (1). The growing interest in food quality leads to a pressing need to develop new methods and technologies to ensure the authenticity of geographic origin of food products aimed to guarantee protection from fraud cases.

In this context the results obtained by the use of a field portable energy dispersive X-ray fluorescence spectrometer for traceability purposes are presented. Specifically, we applied this methodology to mark the geographic site of origin of wild edible mushrooms with a particular interest to one of the most market-demand species: the *Boletus edulis* group (2). Some studies (e.g. 3, 4) emphasized the importance of studying the geology, soil-mineralogy and soil-chemistry of an area for the identification of the geographic site of origin of non-timber forest products, like mushrooms.

Some samples of *Boletus aestivalis* (Paulet) Fr. and underlying soil portions, referring to both the surface horizon and to deeper soil, were recorded in two different areas of Liguria (NW Italy). The first site is dominated by *Quercus cerris* L. and lies in a geologically complex area characterized by different parent rocks, such as: serpentineschists, calceschists, chlorite-actinolite schists and conglomerates. The second site instead, is characterized by the presence of *Fagus sylvatica* L. and lies in a geologically homogeneous area characterized by soils developed exclusively on calceschist.

Chemical analyses performed on both sporomata and soil specimens revealed that the soil mineralogy and geology of a site deeply influence the elements content in wild edible mushrooms. More specifically, statistical analysis revealed that the concentration of major, minor and traces elements detected in each sporomata is directly correlated with their concentration in soil portions. Moreover, the trace content allows to distinguish sporomata samples on the basis of the geographic site of origin. In addition, the positive correlation existing between minor elements and traces found among sporomata and soil portions indicates that these elements can be considered potential soil geo-marker.

The results obtained by this study suggest that geological and chemical soil information can fruitfully be used for traceability purposes to distinguish the authenticity, the quality and the provenance of food products and to potentially protect them from the uncertified and the unknown-origin products.

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FATTY ACIDS COMPOSITION OF KERNEL OIL FROM SIX DIFFERENT HAZEL (*CORYLUS AVELLANA* L.) POPULATIONS IN ITALY

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Hazelnut kernels are rich in fats, proteins, and vitamins and play a relevant role in the agricultural market, mainly because of their use to provide flavor in dairy, bakery, candy, confectionery, and chocolate products. Hazelnut have functional properties due to their content in fatty acids and phenolic compounds that could positively affect human health. Italian hazelnut production is 85,232 10^3kg year^{-1} for in-shell product (1). Particularly, Campania, Lazio, Piemonte, and Sicilia account for 98% of the national production. About 90% of the world crop is sold as kernels and processed in the food industry (i.e. chocolates, bakery, dairy), and the remaining 10% is sold as in-shell product and consumed fresh, blanched, or roasted (2). The food industry requires uniform high-quality nuts and precise morphological, chemical, and physical kernel characteristics, as well as absence of defects (3,4).

In this study, the fatty acid content of kernel is investigated in six hazel populations from different sites of Italy. The 3 sites of Northern Italy are: Natural Reserve Bosco Siro Negri (BN - Lombardy, natural broadleaved Oak forest of Ticino alluvial plain), Lequio (L - Piedmont, hazel plantation, 650 m a.s.l., Langhe hills) and Alba (A - Piedmont, hazel plantation, 250 m a.s.l., Tanaro alluvial plain). In site BN wild type *Corylus avellana* is present while in sites L and A the hazel cultivar is 'Tonda Gentile Trilobata' (previously 'Tonda Gentile delle Langhe'). The 3 sites in Central Italy (Lazio, Tuscia hills) are: Nepi (N1 – hazel plantation with surface drip irrigation, 250 m a.s.l.), Nepi (N2 – hazel plantation with sub-surface drip irrigation, 250 m a.s.l.), Capranica (C unmanaged hazel cultivation, 350 m a.s.l.). The hazel cultivar 'Tonda Gentile Romana' is grown in sites N1, N2 and C.

In each site fully ripe hazelnuts were collected. Crude oil was obtained from finely chopped nuts, using the Soxtherm automatic extraction instrument (Gerhardt Analytical system, Germany).

The fatty acid composition of hazelnut oil samples was determined from total lipid extracts as methyl esters (FAMEs) by gas chromatography (GC) coupled to mass spectrophotometry, according to methods described in regulation of EEC 2568/91 (5-6). The GC reference standard FAME mix GLC-10 was used to identify and quantify the FAMEs.

Three extracts have been prepared from each site sample and 2 injections for each extract were performed. Kruskal-Wallis Analysis was used to test the significance of the differences among concentrations and Tukey's HSD to perform post-hoc pairwise tests. The results are reported in Tab. 1.

	N1	N2	C	BN	L	A		N-C	BN	L-A
Palmitic Acid PA	7.5	5.9	6.6	9.4	5.9	7.0		6.6	9.4	6.2
Linoleic Acid LA	4.5	4.7	5.4	16.4	3.6	3.7		4.7	16.4	3.6
Oleic Acid OA	81.3	68.2	68.0	126.1	70.3	71.5		68.3	126.1	70.8
Stearic Acid SA	2.3	2.1	2.0	2.4	2.6	2.9		2.1	2.4	2.6
Total Fatty Ac. TFA	96.6	81.1	81.3	159.1	83.6	85.2		82.9	159.1	83.8

Tab. 1 - Content of fatty acids (mg g^{-1}) in the 6 populations (see text) and in the 3 varieties of Hazel (N-C: 'Tonda Gentile Romana', BN: wild type. L-A: 'Tonda Gentile Trilobata')

The median concentrations of PA, LA, OA and TFA are always significantly different among BN and all the other populations and among BN and the two cultivars (N-C, L-A). The results of the analysis agree with the literature data about the content of fatty acids in hazelnut oil of hazel cultivars (4) but wild type hazel form Natural Reserve Bosco Siro Negri proves to have the highest fatty acid content.

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TOTAL VOLATILE PROFILE OF *ARTEMISIA UMBELLIFORMIS* LAM. AS A PREDICTIVE MARKER OF THUJONE AMOUNT IN GENEPÌ LIQUEUR

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Artemisia umbelliformis Lam. is one of the Alpine *Artemisia* species used to prepare genepì, a traditional liqueur characterized by a bitter taste and a peculiar flavor (1). These properties are known to be related to both sesquiterpene lactones and volatile fractions, whose major components are α- and β-thujones (2, 3). Thujones are natural terpenoids also associated with absinthe-based products (*Artemisia absinthium* L. and *Artemisia pontica* L.) whose toxicity is currently the object of a heated debate (4). European Union legislation has imposed a limit of 35 ppm on the total amount of these compounds in *Artemisia*-based beverages (5) therefore, thujone-free chemotypes of *A. umbelliformis* have been horticulturally selected to overcome this issue and the chemical and biomolecular differentiation between differently cultivated chemotypes was investigated (4).
Since genepì producers prefer thujone chemotypes in the preparation of their precious liqueur to obtain a better flavor and because of the chemical variability of *A. umbelliformis* also when cultivated, a plant-based prediction technique is required to produce liqueurs in agreement with the European legislation. The aim of this study was to create a prediction model to determine the thujone concentration in the liqueurs before their production. The model here proposed results from the development of an analytical platform based on the combination of fast analyses of volatiles, advanced technologies of quantitation, and multivariate statistical modeling.
Forty *A. umbelliformis* individual plants cultivated at 1300 m a.s.l. in Pragelato (Turin, Italy) were used as experimental set to correlate α- and β-thujone amounts both in plants and in the liqueurs prepared with them. Multiple Headspace Solid Phase Microextraction (MHS-SPME) coupled to Gas Chromatography and Mass Spectrometry (GC-MS) was used to quantify the two compounds in both matrices (6 and references cited therein). MHS-SPME collected data were first analyzed trying to find a linear correlation between thujone amount in plant(s) and liqueur(s) but a multivariate approach had to be adopted, since the results were not satisfactory.
Each plant was then analyzed by Headspace (HS)-SPME-GC-MS and 27 volatiles were used to create a solid multivariate matrix. The plants then were divided in two groups as categorical response variable, in function of the 35 ppm EU limit. Linear Discriminant Analysis (LDA) (7) was used as the main statistical tool to discriminate between different thujone-level groups. This is a linear modeling technique whose goal is to create a multidimensional algorithm that could describe the distribution of samples known to belong to different groups. LDA, which is commonly performed in two steps, calculates the significance of the model basing on known samples (Cross Validation, CV) and then the model predictive power based on unknown samples. Results of multivariate correlation were surprisingly good with a mean CV of 0.97 (median CV = 1.00) and mean prediction of 0.87 (median =0.89).
The last step of this study was the development of a non-separative method to speed up the analysis so that the 70 minutes required from the HS-SPME-GC-MS analysis were reduced to 10 minutes with HS-SPME-MS analysis. The 315 MS fragments resulting from HS-SPME-MS were used as variables to create the multivariate matrix and to perform a successful LDA prediction model, i.e. 0.98 of CV mean (median CV = 0.98) and mean prediction of 0.81 (median = 0.81).

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METABOLITE PROFILING AND ANTIBACTERIAL ACTIVITY OF *MYRCIANTHES HALLII* (MYRTACEAE) USED IN TRADITIONAL MEDICINE IN ECUADOR

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Myrcianthes hallii (O. Berg) McVaugh (Myrtaceae), is a medicinal and aromatic plant commonly known in Ecuador as "arrayán" [1]. In traditional medicine, arrayán is used for its astringent, antiseptic, haemostatic, hypoglycemic and balsamic properties. Moreover, it is effective against pulmonary disorders, diabetes and it is used to treat night sweats of tuberculosis and as wound healing. Despite the long history of use as traditional medicine, no literature data are available regarding its chemical composition and antimicrobial properties. Therefore, the aim of the present study was to evaluate the metabolic profiling and antibacterial activity of arrayán.

The investigation was performed on a hydromethanolic extract obtained from dried leaves of *Myrcianthes hallii*. The first aim was to determine the phytochemical composition of arrayán. Consequently, the extract was submitted to a preliminary purification through dialysis (molecular weight cut-off of 3500 Da) and then analyzed by UHPLC-PDA-hESI-MSn. Moreover, the extract was submitted to microbiological assays to determine the antibacterial activity. Ten clinical *Staphylococcus aureus* strains (five methicillin-resistant and five methicillin-susceptible) were studied. Among those resistant, three strains were multi-resistant (resistant to at least three classes of antibiotics). The antibiotype was determined using the disk diffusion test, according to the Clinical and Laboratory Standards Institute guidelines. Minimum inhibitory concentration (MIC) was determined by using the broth microdilution method, according to the Clinical and Laboratory Standards Institute guidelines.

The results showed that the extract exerted antibacterial activity against both methicillin-resistant and methicillin-susceptible *Staphylococcus aureus* strains (MIC > 1 mg/mL). Although methicillin-resistant strains usually display resistance to several drugs, no relevant differences were observed between methicillin-susceptible and resistant strains. These data ***agree with*** earlier studies carried out on another plant species belonging to *Myrcianthes* genus *(Myrcianthes cisplatensis),* which showed antibacterial activity against methicillin-sensitive and resistant *Staphylococcus aureus* strains.

The UHPLC-PDA-hESI-MSn analysis allowed the identification of more than thirty compounds, including organic acids, phenolic acids, and flavonoids. According to the results obtained from UHPLC-PDA-hESI-MSn analysis, the appreciable antibacterial activity against *S. aureus* of MHE is justified by the presence of a wide spectrum of polyphenols identified in the extract, which act as antimicrobial agent *via* different mechanisms of action (MOA). Many MOA are ascribed to polyphenol, e.g. cytoplasmatic membrane damage, inhibition of nucleic acids, cell wall and cell membrane synthesis. Moreover, in addition to direct antibacterial activity, growing evidence suggests that polyphenols interfere with some bacterial virulence factors such as enzymes, toxins and signal receptors.

In conclusion, since there are no literature data about the phytochemical characterization of arrayán, the present investigation represents the first available study about the chemical characterization of this plant. Overall, the chemical composition and the antibacterial activity of *Myrcianthes hallii* determined in the present investigation can support the medicinal properties ***related to*** its ***traditional*** use in Ecuador [2, 3].

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CONVOLVULUS PLURICAULIS, AN AYURVEDIC HERBAL TOOL FOR HUMAN AND PLANT WELL BEING

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Ayurveda, Indian traditional medicine, is recognized by EU as non-conventional medicine and includes more than 7,000 plants used for phytotherapic purposes. One of the areas of interest that enjoys greater consideration in Ayurveda is linked to rejuvenating practices (*Rasayana*), and focuses on diseases prevention and promotion of a healthy life. *Convolvulus pluricaulis* Sieb. ex Spreng. (Convolvulaceae) is a perennial herb, traditionally used in this branch of Ayurvedic medicine (Singh et al., 2008). Furthermore, it is reported to possess anxiolytic, memory enhancing and mood elevating effect (Agarwa et al., 2014; Verma et al., 2012). *C. pluricaulis* crude drug (dried whole plant) extracts (decoction, formulated with the direction of the Ayurvedic Pharmacopoeia; hydro-alcoholic, chloroform and supercritical fluids extracts) were phytochemically investigated with different chromatographic approaches (GC, HPLC and HPTLC) to detect and quantify the most characterising molecules. Chemical analyses results showed that DEC was characterised by the presence of phenolic acids: caftaric acid, caffeic acid, p-coumaric acid, iso-ferulic acid and tr-ferulic acid. Stigmasterol, β-sitosterol, lupeol, vanillin isomers and derivatives (2-hydroxy-4-methoxybenzaldehyde, 3-hydroxy-4-methoxybenzaldehyde and acetovanillone) were also detected in the same preparation. The hydro-alcoholic extract (HE) showed the presence of p-coumaric acid, vanillin, 2-hydroxy-4-methoxybenzaldehyde, acetovanillone, and vanillic acid. Identification and quantification of the phenolic acids in DEC and HE by gas chromatography was performed following a full validated method (Caligiani et al., 2013). The chromatograms of the botanicals derived from $CHCl_3$ and supercritical fluid extractions exhibited the highest molecular variety. In particular, the main constituents of these phytocomplexes were phytosterols (campesterol, stigmasterol and β-sitosterol) and terpenoids (e.g. lupeol). Among them, two acyclic diterpene alcohol precursors of vitamin E: phytol and isophytol, not present in the preparations considered so far (DEC and HE). The study of *C. pluricaulis* extracts biological activities, performed through a bioassay guided method, was driven both by the ethnomedical traditional uses of the species in the Ayurvedic culture and by those aspects related to modern western phytotherapic culture concerning the prevention of the oxidative stress (antioxidant activity), the evaluation of both genotoxic (safety) and antigenotoxic potential, and cytotoxicity against cancer and normal human cell lines. The antioxidant activity evaluation indicated DEC as the phytocomplex that exhibited activity in every performed test (ABTS, DPPH and β-carotene bleaching test), while HE showed a higher activity than DEC in the ABTS test, no activity against DPPH radical and a lower capacity of blocking the radical propagation than DEC in the β-carotene bleaching test. Regarding the cytotoxic capacity of the considered preparations against A549 and MCF7, cancer cell lines, the growth inhibition experimental data, compared with the IC_{50} threshold stated by the American National Cancer Institute (<30 μg/ml), are not significant. The tests carried out with DEC and HE extracts against two leukaemia cell lines, drug-sensitive (CCRF-CEM) and drug-resistant (CEM/ADR5000), showed, instead, interesting results. The DEC $CHCl_3$ extract and the HE soxhlet extract exhibit experimental data in line with the one stated by the American National Cancer Institute against the CCRF-CEM cell line, but respectively about 2 and 3 fold bigger in the tests against CEM/ADR5000, showing cross-resistance phenomena. These experimental results are innovative in the bioactivity panorama of *C. pluricaulis* and they could be the starting point to a new research extended to other extraction methods or human cancer cell lines. Moreover, it was performed a preliminary investigation aim to establish a possible usage of the considered phytocomplexes as natural defence in integrated or organic agriculture. Therefore, the evaluation of the antimicrobial capacity was performed on four phytopathogens highlighting an interesting activity of the aqueous extract (DEC) against *Clavibacter michiganensis* and *Pseudomonas syringae*.

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POTENTIALLY NEUROACTIVE AMINES IN KIWIFRUITS

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The beneficial effects of a diet rich in fruits and vegetables on human health are generally recognized. The protective effect of a diet rich in fruit and vegetable on cardiovascular diseases and some kind of cancer has been shown in many investigations, including the large scale study using the data coming from the EPIC (European Prospective Investigation into Cancer and nutrition) initiative (Crowe *et al.*, 2011) and the very recent investigation of Oyebode and coworkers (2014). The earlier investigation inspired the launch of various national campaign such as "5-a-day" campaign in UK, France and Germany, the "Fruit and Veggies-more matters" in USA, and the "Go for 2+5" in Australia.
Some very recent investigation highlighted also a positive association between fruit and vegetable consumption and enhanced mood, happiness, psychological well-being feeling (White *et al.*, 2013, Carr *et al.*, 2013; Blanchflower *et al.*, 2012) and decreased depression (Tsai *et al.*, 2001).
However, similar reports referred on specific fruits or vegetables are very rare. Carr and coworkers (2013) reported a specific positive association between the consumption of two kiwifruits per day and less fatigue, more vigour and overall enhanced mood state, while Lin and coworkers (2011) found that kiwifruits seems to improve sleep onset, duration, and efficiency in adults. The precise molecule(s) responsible for these activities have not been yet identified. White and Carr speculated that the observed kiwi fruit effects could be due to the high content of vitamins (mainly vitamin C, D, E), folates, carotenoids, flavonoids, omega-3-fatty acids and micronutrients, while Lin and coworkers speculatively attributed the observed effect on vitamins, antioxidants and serotonin, which has been previously detected in this fruit.
Recently, in a project aimed to the metabolomic- characterization of kiwifruits, we found that, beside the presence of vitamin C and various different polyphenols, an interesting cocktail of metabolites, which potentially could be involved in the psycoactivities of this fruit, have been detected. This phytocomplex included tryptophan, tryptamine, serotonin, N-acetyl serotonin and melatonin, i.e. the complete biosynthetic pathway for the production of phytomelatonin.
The putative gene responsible for tryptamine production in kiwifruit was identified and it was characterized by phylogenetic comparison with that of other plant species and by its heterologous expression in *Nicotiana benthamiana*.

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Aknowledgment: this project was funded by Regione Veneto.

PHENOLIC ACID COMPOSITION AND ANTIOXIDANT ACTIVITY IN EINKORN AND WHITE FLOURS, SOURDOUGH BREADS AND IN-VITRO DIGESTED PRODUCTS

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Epidemiological studies have linked the consumption of whole grain products to the prevention of several chronic diseases such as type 2 diabetes, cardiovascular diseases and obesity (1). Even though the precise mechanism behind this protection is not yet completely known, an antioxidant effect might be implicated. Whole cereals are an important source of beneficial compounds endowed with interesting biological activities, and processing has an important role for the maintenance of both sensory and nutritional quality of grains.

This work has been carried out in the frame of "BAKE4FUN" European project (BSG-SME, proposal number 606476), which has, among others, the aim of designing, validating and developing innovative health promoting bakery products.

Phenolic acid profile has been investigated in an ancient einkorn flour (AF), a standard white flour (SF), and in derived breads obtained through different sourdough fermentation (SD) processes. Moreover, to gain information about the bioavailability of phenolic acids in cereals subjected to different processing procedures, samples derived from a dynamic *in vitro* system simulating gastrointestinal digestion are evaluated.

Original methods have been developed for the determination of ferulic acid, gallic acid, 4-hydroxybenzoic acid, *p*-coumaric acid, caffeic acid, sinapic acid and *trans*-cinnamic acid. Cereal matrices were treated with a methanol/acetone/water mixture, and extraction was carried out separately on supernatant and residue to obtain the free, soluble and insoluble-bound phenolic acids fractions, respectively. *In vitro* digested bread samples have been injected as such after concentration by freeze-drying and water reconstitution. HPLC analyses were performed by with a photodiode array (PDA) detector. Antioxidant activities have been assayed by different *in vitro* methods (DPPH, ABTS, FRAP-FZ, ORAC).

Among the phenolic acids analyzed, ferulic acid (FA) is the most abundant one. It is mainly found in the insoluble bound fraction, where it accounts for 91 to 97% of total ferulic acid, confirming that it is linked to arabinoxylans and to other polysaccharides or proteins located in the cell walls of the aleurone layer of the grain. AF has a significantly higher FA content than SF. Breads prepared from both AF and SF by SD fermentation process showed a significantly higher amount of free FA compared to the corresponding breads prepared by conventional fermentation. Antioxidant capacity was strictly correlated with the FA content in the bound form, thus suggesting that the high content of phenolic compounds is the main contributor to the activity.

Work is in progress to analyze the bioavailability of phenolic compounds in AF and SF breads in order to select the optimal processing conditions for a good maintenance of functional compounds and biological activities.

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FREE FATTY ACIDS IN *TUBER AESTIVUM – TUBER UNCINATUM* SPECIES COMPLEX AND ITS CHEMOTAXONOMIC SIGNIFICANCE

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Tuber spp. are ascomycete fungi that establish an ectomycorrhizal symbiosis with trees and shrubs. Owing to their favourable organoleptic properties, truffles are highly beloved and appreciated in gastronomy and posses huge commercial value. After Chatin's (1) first report of the nutritional components of truffles, more recent studies have revealed 5-9% of their dry weight to be just lipids and a relatively high amount of unsaturated fatty acids in their *Tuber* fruiting bodies (2, 3).
Among the most common edible European truffles are the debated *Tuber aestivum* Vittad. and *T. uncinatum* Chatin (4). Whether *T. uncinatum* and *T. aestivum* are infact a single species or two distinct species has long been the subject of dispute. Given their different smells and tastes, *T. uncinatum* and *T. aestivum* are infact two separate truffle morphotypes with different market values. *T. uncinatum* differs from *T. aestivum* essentially in the presence of hooks in the pore reticulum; the apparently deeper colour of the gleba, the smaller, non-transversally striated facets of the peridium and in the late autumnal maturation on its fruiting bodies. The recent use of molecular markers to typify the ascocarps of these two morphotypes have reached contrasting conclusions (5, 6). In chemotaxonomical studies of truffles compounds such as nucleic acids, proteins, and enzymes have often overshaddowed the subject of Fatty Acid (FA) profiles. Therefore, the aims of this study were to analyze the content and composition of Free Fatty Acids (FFA) present in methanolic extracts of *T. aestivum* and *T. uncinatum* ascocarps, and to evaluate their significance to differentiate truffles belonging to the *T. aestivum* and *T. uncinatum* morphotypes.
The results have shown that linoleic acid was the major FFA detected in both morphotypes. In addition to linoleic acid, oleic acid and palmitic acid were abundant FFAs in the *T. aestivum* and *T. uncinatum*. The *T. aestivum* and *T. uncinatum* truffles gain an added value from the elevated nutritional worth of the linoleic acid and oleic acid present in their composition.
A hierarchical cluster analysis of FFAs present in methanolic extracts from *T. aestivum* and *T. uncinatum* ascocarps underlined the presence of two 'naturally' distinct groups according to the harvesting period: summer and autumn respectively. This research is supported by Fondazione Cassa di Risparmio di Perugia (Perugia, Italy), project code 2014.0094.021.

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POSTERS

1. = PLASTICITY OF THE FLORIGEN ACTIVATION COMPLEX DURING PHOTOPERIODIC FLOWERING IN RICE

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Rice is a short day plant that flowers only when exposed to short days (SD). The reference Asian accession Nipponbare requires exposure to a minumum of 12 SD to be committed to flowering. During this time, two florigenic proteins called HEADING DATE 3a (Hd3a) and RICE FLOWERING LOCUS T 1 (RFT1) reach a peak of expression in leaves and are subsequently repressed after commitment. Hd3a and RFT1 are produced in leaves and migrate to the shoot apical meristem where they determine the transition from vegetative to reproductive development. *Hd3a* and *RFT1* expression in leaves is regulated by the transcription factors HEADING DATE 1 (Hd1) and EARLY HEADING DATE 1 (Ehd1) that are in turn regulated by a complex gene network also influenced by light and the circadian clock. In the shoot apical meristem, Hd3a binds the bZIP transcriptor factor OsFD1 through a 14-3-3 protein, thus forming a transcription activation complex, the florigen activation complex (FAC). The FAC is able to activate several target genes initiating a molecular cascade required to start inflorescence development from the vegetative meristem.
We focused our studies on alternative components of the FAC and their functions. Our data indicate that besides Hd3a, also RFT1 can be a component of such complex. Additionally other bZIP transcription factors closely related to OsFD1 can interact with distinct components of the FAC, thus forming complexes involved in rice flowering. These data suggest a wider combinatorial plasticity between FAC components that leads to the formation of different complexes and increase the robustness of the photoperiodic response. Finally, we will present evidences indicating that the FAC can also be formed in leaves where it affects *Ehd1* expression and, as a consequence, *Hd3a* and *RFT1*. We propose a broader role of the FAC during and after floral transition both at the shoot apical meristem and in leaves.

1.1 = VERTICAL GREENING AND CONSERVATION OF THE MONUMENT SURFACE: THE ROLE OF *HEDERA HELIX* L.

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The ivy (*Hedera helix* L.) is one of the most widespread plants growing on the buildings in the Euro-Mediterranean area. Some authors underline its aesthetic value and claim its use for the direct vertical greening, with the idea to have many benefits (Sternberg et al., 2011), whereas others recognized it as deteriorating agent (Elinç et al., 2013). The biodeterioration and bioprotection processes are sometimes in a precarious balance, and probably the relative effects depends from the ecological and biogeographical conditions.
Then, in this work we evaluated the biodeterioration degree (BD) due to the ivy colonization on the stone monuments in a Roman archaeological area, where it grows spontaneously, abundantly and vigorously.
The selected study area was the Villa of Massentius, where we analysed 4 plants on the North-Est boundary wall of the Romulus's Mausoleum, which have been cut in July 2013, in order to evaluate the BD due to the ivy coverage. These 4 sites differ in microclimatic condition: the sites 1-2 are located in a point affect by shadow-humid conditions, whereas the site 3-4 are in drier ones. Near sites 1-2, we monitored the annual growth rate of 3 young climbing up plants. At the end, we analysed the effects of the capping ivy growing on the top of the ruin in North-Western internal boundary wall of the Romulus's Mausoleum.
We applied a new quantitative method, derived by the method proposed by Fitzner et al. 2001 in order to estimate the weathering forms (I, P/O, W) due to the ivy on vertical surfaces. A Damage Degree for each weathering form (D_v) was calculated to obtain the Total Damage Degree for each site (D_s). The growth rate was calculated trough a photographic monthly survey (Jan-Dec 2014), and subsequent measure of the ivy coverage percentage for each month. For the capping ivy we evaluated its deterioration ability trough the analysis of: walls penetration rate of ivy stems, paedogenesis effects of ivy cover, secondary growths of ivy due to seeds germination.
The spontaneous ivy growth in this area is wide where the microclimatic conditions are very humid. The maximum value of growth rate correspond to 11% per m^2, for the range months March-May. The mean of Back Weathering (D_W) correspond to 58% per m^2 of surface loss, and the Crumbly Disintegration with the Break Out ($D_{P/O}$) shows a loss of surface of 14% per m^2. The highest values of D_s (Total Damage Degree) resulted for site with higher ivy cover percentage and in shady condition. At the end, in the situation of the capping Ivy, we evaluated on average the stone loss of 23.87 cm^3 on a ivy covered surface of 5,5 m^2. Consequently, we can state that the deteriorative action ivy on the walls is not negligible, and its dangerousness changes in relation to environmental variables and cover degree. It was demonstrated that the damage degree increases in humid and shady conditions that correspond to the optimal ecology characteristics of this species. For plans in conservation of stone monuments and projects of vertical greening, in order to maximize the results, and to avoid negative effects, it is necessary consider the auto-ecology of the species involved, such as methods to reduce the interaction.

Fig. 1 Stereomicroscope image air rootles of ivy

Fig. 2 Ivy's branches growing inside the wall

Fig. 3 Growth rate of ivy

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1.1 = FROM GRAPEVINES TO WINES: THE "SECRET" OF SARDINIAN VITICULTURE

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Vitis vinifera L. is largely diffused in Sardinia, both as a spontaneous wild plant (*V. vinifera* subsp. *silvestris*) and as a cultivated crop (*V. vinifera* subsp. *vinifera*). The large number of grapevine cultivars recorded (1) is the result of the complex history of Sardinia. Sardinia was part of the Spanish Empire from 1479 to 1714 and, during this period, commercial and cultural exchanges between the two regions resulted in modifications to its agricultural products. In the case of viticulture, it is notable that Spanish and Sardinian grapevine varieties often have similar-sounding names; for example, 'Bobal' and 'Bovale', 'Cariñena' and 'Carignano', and 'Garnacha', 'Granaccia' and 'Granazza'. We may assume that, due to politic and economic relationships, exchanges of grape varieties could have been occurred between Sardinia and Spain, improving the genetic composition of the local germplasm. At the same time, certain Sardinian cultivars, some of which derived from the domestication and cultivation of wild local grapevines (2), could have been introduced into Spain. The first aim of our work was to evaluate the genetic structure of grapevine germplasm of Sardinia and its relationships with Spanish cultivars. Simple Sequence Repeat (SSR) markers were applied to characterize each accession and to study the relationships among Sardinian and Spanish cultivars. Results suggest the occurrence of several cases of synonymy such as 'Cannonau' and several Spanish accessions of 'Garnacha Tinta'. The genetic origin of cultivars is only one of the elements useful in wine traceability. Wine quality relies on a precise equilibrium among different factors and it is largely influenced by the environmental conditions of the production area. The second goal of this study was the characterization of 'Cannonau' wine produced in different Sardinian localities by studying the microbial diversity of berries and musts. Preliminary data suggest that microbial biodiversity, not only yeast, over time and space may play a relevant role in wine fermentation and largely influences the organoleptic characteristics of Sardinian wines.

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1.3 = ASSESSMENT OF EU AGRI-ENVIRONMENTAL REGULATIONS BENEFITS ON PLANT BIODIVERSITY IN LOMBARDY

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Over the last decades agro-biodiversity conservation has gained prominence in both scientific and policy-making circles and agro-environmental issues have been the subject of important common policies within the European Union (EU).
Since 1992, the concept of multifunctionality is promoted by the EU reformed Common Agricultural Policy (CAP) and is stated in the agro-environmental regulations (EC) No. 2078/92, No. 1257/99 and No. 1698/2005, according to which each EU member country is expected to encourage production of agricultural public goods through the development of a National Agri-Environmental Programme (NAEP).
Since 1995 Lombardy Region applied the Council regulations with the 1993-2000, 2000-2006, 2007-2013 Rural Development Programmes (RDPs). the new RDP for 2014-2020 is awaiting approval from the European Commission.
One of the goals of the agri-environmental measures proposed by the EU is the recovery of an ecological network in order to re-establish the relationships between natural elements fragmented and reduce the loss of biological diversity, so that the agro-ecosystem provides a variety of ecosystem services such as support for biodiversity. This target is a constant work to address the productive activity towards greater sustainability while allowing agriculture in Lombardy to achieving adequate levels of competitiveness.
This study was part of a research project funded by Lombardy Region, Directorate-General for Agriculture, Research and Technology Iinnovation Structure, within the Regional Programme for Agricultural Research 2007/2009. It was aimed at assessing the environmental impacts of EU agri-environmental regulations through biodiversity parameters. Plant biodiversity assessment was carried out on four different types of farming systems (FS): organic FS, FS characterized by extensive crop production, maintenance of hedges, plant rows and wooded buffer strips, integrated FS (integrated agricultural production, balanced fertilization and crop rotation), conventional FS. Phytosociological relevées were carried out in cultivated fields of rice, maize and, soya, polyphyte meadows, water-meadows, hedges and tree-lines, following the Braun-Blanquet method (1). The quantitative aspect of specific biodiversity was evaluated for each farming system plant community using STADIV pc-program (2) to calculate species richness, diversity, equitability and dominance indices. Species average cover-abundance values for each community were subjected to importance-values distribution analysis using DIVFIT pc-program (2), producing dominance-diversity curves (3).
The results show that the best plant biodiversity has been detected in organic and FS characterized by extensive crop production and maintenance of hedges, plant rows and wooded buffer strips. The percentage of alien and invasive species is lower in these two management types with respect to the integrated and conventional farming systems.
Change in plant biodiversity can provide small-scale elements as a measure of ecosystem response to land-use resulting from policy decisions affecting management of the agricultural landscape. So biodiversity assessment is a useful tool to assess multifunctionality and sustainability of different farming systems and agro-ecosystem.

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1.3 = FUNGAL BIODIVERSITY IN ARCTIC BIOLOGICAL SOIL CRUSTS

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Biological soil crusts (BSCs) are communities comprising diverse assemblages of cyanobacteria, eukaryotic microalgae, fungi, lichens, mosses, as well as heterotrophic bacteria; recently, archaeal populations have also been documented from BSCs [1]. They play many important ecological roles worldwide in all semiarid and arid lands which cover more than 30 % of the Earth' surface. Within arid environments, BSCs have even been termed 'mantles of fertility' as they are considered hotspots of biogeochemical inputs [2].
BSCs enhance soil quality by aggregating soil particles, thereby reducing wind and water erosion; they may also increase soil surface temperature and modify the water run-off infiltration balance, shuttling run-off to run-on zones in some cases and increasing soil fertility by N and C fixation. Moreover BSCs may facilitate succession to later series, suggesting that assisted recovery of BSCs could speed up succession [3].
Fungi typically form networks of highly branched hyphae, and those associated with BSCs may play several important roles within arid land ecosystems. For example, it has been suggested that filamentous hyphae link BSCs with patches of arid land vegetation, mediating nutrient exchange between these systems. Also it was demonstrated that plant nitrogen is strongly affected by interactions with mycorrhizal fungi and biological soil crusts [4]. Yet, fungal assemblage in BSCs have been mostly overlooked and their biodiversity composition remains mostly unknown.
Aim of this study was to examine the fungal biodiversity in thirteen samples of Arctic BSCs collected in different sites in the alpine Tarfala Valley, located on the slope of Kebnekaise, the highest mountain in northern Scandinavia. The BSCs analyzed were rather homogeneous in appearance; fungi were isolated and identified using both morphological and molecular approaches. Isolation data revealed that fungal assemblage composition was also homogeneous among the BSCs analyzed and characterized by low biodiversity; *Mortierella* was one of most recurrent genus of filamentous fungi, while, among yeasts basidiomycetes only were found with the genera *Rhodotorula* and *Cryptococcus*, rather common in polar environments. Notably, some species related with ericaceous mycorrhizas were isolated in all samples analyzed.

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1.3 = CHARACTERIZATION OF THE TRIPARTITE INTERACTION BETWEEN TOMATO, ARBUSCULAR MYCORRHIZAL FUNGI AND CUCUMBER MOSAIC VIRUS

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The mutualistic association established between arbuscular mycorrhizal (AM) fungi and the majority of land plants is considered a natural instrument to improve plant health and productivity; mycorrhizal plants, beside an improved mineral nutrition, often show higher tolerance to biotic and abiotic stresses (1). The AM symbiosis can influence the outcome of plant-pathogen interactions; however, the impact of the AM symbiosis on the infection by viral pathogens is still largely uncertain and little explored (2,3). In this study tomato (*Solanum lycopersicum*, Moneymaker cv) plants were grown under controlled conditions and inoculated with the AM fungus *Funneliformis mosseae*. Once the colonization had developed, plants were inoculated with *Cucumber mosaic virus* (*CMV*), which causes one of the most serious viral diseases of tomatoes worldwide (4). Four biological conditions were set up: control plants (C), CMV-infected plants (V), mycorrhizal plants (M) and CMV-infected mycorrhizal plants (MV). At the time of analysis (28 dpi - days post infection), the mycorrhization level was not significantly modified by virus infection, indicating that the AM colonization was unaffected by the presence and spread of the virus. On the contrary, in MV plants viral symptoms were milder than in V plants at 14 dpi while no difference was found at 28 dpi. At the two time points the concentration of viral RNA was similar in shoots of V and MV plants. Physiological analyses showed that in virus-infected plants the AM symbiosis improved plant performances in terms of stomatal conductance and photosynthetic CO_2 assimilation pointing to a positive role in water use efficiency of the host plant. Genome-wide transcriptomic data obtained by the RNASeq technique are currently under analysis. In their whole the results highlight a systemic effect of the AM symbiosis which alters the responses to the viral infection.

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1.3 = ASSESSMENT OF SOIL QUALITY OF DIFFERENTLY MANAGED ENVIRONMENTS BY CO_2 AND FUNGAL COUNT

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The quality of a specific kind of soil indicates the measure of its capacity to sustain plant and animal productivity, maintain or enhance water and air quality, support human health and habitation within natural or managed ecosystems. A soil with high quality can accomplish its main functions: sustaining biological activity, diversity, and productivity, regulating the water and solute flow; filtering and buffering, degrading, immobilizing, and detoxifying organic and inorganic materials, storing and cycling nutrients and other elements (1). An integrated method for assessing multiple aspects of the soil and their connections leads to better understanding of the grade of its quality. One of the most important aspects that must be taken into account is soil respiration that can be measured as CO_2 production. Soil respiration is an important component of the carbon dynamics of the terrestrial ecosystems (2) and most of the CO_2 production is associated with heterotrophic respiration from fungal and bacterial communities using recently produced organic material as an energy substrate (3). The main object of this research is to measure the CO_2 production of soil differently managed and to evaluate the fungal component as sensitive parameters of the soil functionality.Two study areas were considered, both sited in Lombardy Region (Italy):

- a grassland system with unmanaged (control), replanting and ploughed plots (9 in total); the CO_2 flux ($\mu mol*m^{-2}*s^{-1}$) were measured with CO_2 portable analyser (ADCPro-sd with soil camber)
- 6 plots differently managed, 4 of them for a renaturation process, 1 control plot represented by high disturbed field recently dug-up, and 1 plot in the Natural Reserve "Bosco Siro Negri" representing the reference potential forest condition. The CO2 production was measured by the Solvita Soil-life test (http://www.themeterman.com.au/solvita_soillife.php).

CO_2 production and Colony forming units per g of soil dry weight were analyzed in each area. Non-Parametric MANOVA (PERMANOVA) was applied on the CO_2 data and showed that there are significant differences ($p:<0.001$) between the different plots and methods of management. A similar trend of the CO_2 flux was showed by the mycological data. The soil of the integral natural reserve showed the highest levels of respiration rate and fungal abundance. Analysis based on the results obtained highlighted that CO_2 and fungal abundance are sensitive descriptors of soil condition and they can be helpful to pick out the right management practice and to monitor renaturation processes.

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1.3 = PROTECTING BEECH FORESTS AND ENHANCING ECOSYSTEM SERVICES: THE ROLE OF FUNGAL BIODIVERSITY IN APENNINE PRIORITY HABITATS

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The FAGUS LIFE Project (LIFE11/NAT/IT/135) targets two European priority habitats, i.e. Habitat 9210* Apennine beech forests with *Taxus* and *Ilex*, and Habitat 9220* Apennine beech forests with *Abies alba*, within two National Parks: Cilento, Vallo di Diano and Alburni, Gran Sasso and Monti della Laga. These habitats have a limited distribution due to the long-term impact of human activities, such as harvesting and grazing, which simplified the structure and composition of these ecosystems. Conventional management in these forests impacts a wealth of taxonomic groups, such as saproxylic beetles and fungi, which are threatened throughout Europe by the lack of deadwood and of senescing trees, and by the homogeneous structure of managed forests. Biodiversity plays a key role in providing desired rates of multiple ecosystem services (e.g., timber harvest, carbon storage, soil and drinking water supply protection, non-woody forest products), and thus synergies are expected between the provision of such services and conservation efforts. With this regard, fungi play a prominent role in several ecological processes, creating the basis for the cycling of photosynthetic energy, carbon, and nutrients stored in woody material. The FAGUS project aims at developing and testing management strategies able to integrate the conservation of priority forest habitats (9210* and 9220*) and the sustainable use of forest resources for multifunctional purposes. In order to assess the responses to different management treatments a BACI monitoring design (Before-After, Control-Intervention) has been applied on forest structure and diversity of focus taxa before and after experimental harvesting treatments. The before-intervention field sampling was carried out in autumn 2013 in 33 monitoring plots across the two National Parks. The occurrence at plot level of both saproxylic and epigeous *Ascomycota* and *Basidiomycota* sporocarps was surveyed. All standing and downed deadwood with a minimum diameter of 10 cm was sampled for sporocarps larger than 1 mm, and information on decay class and fungal morphogroups was recorded. We identified species of high scientific concern, in both National Parks, specifically 64 species of saproxylic fungi, with 35 exclusive species in Cilento and 52 species with 23 exclusive species in Gran Sasso. As regard epigeous fungi 47 species with 26 exclusive species in Cilento and 42 species with 20 exclusive species in Gran Sasso were recorded. Within the "Gran Sasso and Monti della Laga National Park", the area of Incodara is of special interest due to the occurrence of the species *Ossicaulis lignatilis*, which is among the 21 identified indicator species for assessing conservation value of beech forests in Europe. Cilento has an alpha and gamma diversity higher than Gran Sasso, both for saproxylic and epigeous fungi. Species composition was visualized using Non-metric Multidimensional Scaling (NMDS), based of Jaccard dissimilarity. The results show a clear separation between Cilento and Gran Sasso NP at least for saproxylic fungi probably in relation to the topografical position of the plots. The dendrograms from hierarchical cluster analysis of epigeous and saproxylic fungi for both National Parks respectively illustrate a different arrangement of the clusters; in fact, the plots resulted associated in different ways the case of epigeous in respect to saproxylic fungi. The results of this study shed light on the management measures required to ensure the survival of wood decay fungal communities. Substrate heterogeneity is achieved, at both the stand and landscape level, over time by maintaining and enhancing ecosystem services and ecological continuity in beech forest ecosystems; this ensures the presence of multiple habitats and microhabitats for fungi.

1.3 = ARE FUNGAL COMMUNITIES DECOMPOSING CELLULOSE RANDOMLY ASSEMBLED THROUGH DISPERSAL AND PRIORITY EFFECTS?

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The overall response of all ecosystem functions (ecosystem multifunctionality) showed a positive correlation to soil biodiversity, thus confirming that soil community composition is a key factor in regulating ecosystem functioning [1]. Simplification of soil community composition can hypothetically impair some ecosystem functions, including decomposition and nutrient cycling. The understanding of the mechanisms that govern species distribution and dispersion patterns in soil is thus fundamental to explain the role of some functional groups in ecosystem functioning. Fungi have a great role in soil carbon cycling, plant nutrition, and pathology, they are widely distributed in all terrestrial ecosystems. Besides their diversity and importance, the distribution of species, phyla, and functional groups has been poorly documented. Some fungi seemed to follow similar biogeographic patterns as plants and animals, but other taxonomic and functional groups showed to run counter to overall patterns [2]. Thus some *taxa* can theoretically be everywhere, but apparently only a few small number of them can exert a huge influence on particular environments. These species are supposed to act as keystone species, namely organisms that play a unique and crucial role in the way an ecosystem functions. Some key-stone and widely distributed species could have their force in the fact that they behave as pioneer species on particular substrates (like lignin, cellulose, collagen, etc.). Actually several studies showed that micro-habitats and substrates are of considerable significance in determining fungal species' distribution [2] and for some authors (quite in contrast with what showed by metabarcoding studies), local differences in distribution are greater than continental differences. This means that the scale at which fungal ecology concepts are tested against environmental sampling is determinant, and the comprehension of patchiness in fungal partitioning, competition and variability is fundamental. The aim of this study was to test two opposed niche theories in the colonisation of pure cellulose patches by fungi. Classical niche theory [3] predicts that species assemblages will be structured by the ability of individual species to persist in a given set of abiotic conditions, while successfully competing with other species for resources and space. Neutral theory [4] predicts that assemblages will be structured by stochastic processes such as initial dispersal into an area. In other words, are fungal communities decomposing cellulose randomly assembled through dispersal and priority effects, or do species interact with each other leading to positive and negative associations? The two hypothesis were tested with cellulolytic fungi growing in a model natural environment using both culturing techniques, direct microscopic observation and a functional metabolic approach. Co-occurrence and exclusion between fungal species, pioneer role of some *taxa* and patchiness of species distribution were considered. In particular the relationship between the potential activity (metabolic profiles) shown by pure fungal isolates (in vitro) and their ecological role in the field was evaluated according to the functional approach (Phenotype Microarray, Biolog^{TM}) used in Di Lonardo et al. [5]. What emerged was that at an early stage of cellulose colonization, the fungal species occupy different spatial niches on cellulosic resources, and it is present a potential overlap in the overall metabolic function. Moreover, a sort of founder effect could be hypothesised for microfungi when they colonise sterile cellulose, suggesting a strong influence of the scale, in the causative mechanisms that link genetic fungal diversity to specific ecosystem functions and, at a broader scale, to ecosystem multifunctionality.

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1.4 = PRELIMINARY STUDY IN OPEN RACEWAY FOR PRODUCTION OF *SPIRULINA* BIOMASS IN SOUTHERN ITALY. FIRST EXPERIENCE IN CALABRIA

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Arthrospira platensis (Spirulina) is an economically important filamentous cyanobacterium that is commercially produced as a source of human health food, animal feed, and cosmetic colorants. (1). Extensive studies on improving its growing condition and cultivation have been carried out (2,3). Nowadays there are no literature data, which report experiment in Calabria. Therefore, to our knowledge this paper is the first report for Calabria. We did a preliminary evaluation of the growth characteristics of *Arthrospira platensis'* strain in open-raceway in order to evaluate the potential economic impact. *A. platensis* used in this study was collected from "Provence Spirulina" farm in the south of France and maintained in Zarrouk's medium (pH 9) under rotatory conditions and illuminated continuously at a light intensity of 3200 LUX (4). When the culture was at the stationary phase of growth, a 20 l bottle was used to start to scale-up process. We started with experiment when the scale-up process achieved a volume of 250 l (fig. 1). We evaluated the growth in open raceway from April 2014 to March 2015. Every 10 days the biomass in culture was harvested and renewed for 50 percent of volume medium. We calculated correlation between weight and volume as g/L and improved microscopic examination of sample using light microscope. The growth curve of *A. platensis* represents a typical microbe growth curve. It shows a lag, log, and stationary phase, where the log phase is faster in summer than in winter months. The total yield of *A. platensis* biomass was 0.98g/L (245g/250 l) harvested after about 240 h. From literature data, the optimum temperature for the growth of *A. platensis* is around 36 °C (5). Thus, the production in open raceway is limited by temperature. The production is favorite at the lower latitudes, but limited at higher. Therefore, the temperature is the most important environmental parameter that limit the growth of *A. platensis* in open raceway. However, in south Calabria the average temperature allow the growth in open raceway as our data shown). The national investments in green economy improved in last years (6). Favorable environmental conditions and strategic location of south Calabria might draw more funding in the green enterprise.

Fig. 1 Pilot scale raceway

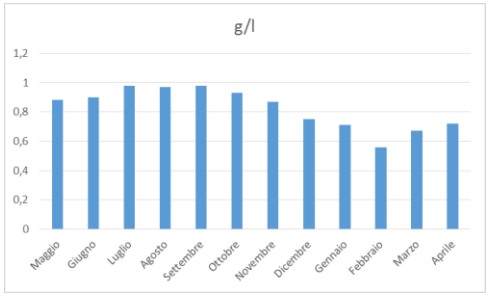

Fig. 2 Data shown growth in open raceway

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1.6 = POLLINATION OF TWO TOMATO CULTIVARS IN UNHEATED GREEHOUSES: FROM POLLEN TRANSFER TO FRUIT DEVELOPMENT AND BUMBLEBEES FORAGING ACTIVITY

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The purpose of this study is to determine causes of inter-annual yield variability in tomatoes grown in unheated greenhouses. Certainly, yield reduction is due to mid-winter period and to sudden change in temperatures. Nevertheless, it is unclear which are keys factors affecting production and if they are linked with pollination quality or pollen viability. For this reason all parameters related with pollination were considered in the present study.
Two common tomato cultivar that have shown changes in production in previous years, were chosen for the present study: "fragolino" and "cupido" variety; experiments were carried out in 10 greenhouses, five for each cultivar.
Experiments were conducted as follows:
1) In pollination experiments natural pollination performed by commercial bumblebees were compared to hand pollination. Thereafter, after ripening, fruit were counted, size measured and these parameters were compared in the two pollination test in order to assess pollination efficiency.
2) At the same time bumblebees foraging activity was recorded through standardized counting of flower in order to assess the number of visits. In addition flowers with necrotic discoloration in anthers cones, which are result of buzzing pollination by bumblebees, were counted. Bruising level were included in five categories (0-4).
3) Pollen germination was also considered and experiment where carried out throughout the production period (September-June).
The study is ongoing and preliminary results shows:
1) Pollination experiments: the flowers proportion that reached fruit maturity was compared in hand ad natural pollination. In three out of ten greenhouses pollination has not been optimal ($p=0.013$; $p=0.003$; $p=0.021$) while in the other seven there were no significant difference between hand and natural pollination ($p>0.05$). Fruit size was compared in hand and natural pollination. This experiment highlights the relationship between fruit size and pollination efficiency. Preliminary results are contrasting in the two cultivar. "Fragolino" shows in three out of five greenhouses that hand pollination has produced fruit with significant larger size ($p<0.0001$ in two; $p=0.034$), while in "Cupido" there was not difference with one exception of larger fruit in natural pollination ($p<0.001$). For the statistical procedure was used " t – test".
2) Bumblebee foraging activity: the number of flower visited and necrotic discoloration level were compared both to fruit development and size. Preliminary results show no relation between the percentage of bruised flowers and the number of fruits ($p=0.353$) as well as diameter ($p=0.42$). Result changes while high degree of necrotic discoloration occurs ($p=0.041$) highlighting the importance of a high bumblebee activity in greenhouses. For the statistical procedure linear regression was used.
3) Pollen germination analysis are ongoing.
Preliminary results shows how an inconstant or low efficient bumblebees pollination can affect the yield in some cases, while an efficient pollination can increase production both in terms of number of fruit and their size.

Acknowledgements
This communication has been produced as part of the research project "Pollination of two tomato cultivar in unheated greenhouses: from pollen transfer to fruit development and bumblebees foraging activity" developed at the "Cooperative Santa Margherita Terra e Sole, Pula, Cagliari, Italy", by a research grant financed from the resources P.O.R. SARDEGNA F.S.E. 2007-2013 - "Obiettivo competitività regionale e occupazione, Asse IV Capitale umano, Linee di Attività I.1.1. e I.3.1."

1.8 = PRELIMINARY GEOMYCOLOGICAL INVESTIGATIONS IN THE LIBIOLA MINE (LIGURIA, NW ITALY)

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Contamination of soil by heavy metals through industrial and human activities may lead to high concentrations of toxic metals (1). In particular, sulphide mines represent hazardous and dynamic environments. In fact, metals pollution continues even after the mine activity closed (2). This is due to the reactions between rain waters and waste rock dumps sulphide ores. The process, called Acid Mine Drainage (AMD), leads to the soil nutrients loss, causes its acidification (pH $\leq$ 4), and determines ecotoxic metals accumulation in soils (3). Hence, pioneer microorganisms such as fungi may play a central role to remediate these polluted soils and promote their plant recolonization, reactivating biogeochemical cycles (4, 5, 6).
In this preliminary work, we studied the fungal diversity and the potential interactions between a *Trichoderma harzianum* Rifai strain and dump soil in the Libiola mine (Liguria, NW Italy). Libiola is a derelict Cu-Fe sulphide mine subjected to AMD processes. In particular, previous studies about its waste rock dumps showed physic, chemical, and edaphic critical conditions (2). Libiola sulphide mineralizations are associated with basalt rocks (basalts, basaltic pillows, and basaltic breccias), and serpentinites (2).
Libiola dump soil samples were collected and some fungal strains were isolated by the Gams modified dilution plate technique (7). Later they were identified by macro-micromorphological method and DNA molecular analysis. A number of strains belong to the *Penicillium* genus, but also *Trichoderma*, *Cladosporium,* and *Aspergillus* genera. *Trichoderma harzianum* resulted one of the most frequent among the isolated species. Moreover, previous studies showed its Cu bioaccumulation capability and high tolerance (6), so this native strain was later selected and its growing capability in a media including sample of Libiola soil was tested to verify the possible and potential interactions among this fungal strain, rocks, and minerals. Four flasks containing Libiola dumps soil were autoclaved and then inoculated with fungal solution. Then, the flasks were stored in the dark at 24° C for 3 months. After this period, the major of soil pebbles resulted completely covered by *T. harzianum* mycelia. Some serpentinitic and basaltic pebbles and some mycelia portions were analysed by a stereomicroscope and scanning electron microscopy (SEM) and energy dispersive X-ray analyzer (EDXA). The results highlighted the actual and close interactions among this native fungal strain and both sulphide mineralizations and host rocks. In fact, the analysis of the basaltic pebbles covered by mycelium showed the presence of calcium sulphate minerals wrapped by *T. harzianum* hyphae and conidia.
The study of fungal and soil interactions may allow understanding the role played by fungi in the Libiola mine soils remediation. However, other studies should be conducted to prove the effective interactions and understand how to take advantage of them.

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1.8 = ISOLATION OF BLACK FUNGI FROM CONTAMINATED SITES: NEW STRAINS WITH BIOREMEDIATION POTENTIAL

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Aromatic compounds represent a serious problem for the public health for their toxic and carcinogenic potential. They are extensively used in industrial processes and are also present in fuels, therefore may be easily released in the environment and cause severe soil and water contaminations.
During the last decade, bioremediation has become an innovative technology for removal of xenobiotics from the environment and fungi, because of their wide degradative abilities, are successfully used both in bioremediation [1; 2] and biofiltration [3] processes representing thus a useful tool in environment protection.
The ability to metabolize aromatics has been already proved in a restricted group of black yeasts belonging to the family *Herpotrichiellaceae* in the order *Chaetothyriales* [4,5].
In the present study we sampled habitats never studied before for searching new black fungal strains able to mineralize aromatic substrates; selective growth conditions, such low temperatures and naphthalene-based substrata, were applied in order to prevent the development of fast growing cosmopolitan fungi and select strains with potentialities in degrading aromates.
80 black fungal strains were isolated from car fuel tanks and pumps (gasoline and diesel); along with some recurrent species, such as *Exophiala xenobiotica* and *Exophiala crusticola*, revealed also the presence of few isolates belonging to a possible new genus and species next to *Ochroconis*.

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1.8 = SEED GERMINATION UNDER NICKEL STRESS IN A FACULTATIVE HYPERACCUMULATOR

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'Facultative hyperaccumulators' (1) have been documented to hyperaccumulate metals when occurring on metalliferous soils, including serpentine, but can also be found outside those metal-rich areas (2). It is well known that high nickel (Ni) levels exert toxic effects on seed germination (3), but literature investigating the same on hyperaccumulators is very few (e.g. 4). As a consequence, we investigated the effect of native soils with high or low Ni levels on seed germination of *Alyssoides utriculata* (L.) Medik. (Brassicaceae), a new Ni hyperaccumulator (5, 6), distributed in the North-eastern Mediterranean area on serpentine and non-serpentine sites.
Seeds of *A. utriculata* were collected from 3 populations in Liguria (NW Italy) on different Ni-rich soils (A, serpentine; B, serpentine with Fe and Cu contamination; C, non-serpentine), according to international guidelines (7, 8). Native soils were also sampled, dried at 60°C 48h and sieved at 4 mm; neutral growing substrate and vermiculite were used as control for comparison. Experiments were replicated in microcosm (growing chamber, 16 h d^{-1}, T=20°C±1 in Petri dishes, 2 weeks, n=400) and in mesocosm under drought stress (greenhouse, natural photoperiod, T=20°C±4 in plant trays, 8 weeks, n=3000). Soils were autoclaved to assure basic sterility. Seeds from A, B and C populations were sown on each soil type, both in microcosm and in mesocosm.
In microcosm, the germination index (GI) is significantly high (GI>>50%, $p<0.05$) in A, B and C populations after 2 weeks, with the C population less viable. Post-hoc test showed no significant difference in GI for all times, for all soils and for their combinations. In mesocosm, on the contrary, GI<50% under both Ni stress and drought stress in all populations, although GI had a significant positive correlation with time ($r>0,039$; $p<0.05$). Post-hoc test showed no significant effect in relations with A, B, and C soils.
In addition, all plantlets grown on A, and B substrate gave a positive reaction to DMG test for Ni detection (9), suggesting that Ni-hyperaccumulation is a species and not a population trait. Our results support the hypothesis that hyperaccumulation is genetically determined in the species.
The results suggest the use of this species to develop *in-situ* phytoremediation and habitat restoration avoiding ecosystem disruption and minimising interventions and costs in the Mediterranean habitat.

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1.8 = PRELIMINARY CHARACTERIZATION OF FUNGAL ENDOPHYTES FROM *NEPHROLEPIS CORDIFOLIA* (L.) C.PRESL AND THEIR ROLE IN FERN TOLERANCE TO INORGANIC POLLUTANTS

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Nephrolepis cordifolia is a fern worldwide diffused as an ornamental plant, but its biological and ecophysiological characteristics have so far been poorly studied. Even if its resistance to biotic and abiotic stress lacks consolidated support, parasite attacks have been occasionally observed (1). On the basis of a) antimicrobial substances production (2), b) ability to accumulate heavy metals (3,4,5), and c) tolerance to some rare earth elements (6), *N. cordifolia* had been proposed as a possible candidate for phytoremediation in polluted environments (4). The presence of microbial endophytes, in either roots end rhizomes, possibly helping accumulation of inorganic pollutants in plants, was reported (7,8). The aim of this research was the isolation and identification of fungal endophytes obtained from *N. cordifolia* collected in different sites in southern Italy. Preliminary *in vitro* studies on responses of *Trichoderma harzianum* Rifai associated with *N. cordifolia* to some pollutants were also performed. The results confirm that *T. harzianum* acts as an endophyte in tubers and roots of *N. cordifolia* populations grown in different locations (Bari, Taranto, Marina di Leporano (TA), Montalbano Jonico (MT), and Portici (NA)). *T. harzianum* strain Th1, isolated from *N. cordifolia* populations grown in Bari, was exposed in either liquid and solid media to $NaAsO_2$, $Ce(NO_3)_3$, and $Pb(NO_3)_2$ at concentrations ranging from 0.01 to- 10 mM. A dose-dependent effect was observed. The Th1 strain tolerates each pollutant at concentrations up to 1 mM, displaying a biomass increase, while it was completely inhibited at 10 mM concentration. A possible role of the fungal endophytes in the tolerance of *N. cordifolia* to tested metals is hypothesized.

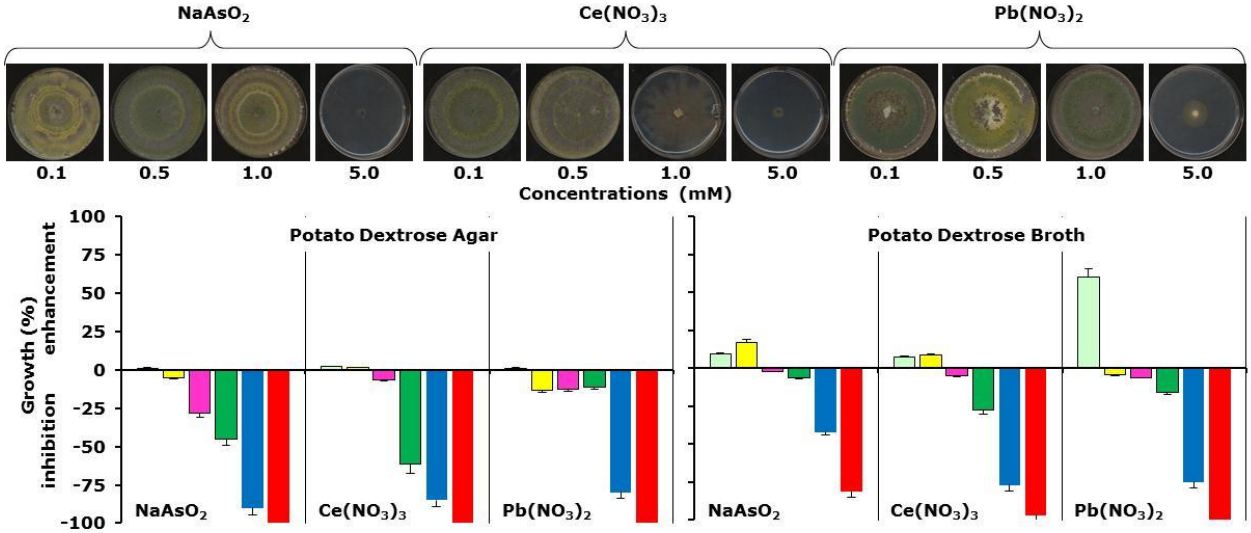

Fig. 1 Effect of $NaAsO_2$, $Ce(NO_3)_3$, or $Pb(NO_3)_2$ assayed at 0.01 (■),0.1 (■), 0.5 (■); 1 (■), 5 (■) or 10 (■) mM on *Trichoderma harzianum* strain Th1 grown in Petri dishes on Potato Dextrose Agar or in liquid Potato Dextrose Broth.

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1.9 = A CASE OF ECOLOGICAL RENATURATION IN A DRAINED MEDITERRANEAN PEATLAND: THE CASE STUDY OF THE MASSACIUCCOLI LAKE BASIN (TUSCANY, IT)

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The Massaciuccoli Lake floodplain is located in the Natural Park of San Rossore, Migliarino and Massaciuccoli in NW of Tuscany (Italy). Since the 1930s, a large part of the Massaciuccoli floodplain has been drained for agricultural purposes. To ensure a water table depth suitable for cultivation, a complex network of artificial drains and pumping stations has been used to drain the superficial aquifer and rainwater. Land use is characterised by conventional agriculture (covers 80% of the area) and periurban infrastructures, such as a wastewater treatment plant. In the peatland area, cropping systems are based on continuous production of maize (*Zea mays* L.), sunflower (*Heliantus annuus* L.), wheat (*Triticum spp*. L.). As a consequence of land use, several environmental concerns arose in the last 50 years. The most important concerns are those related to:
I. eutrophication of the lake due to nutrient enrichment (N, P) in the surface- and groundwater.
II. the subsidence rate (2-3 m in 70 years) due to compaction and increased mineralization of peat.
The project RestoMedPeatland (https://sites.google.com/site/restomedpeatland/) started in 2011, identified rewetting and setting-up a phyto-treatment system as the solution for improving water quality,slowing down soil organic matter (SOM) mineralization, and, therefore, a method to restore the ecological functions of this site. A 15 ha experimental area was used to compare the efficiency of three different systems (A: constructed wetland, B: paludiculture system and C: natural wetland) (Fig.1) in treating the eutrophic drainage water coming from a sub-watershed in the reclamation district. Quantity and quality of water was monitored both at the entrance and the exit of each system.

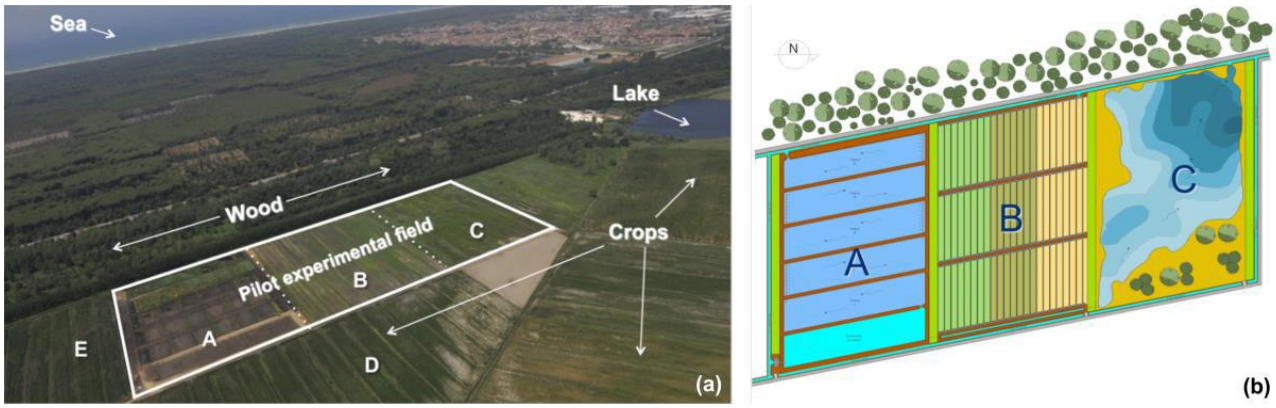

Fig. 1 Areal view (a) and set-up (b) of the pilot experiment with three different management systems: constructed wetland (A), paludiculture system (B) and natural wetland (C).

In the natural wetland (C), after top soil removal, excavation and rewetting with drainage water, the vegetation has evolved naturally. The surveys carried out during the four years after pilot experimental field, highlighted the development of a spontaneous hydro-hygrophilous vegetation. This was mainly composed of helophytic communities dominated by *Phragmites australis* (Cav.) Trin. and *Typha latifolia* L., in addition to monophytic populations of *Myriophillum* spp., occupying different areas of the system according to the depth of excavation, for about 50% of the whole surface. The flora, currently, consists more than 30 species, as hydro-hygrophilous *Alisma plantago-aquatica* L., *Juncus effusus* L., *Apium nodiflorum* (L.) Lag, *Lemna minor* L., characteristic of the nearby Massaciuccoli Lake. The data collected so far showed a high and progressive growth of the biomass of the natural area, a dynamic upward trend of floristic-vegetation biodiversity, and, from a technical standpoint, a large phyto-treatment capability.

1.9 = BIOINFORMATIC AND MOLECULAR TOOLS TO IMPROVE BIOREMEDIATION OF POLLUTED AIR AT INDUSTRIAL SITES

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The aromatic hydrocarbon benzene and other non-methane volatile organic compounds (NMVOCs) represent a major source of environmental pollution and the main targets in bioremediation studies (1). Bioremediation appears as the most cost-effective technique for treating industrial polluted gaseous streams containing organic compounds and remediation of polluted air is generally performed using conventional biofiltration techniques (2). Traditional biofilters do not represent suitable tools for the treatment of hydrocarbon-contaminated air since the biofilter beds are often subjected to clogging due to biomass overgrowth while the rotating biofilter reactor-based technology, e.g. 'Biowheel 2.0', represents nowadays a promising alternative (3). The bioremediation performance of the 'Biowheel 2.0' rotating biofilter relies on the activity of microbial consortia which carry out pollutant degradation at extremely high efficiency.
To date, plant inoculation with microorganisms having high potential for pollutant degradation combined with plant growth-promoting ability represents a promising strategy for phytoremediation purposes. The present investigation deals with the microbiological and molecular characterisation of bacterial isolates provided by BMB Technologies & Services srl, as putative candidates for future phytoremediation strategies.

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1.9 = GENETICS OF *ANADENANTHERA COLUBRINA* VAR. *CEBIL* (FABACEAE), A TREE FROM SALTA (NORTHWESTERN ARGENTINA)

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Anadenanthera colubrina var. *cebil* (Vell.) Brenan belongs to the Fabaceae/Leguminosae family, Mimosoideae subfamily. This tree species has economic, medicinal and cultural applications in South America. It grows in Brazil, Paraguay, Peru, Bolivia and Argentina.

The wood contains tannins, is hard and resistant to termites and is used for wood strcture and furniture, as poles and for firewood. In popular medicine, it is used to treat respiratory problems and inflammations. It is considered a sacred tree by local cultures. The seeds have been used in rituals and popular medicine, as they contain alkaloids derived from dimethyl tryptamine, by shamans for over 3000 years (1).

Studies carried out on wild plant species have revealed that the distribution of morphological and chemical characteristics present patterns that can be related to geographic regions (2). These variations may reflect the phenotypic plasticity of the individuals, genetic adaptations of the populations to different environments or both (3).

The aim of this work is to study the morphological descriptors and the genetic diversity of *A. colubrina* var. *cebil*, collected from different locations over its distribution area in Salta Province (North Argentina) and conserved in the Germplasm Bank of Native Species (BGEN-INEAH) of the National University of Salta.

As demonstrated in previous results (4), the analysis of the morphological traits of the fruits showed that the most distant populations were Metán and El Cebilar (Euclidean distance 4.49), whereas San Bernardo and El Gallinato were closer. In addition, these two populations were more distant from Metán than from El Cebilar. The distribution referred on weight of seeds, were normal in El Cebilar and El Gallinato, whereas in San Bernardo and Metán seed weights were skewed and negative leptokurtic. Seed weight varied between populations and was significantly higher in San Bernardo, lower in El Cebilar and intermediate in El Gallinato and Metán, similar to the reported distributions in fruits. The other morphological descriptors (width, length and thickness) varied between populations. Then, we investigated the phylogenetic relationships among the ITS-rDNA regions of four different individuals from the same species of *A. colubrina* var. *cebil*. The highest similarity of nucleotide sequence was found between San Bernardo and Metán, and between El Cebilar and El Gallinato, in agreement with seed descriptors analysis; outgroup is the Brazilian *A. colubrina* (AcBr).

In this work, we analyzed the ITS rDNA nucleotidic sequences of 29 individuals collected from three of the four sites.

Fig. 1 *Anadenathera colubrina*.

Fig. 2 e 3 Flower and seeds.

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1.9 = DENDROENERGETIC PLANTS OF THE COLOMBIAN AMAZONIA: THEIR USE AND MANAGEMENT

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Dendroenergetic plants are woody species used as fuel and are considered as the main energy source by a large part of the world population, especially the rural one, due to their availability and low cost (1). In this study the most used dendroenergetic species in the Colombian Amazonia, are presented and discussed. We analyzed their supply, use, and management in the tradition of the indigenous community Piapoco from Laguna Colorada (Gianía, Colombia). The review of the biogeographic and sociocultural features of the study area was carried out as well as the ethnobotanical aspects related to firewood. Indirect and direct surveys (2002) regarding home features, charcoal knowledge, use and management of dendroenergetic plants, transmission way of this knowledge were recorded during the field research work. Moreover, a group of informants (162 family groups) was chosen in order to be interviewed. Area, distance and direction of firewood collection were recorded with the collaboration of people. Likewise pictures of firewood use, management and consumption were collected together with botanic and ethnobotanic data of all dendroenergetic species used. Samples of each species were dried and labeled and kept in the Universidad Pedagógica y Tecnológica de Colombia (UPTC) Herbarium, Colombian National Herbarium (COL) and Colombian Amazonian Herbarium (COAH). As a result, an informative catalogue was created. Inhabitants from Laguna Colorada get the forest resource from four habitats: conuco, stubble, high mountain and banks. Conuco refers to the vegetation left in a place as a consequence of slash and burn system for agricultural production. Stubble is the land that hosted conuco and has been abandoned to let the soil rest and making possible the recovering of vegetation. Regarding the complementary sources, high mountain includes vegetation areas which have been subject to long time regrowth after being cut down. Banks deal with of lagoon surrounding areas in which plant-based fuel resources increase during the dry seasons. 95 dendroenergetic plant species were recorded and catalogued. The most represented families are *Myrtaceae* (11%) with 4 genera and 10 species, *Moraceae* (7%) with 5 genera and 7 species, *Annonaceae* (6%) with 4 genera and 6 species and *Mimosaceae* (6%) with 3 genera and 6 species. The most frequently used species are *Nectandra cuspidata* Nees. & Mart., *Inga* sp. *Theobroma subincanum* Mart. and *Bocageopsis multiflora* (Mart.) R. E. Fr.. It was observed that the mostly used firewood species correspond to those used ten years before (1992). However, some of them are not used anymore such as *Fourcraea* sp. and the use of *Alchornea fluviatilis* Secco, *Eugenia micrantha* (Kunth) DC., *Inga* sp. and *Triplaris surinamensis* Cham. is now reduced. Piapoco indigenous people bear in mind the firewood quality, the species abundance and their distribution in the gathering areas. Most of the dendroenergetic species used in Laguna Colorada are catalogued as excellent or highly qualified. Firewood enables to light, to heat and to produce coal or ashes. Preferred lighting species are: *Nectandra cuspidata*, *Vismia ferruginea* Kunth, *Alchornea fluviatilis* and *Cochlospermum orinocense* (Kunth) Steud. Nonetheless, being soft wood, they are considered low quality firewood species due to their little density and fast combustion. Heating species are *Eugenia micrantha*, *Inga* sp. and *Eschweilera* sp.. They are catalogued as excellent or highly qualified firewood. Despite being hard wood, difficult to be cut down and carried, they guarantee a prolonged combustion. *Licania* sp. is well-known for ashes production that can be used for handicraft. Firewood gathering, in the indigenous community from Laguna Colorada, is carried out by slash and burn system to foster conuco cultivation. This process implies the selective use of secondary vegetation which is one of the most important sustainable management strategies. The studied Piapoco community recognizes and differentiates various uses and properties of dendroenergetic species. This indicates the richness and complexity of firewood knowledge and management which depends not only on the environment, but also on the cultural and social contexts.
Indigenous people from Laguna Colorada choose the firewood according to its availability, the diversity of sources and combustible species. This contributes to the vegetation regeneration processes. However, it is necessary to inquire into the low availability of some plant species such as *Vasivaea* sp. If the use of firewood is based on diversity and preservation of ancestral traditions, it implies the biological richness conservation. The use of firewood in Laguna Colorada is an epitome of integral management of resources inherited by Piapoco ethnic group and represents a relevant cultural patrimony that has enabled them to support themselves for ages.

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1.10 = WATER USE IN THREE *SEDUM* SPECIES UNDER DROUGHT CONDITIONS IN GREEN ROOF SUBSTRATES AND THEIR ROLE IN GREEN ROOF PERFORMANCES

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One of the main goal of the green infrastructures is to mitigate the urban heat island effect. Green roofs, particular rooftops that include substrate layer and plants, could have a big role in this because of the cooling effect (1) due to plants evapotranspiration. In Mediterranean climate, with high summer temperature and scarce precipitations (2), the green roofs are characterised by hard environmental conditions. Crassulacean species are the most utilized in such stressing environments because are drought resistant CAM species. CAM (Crassulacean Acid Metabolism) is a water-saving way that succulent species use to fix carbon dioxide (CO_2). In particular, CAM species reduced their daily transpiration. In this study we utilized *Sedum lydium* Boiss, *Sedum album* L. and *Sedum kamtschaticum* Fisch. They are CAM facultative, so they can switch between C3 photosynthesis and CAM to respond to particular environmental conditions, for instance low water availability (1). The aim of this study is to better understand the water use of these three species and how a low transpiration can influence the green roof cooling effect due to evapotranspiration. For the experiment we utilized 34 pots filled with 500 g of green roof's substrate (Harpo/SEIC substrate); 10 pots for each species, 4 pots were with substrate-only to determine the water loss due to evaporation. Plants grew in greenhouse at 16/28°C night/day. The experiment lasted 31 days; the first and the 16th day of the experiment, pots were irrigated until their field capacity. For each pot with plants we measured: 1) daily evapotranspiration rate calculated following gravimetric method (1); 2) leaf RWC (Relative Water Content) following (3), from the 15th day, every three days. For each pot with substrate-only we measured daily substrate evaporation rate following gravimetric method (1). Moreover, we calculated relative humidity (RH) of substrate ((wet soil – dry soil)/dry soil)*100) of plants' pots. After 16 days, pots with plants showed a higher loss of water compare to substrate-only pots. Plants lost on average the 99,6% of water, whereas the substrate–only pots, lost 91,7%. However, from the 9th day until the end of experiment, we noticed a slowdown in total loss of water in the pots with plants, they lost on average 13,8% of the total water, while substrate-only pots lost 22%. Regarding the species behaviour, *S. kamtschaticum*' pots began to lose more water than substrate-only one day before (the 8th day). After the second irrigation**,** the evapotranspiration rate showed a particular trend: from the 17th to the 20th day, plants' pots lost less water (on average 10,5%) than substrate-only pots. Then, evapotranspiration of plants' pots was higher than evaporation of substrate-only pots. After the 27th day, again, we noticed a slowdown in total loss of water in the pots with plants. RWC data showed an increase from the 17th to the 23th day. From the 27th day to the end of the experiment, RWC decrease. These data allow us to hypothesize the behaviour of the three *Sedum* species in green roof's substrate and under water stress. We hypothesize that at the beginning of the experiment the three species were doing C3 photosynthesis. After the irrigation, RWC data showed an increase of water in the leaves and at the same time the evapotranspiration rate decrease**.** Probably, stomata were still close and plants held water inside their leaves. From this moment until 27th day, the three species restart to behave as at the beginning of the experiment**,** showing an evapotranspiration rate higher than the evaporation of substrate alone**.** We could identify, for every species, the days of the beginning of the two slowdown's periods: the 9th and the 28th day. Finally, we calculated the average RH of soil at the 9th day for *S. lydium*'s and *S. album'*s pots, at the 8th for *S. kamtschaticum*'s pots and also at the 28th day for every plants' pots. Looking closer we notice that *S. kamtschaticum* lose more water than other *Sedum* species after the two irrigations. This is confirmed by RH data. These show that the average RH of soil obtained at the 8th day for *S. kamtschaticum*' pots are statistically equal to *S. album* and *S. lydium* data obtained at the 9th day (the average data was 5,2%). *S. kamtschaticum* has a well water use efficiency (among of carbon fixed relative to water transpiration) with high soil moisture (1). In general, all three species show a high plasticity regarding the use of water, maybe this could be also correlated to a shift from C3 photosynthesis to CAM metabolism. When the RH of soil is scarce (around to 5,2%), every species limited transpiration and lost less water respect substrate only. This has a negative influence on green roof's cooling effect. However, when water is available, green roof with plants lose more water than substrate-only because of transpiration. Thus, a green roof with these species needs some water to guarantee a cooling effect higher than a roof top with only substrate layer.

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1.10 = URBAN AND PERI-URBAN FORESTS IN THE METROPOLITAN AREA OF ROME: ECOPHYSIOLOGICAL RESPONSE OF *QUERCUS ILEX* L. IN TWO GREEN INFRASTRUCTURES, IN AN ECOSYSTEM SERVICES PERSPECTIVE.

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Green Infrastructures (GIs), such as urban forests, deliver important benefits for humans, collectively known as Ecosystem Services (ESs) (1, 2). Among these ESs, the amelioration of urban air quality through the removal of air pollutants deserves large attention, owing to the positive impact on human health and well-being (3, 4, 5). Experimental data, such as detailed descriptions of functional parameters, are needed for a reliable quantification of ESs. The present study was carried out in the metropolitan area of Rome, in the frame of the Project PRIN "TreeCity: Planning the green city in the global change era: urban tree functions and suitability for predicted future climates" (6). An urban and a periurban forest has been considered, both dominated by *Quercus ilex* L., which has been chosen as target species for its wide natural distribution in the Mediterranean Basin, as well as for its widespread use in urban contexts. The two studied sites were characterized by different environmental stressor and forest management practices, resulting in different trends of leaf gas exchanges, photosystems functionality and plant water status. During spring, gas exchanges were lower in the urban than in the periurban forest, due to higher air temperature and Vapour Pressure Deficit in the latter site. During summer, instead, in the periurban area the functionality of *Q. ilex* was affected by drought, which did not occur in the urban forest due to higher summer rainfalls as well as periodic irrigations. The water use efficiency was basically lower in the urban park, as well as the photosystems functionality (PSII and PSI). Differences in the intensity of the main phenological phases were also highlighted. Our results point out that the two GIs fulfill a complementary role in the ESs provision in the metropolitan area of Rome, in relation to the ozone removal and the resulting air quality improvement and climate regulation.

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1.10 = GREEN INFRASTRUCTURES FOR THE URBAN AREAS: SOME RESULTS FROM A ONE-YEAR MONITORING ON THE EXPERIMENTAL GREEN ROOFS OF THE UNIVERSITY OF BOLOGNA

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The European Communication (1) on Green Infrastructures (GI) encourages their utilization in various contexts, such as the mitigation of critical environmental situations (high temperatures during the summer period, runoff phenomena following heavy rainfalls, etc.) due to the artificiality of urban environment. This Communication represents a right framework for a significant improvement of the European environmental conditions, as well as for the development of a green economy. It needs, however, to be accompanied by solid technical and scientific skills, tightly integrated among plant ecology, plant physiology, civil and environmental engineering. Especially if the GIs are used in urban areas, this is undoubtedly an essential step for the production of quality material and, possibly, certified for the performances it provides. The wild plant species represent a rich heritage, which can be used for the production of high quality green infrastructures.
They can give excellent results, especially if used in environments whose ecology is similar to that of the original environment. To reach this goal, a wide and not too expensive production of GIs, with wild plant species, must be available, as well as a good knowledge of their responses to the main environmental factors.
In this perspective, the Department of Agricultural Sciences and the Agricultural Farm of the University of Bologna (AUB) are participating in the Project "Green Roofs", promoted by the Department of Civil, Chemical, Environmental and Materials Engineering, the CIRI Building and Constructions and the Technical Office (AUTC) of the same University.
Through this project a first core of experimental green roofs has been realized. Part of the plant species used for the roofs cover, comes from cultivations maintained at the AUB nursery. They have been obtained from seeds and/or cuttings, coming from wild populations of arid and semi-arid grasslands. Part is of commercial origin. Some roofs are instrumented for a continuous recording of the main meteorological parameters, as well as the water content of the substrate and the water runoff.
The results after the first year of monitoring show that green roofs with wild species (*Bromus erectus* Huds., *Lotus corniculatus* L., *Dorycnium pentaphyllum* Scop.) have better performances for the mitigation of summer temperatures, as well as for the mitigation of the runoff phenomena, if compared to green roofs with commercial mixtures of *Sedum* species. Greater maintenance costs (emergency irrigation, cutting, fertilizing) requested by the above mentioned wild species, seem to be largely offset by their better environmental performances. The commercial roof covers with *Sedum* species, preferred because of their low maintenance costs, are, only apparently, more advantageous.
These results are in accordance with those coming from the recent scientific literature on the subject (2, 3, 4, 5) which suggest a careful choice of the plant species, depending on the results to be obtained. The use of stress-tolerant species does not seem always appropriate, especially if the GIs are required for ecological services involving an active interaction plant-environment, even at times of stress. The species most suitable therefore, seem to be those with intermediate strategies (6), such as, for example, the competitors-stress tolerators.

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2 = DYNAMICS OF SUMMIT FLORAS OVER THE LAST CENTURY IN THE N-WESTERN ALPS

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Climate warming over the past century has altered biotic communities and distributional range limits of plants and animals (1). Mountain habitats and ecosystems seem to be particularly threatened and substantial loss of potential habitats for a majority of alpine plant species is forecasted for the 21th century. The species from higher elevation are more exposed to this phenomenon than species at lower elevations (2).
Alpine plants may respond to rising temperatures and longer growing seasons either by adapting to the new conditions or by an upward migration in their distribution ranges to more suitable habitats, leading to an increase of species richness on high elevation areas. Simultaneously with the upward shift of alpine species, a decline in cold-adapted species is expected (3) and rare and endemic high altitude species with narrow distribution ranges are regarded as especially threatened.
Summit areas offer good opportunities for studying the influence of climatic change on plant migration, as plant life at high elevation is mostly dominated by abiotic factors like temperature and snow cover, which are directly influenced by global warming. Furthermore, human impact that potentially masks climate change effects is low compared to lowland regions.
Summits with old records of their flora can be considered as permanent plots (4), and re-surveying of such mountain summits results very suitable to study long-term changes in the flora of high elevation areas.
In order to assess the dynamics of the summit flora, we revisited 13 mountain summits located in the NW-Alps (Valle d'Aosta, Valle di Susa, Vallese) over the past two summers and compared these data with historical floristic inventories from the same mountains, compiled by Vaccari (5, 6, 7), Mussa (8), Braun-Blanquet (1919), Pawlowski (10). Comparisons with recent findings enabled us to investigate 1) plant diversity changes (α- as well as β-diversity), 2) altitudinal shifts of species i.e. migration rates and 3) taxonomic and functional groups associated with migration and colonization success.
The surveyed mountains ranges between 2737 m and 3559 m in elevation and includes 7 siliceous summits and 6 calcareous.
In total the species dataset consisted of 226 species recorded during the historic and recent periods. 20 species were only in historical records while 76 species were found for the first time in the recent records.
Species richness has increased on 11 of the 13 mountains surveyed, with a mean increase of 14.8 species per mountain, while has decreased on 2 mountains.
The sum of the plant occurrences on all summits in the historical period was 483 with a mean of 37 species per summit. In the recent period, there were 662 plant occurrences in total with a mean of 51 species per summit. These data indicated that the average species richness on the considered summits has increased by 37 % in the last century.
Preliminary results indicate that the increase in species richness (α-diversity) of the surveyed summits is not accompanied by a decrease in differentiation between summits (β -diversity or heterogeneity), as expected. This finding suggest that the pool of species which benefit from warming by expanding their ranges upward in a specific summit is not the same for all summits and, therefore, floristic homogenization has not still occurred.

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2.2 = PLASTIC RESPONSES TO ENVIRONMENTAL CHANGES IN THE LONGEVITY OF SEEDS

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Seed longevity is an important plant trait for the *ex situ* conservation and persistence in the soil of many species. Using Controlled Ageing Test (CAT) at high temperature and humidity (e.g. 45 °C, 60% RH) seed longevity has been found to vary considerably across species and the climate at their origin. In particular, plants growing under warm/dry conditions tend to produce seeds with greater resistance to ageing than plants from cold/wet climates (1, 2), suggesting a different ability to withstand ageing after dispersal too. Interestingly, species that are long lived under CAT show a long soil seed bank persistence (3). However, whether ageing processes are similar under CAT and in nature is still not well understood. More recent studies found significant transgenerational changes in seed longevity associated with environment-induced effects (4), indicating that differences between seed lots may be due to plastic responses to the local environment. Our study investigated the extent to which the variation of seed longevity between seed lots that experienced different climate during development and maturation are the results of genetic differences and/or adaptive responses to environmental changes. Using experimental approaches in the lab and in the field we show that seed longevity may depend on both the species-genetic background and environmentally-induced parental effects, with this latter acting through differential mRNA accumulation (5). Furthermore, we suggest that such changes could be adaptive, enhancing the offspring (seeds) to cope with the post dispersal environment. Transgenerational plasticity driven by parental effects may play a fundamental role in survival and persistence of the species in the face of future environmental challenges, such as climate changes.

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2.3 = MODELING THE POLLEN SEASON START IN *CORYLUS AVELLANA* AND *ALNUS GLUTINOSA*

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Hazelnut and black alder are important sources of airborne pollen and represent an allergen threat during the flowering period. Hazelnut and alder act as primers of allergic sensitization to Betulaceae and other pollen allergens and the clinical symptoms become more marked during the birch-pollen season (3). Pollen forecasts are a helpful tool for allergy patient to plan pollen avoidance strategies and the medicine intake which reduces the symptoms. Moreover, researches on hazelnut are conducted also for their importance in fruit production.
In Italy, the pollen seasons of *Corylus avellana* and *Alnus glutinosa* typically last from the middle of December to the end of March or early April. The presence of pollen in the air can be used as a proxy of male flowering. Several researches take into account the relationship between environmental factors, mainly temperature, and the start and trend of the pollen season (3).
The aims of the present study are (i) to investigate the relationships between environmental conditions and the pollen concentration of two early flowering species (hazelnut and black alder) during the flowering period by a multiple regression method (ii) to test a method to predict the pollen season start in order to obtain a helpful tool in allergology and hazelnut cultivation. This method is based on a model proposed by Cesaraccio (1,2) to predict the bud burst in orchards and deciduous forest species and it has been applied to predict the pollen season start of hazelnut and black alder. Moreover, estimating the chilling requirements of hazelnut and the amount of winter chill available at a given location is a central topic in agricultural research, ever since the cultivation ranges of this species have been expanded in the last 20 years beyond its native range.
The aerobiological monitoring was carried out continuously in Turin, in the period 1983-2014, using a Hirst spore trap, VPPS 2000 Lanzoni, placed in the centre of the city at 12 m above the ground level and absorbing 10 l air/min (3). The location of the spore trap did not change in the monitoring period. The standard method of collection was used, and average daily pollen concentration was expressed as grains/m^3 of air. The yearly pollen concentration time series of the two different taxa were manually inspected and it was decided to consider the period from 2001 to 2014 for *C. avellana* and the period from 1996 to 2014 for *A. glutinosa*.
The temperature values for the sampling area throughout the study period were obtained at the meteorological station network of the Piedmont Regional Agency for the Protection of the Environment (ARPA Piemonte). In this study, the methods applied (3) define the pollen season with three different thresholds as the period in which 90%, 95% and 98% of the total season's catch occurred. Moreover, the main peak appearance date (MPA) was calculated for each year as the day in which the maximum value of daily pollen concentration is reached. The relationships between meteorological parameters measured on daily based (mean temperature, rainfall with 1, 3, 5, 10 mm as threshold, wind and mean humidity) and the DOY of pollen season start (PSS) of *C. avellana* and *A. glutinosa* were investigated using correlation and regression analysis to identify the pollen season start. Correlation and regression analysis were carried out for each percentage calculated (90%, 95% and 98%). Spearman's correlation test of the mean daily values was used to evaluate these relationships.
The model proposed by Cesaraccio (1,2) to predict the bud-burst is a sequential model called Chill days model (C_D) based on the concept of chill days (C_d) to break rest and accumulation of anti-chill days (C_a) to overcoming quiescence. Negative C_d values are accumulated until they reach a value called chilling requirement (C_R). The chill days and anti-chill days both depend on the selection of a temperature threshold (T_C) and C_R, so these parameters are iterated to find the combination that best predicts the bud-burst dates.
The accuracy of Chill days model (1,2) and the agreement between observed and estimated data of PPS were evaluated with the mean absolute error (MAE), the relative root-mean-square error (RRMSE), the efficiency index (EF), and the coefficient of residual mass (CRM) index.

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2.3 = MICROTOPOGRAPHY ENHANCES THE COEXISTENCE OF DIFFERENT PLANT FUNCTIONAL TYPES AND CONSEQUENTLY EXTENDS THE GROWING SEASON IN SUBALPINE GRASSLANDS

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Mountain ecosystems are generally characterized by complex topography, where the presence of microhabitats and related differences in soil characteristics and snow dynamics induce patches with uneven distribution of species of different plant functional types.
In subalpine and alpine grasslands where the growing season is generally limited to 3-4 months or less, different functional types clearly reflect distinct adaptations to resource use efficiency, reproductive competition and life cycle completion strategies (1).
In this study the vegetative phenology of a subalpine grassland located in the Alps, in the LTER site of Torgnon (Aosta Valley) at 2160 m a.s.l., has been detected from 2009 to 2014 applying direct (biomass and LAI) and indirect methods (nadiral repeated photography, digital camera images, eddy covariance data) (2, 3, 4, 5, 6) in the framework of the PHENOALP EU Interreg Project (www.phenoalp.eu). The vegetation is characterized by the dominant oligotrophic grass *Nardus stricta*, the keystone species, and the forbs *Geum montanum, Arnica montana, Trifolium alpinum, Ranunculus pyrenaeus* and *Leontodon hispidus*.
A recent analysis carried out with digital images showed that differences in snowmelt dates were unexpectedly negatively related to the beginning of the growing season (6). The negative correlation was explained considering that areas characterized by different microtopography have also a different species composition: on the convex areas *Nardus stricta* is dominant (with average cover of 93%) and the concave areas are characterized by forbs, the most abundant species being *Geum montanum* and *Arnica montana* while *Nardus stricta* has an average cover of less than 50%. The forbs are distinctly dominant in the concave areas and are clearly characterized by an early greening after snowmelt, while the grass *Nardus stricta* is dominant in the convex areas and its greening is delayed. This behaviour was especially evident in 2011 that was characterized by an extremely anticipated snowmelt, when the forbs developed 4-5 weeks before the greening of *Nardus stricta*.
A detailed micromorphological analysis of the soil properties in convex and concave areas has been carried out in order to show the relationships between the plant functional types and the heterogeneous environmental conditions. The analyses highlighted the presence of hummocks, small cryogenic mounds closely spaced in the grassland, which established specific variations on soil chemico-physical properties and different microclimatic conditions compared to the interhummock positions which have higher nutrient and water availability.
In addition, to investigate different litter decomposition rates in relation to microtopography and plant species, litter bags containing only forb and only *Nardus stricta* were located in hummocks and interhummocks positions. The results show a faster decomposition of forbs in the nutrient-richer interhummocks topsoils compared to the podzolized hummocks positions, and an overall slower decomposition rate of *Nardus* litter.
Our work indicates that microtopography enhances the coexistence of complementary plant functional types and consequently increases the possibilities of extending the growing season length and influencing the specific composition of the community, expecially when an early snowmelt occurs.

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2.4 = TCP15 REGULATES GERMINATION TOGETHER WITH DELLAs

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The TCP transcription factors contain a 59 amino acid basix-helix-loop -helix (bHLH) DNA binding domain. The acronyms “TCP” derives from TEOSINTE BRANCHED (TB1, Zea mays), CYCLOIDEA (CYC, Antirrhinum majus), and from the rice PROLIFERATING CELL FACTORS 1 and 2 (PCF1 and PCF2).
Here we describe the role TCP14 and TCP15 as key regulators of Arabidopsis seed germination. The *tcp14* and *tcp15* single mutants and the *tcp14/tcp15* double mutant are characterised by a strong delay in germination. However such phenotypes can be partially rescued by either adding gibberellins or by prolonged vernalisation, suggesting a possible role of these two transcription factors in gibberellin homeostasis.
DELLA proteins are negative regulators of gibberellin signalling and they act immediately downstream of the GA receptor. Both TCP14 and TCP15 heterodimerise with DELLA proteins. All together our data indicate that the joint regulation of germination, by gibberellin and TCPs, occurs through physical interactions with DELLAs. Our data indicate these two TCPs participate to regulate cell cycle related genes in response to GA.

2.4 = TRANSCRIPTIONAL PROFILING OF THE SHOOT APICAL MERISTEM OF RICE DURING TRANSITION TO REPRODUCTIVE GROWTH

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Rice (*Oryza sativa*) is a short day plant in which flowering is promoted when day length falls below a critical threshold. Changes in day length are perceived by leaves and result in the synthesis of long-distance systemic signals called florigens. The rice genome encodes at least two florigens, *HEADING DATE 3a* (*Hd3a*) and *RICE FLOWERING LOCUS T 1* (*RFT1*), whose cognate proteins move to the shoot apical meristem (SAM) to induce flowering. During floral transition, the SAM initiates extensive morphological changes, ultimately leading to panicle development, and the internodes elongate to expose the mature panicle in a process called heading. Currently, very little is known about the dynamics of gene expression at the SAM upon photoperiodic induction. In order to elucidate the transcriptional reprogramming of the SAM during floral transition, we have performed RNA-sequencing experiments on apical meristems of the *temperate japonica* cultivar Nipponbare during the early stages of floral transition. We identified several genes whose expression is dependent upon the photoperiod and/or florigenic proteins. *DOWNREGULATED DURING TRANSITION 1* (*DDT1*), a transcription factor belonging to the C2H2 zinc finger family, is strongly repressed at the meristem during transition from the vegetative to the reproductive stage. Transgenic rice plants overexpressing *DDT1* under the control of the *pACTIN* promoter develop extra glumes during flower development, a phenotype consistent with prolonged expression of vegetative characters. We generated targeted lesions at the *DDT1* locus using genome editing technologies and isolated frame-shift mutations in the coding sequence of the gene. *ddt1* mutant plants failed to repress internode elongation under long days, resulting in a prostrate phenotype. These data indicate that *DDT1* limits the vegetative-to-reproductive phase change and that flowering and internode elongation can be genetically uncoupled.

2.4 = EFFECTS OF HERBIVORY ON POLLEN FLOW, REPRODUCTIVE SUCCESS AND TROPHIC INTERACTIONS IN BRASSICA RAPA

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Multi-trophic interactions involve several players like plants, pollinators and herbivores with specific inter- and intra-relational dynamics. The introduction of exotic herbivores, that interact with native species, may have an impact on the strength of the existing interactions (1). At the present, we know that herbivory imposes a strong re-modulation of the available resources and has a direct cost for the plant, through the removal of tissues and the reduction of available resources that can be allocated to fruits and seeds (2). In addition to its direct impacts on fitness, herbivory can alter floral signaling, such as morphological or chemical floral traits. This could influence pollinator behavior, which may indirectly affect pollinator-mediated fitness. In a recent study (3), we showed that herbivory reduces the quantity of several floral volatile organic compounds (VOCs) and this decrease could potentially reduce attractiveness of flowers to pollinators. However, plants and native herbivores often share a long history of antagonistic co-evolution, where selection is expected to minimize the effects of herbivores on overall plant fitness. In contrast, plant responses to exotic herbivores may differ from responses to native herbivores and affect more negatively the reproductive success (4). The extent to which native and exotic herbivores affect pollinator behavior and then reproductive success in plants is still poorly understood. To get more insights into these interactions, we are investigating the effect of native and exotic herbivory on plant pollen flow and reproductive success by using *Brassica rapa* as model system. In particular we asked: What is the indirect effect of the native specialist herbivore *Pieris brassicae* on the reproductive success of the plant? In the same way, what is the indirect effect of the native generalist herbivore *Mamestra brassicae*? Could the introduction of the exotic herbivore *Spodoptera littoralis* in the native contest change pollinator behaviors and ultimately the reproductive success of *Brassica*? In order to detect whether the fitness of *Brassica* is affected by local and exotic herbivores, we exposed to diurnal pollination artificial plots of *Pieris*-infested, *Mamestra*-infested, *Spodoptera*-infested and control *Brassica* plants. The experiment was performed in several replicas and for each plant treatment we measured the number of flowers, pollination time, foliar damage, reproductive success in terms of fruits number and seed set, and the pollen transfer through the use of powder fluorescent dyes. A preliminary analysis of the relative fruit reproductive success (ratio fruits : flowers) shows no significant difference between herbivore-infested and control plants, both for local specialist-, generalist- and exotic herbivores. However, by measuring the pollen transfer with fluorescent dyes, we found a trend where *Spodoptera*-infested donor plants exchange less pollen then local herbivore-infested and control plants but this apparently do not affect the overall fruiting success. At the present, we cannot conclude whether exotic herbivores influence the pollinator behavior and the reproductive success of *Brassica* in a different way than local herbivores.

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2.5 = EFFECT OF SALINITY ON SEED PROTEIN COMPOSITION OF THREE LANDRACES OF *CHENOPODIUM QUINOA*

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Considering the current models of climate change, lands under salinization are expected to increase during this century (1). Even if salinity effects may not always be negative and in some cases have some favorable effects on quality and disease resistance of the crop, usually salt stress is one of the main abiotic factors limiting crop productivity and plant distribution worldwide, since most species are salt sensitive (2). Despite this, some plants (tolerant glycophytes and halophytes) can tolerate high levels of salt by efficient morphological, physiological and molecular mechanisms (3).
Quinoa (*Chenopodium quinoa*) is an Andean native crop grown primarily for its edible seeds. It exhibits remarkable tolerance against several abiotic stresses such as salt, drought and extreme temperatures. Moreover, quinoa plant is regarded as one of the crops that might sustain food security in this century for its excellent nutritional features, such as high protein content, the unique amino acid composition and the absence of anti-nutritional factors (4). Although the nutritional properties of quinoa seeds have been described and different genotypes analysed, the effect of salinity on the seed protein content and profile has not been thoroughly investigated.
In the present study, seed harvested from plants growth at two levels of salinity (100 and 300 mM NaCl) were used to isolate and characterize the major seed storage proteins. The three quinoa landraces used (R49, VI-I and VR) were originating from different areas (along a north-south gradient) of Chile.
Differences between genotypes were evident, but in general, the yield in terms of seeds dry weight was significantly decrease by salt treatment, as well as the total protein amount. The total protein pattern showed genotype-specific bands that were drastically increased, in a dose-dependent manner, by salt treatment. These bands corresponded to the protein fraction enriched in albumins and globulins, and in particular to the chenopodin, the 11S globulin quinoa's major seed storage protein. The general reduction of protein content in relation to salt stress was instead due to the general decrease of other seed storage proteins, the 7S globulins.
As quinoa is receiving considerable attention as alternative for the formulation of gluten-free products, the aim of this study was also the characterization of protein profiles of three different quinoa varieties. Gliadins and glutenins, the toxic protein for celiac subjects, were barely extractable and hardly detectable as present in very low amounts. Moreover, these proteins were shown not to be the preferred substrates of the mammalian enzymes Transglutaminase (TGase) that plays a crucial role in the initiation of celiac disease.

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2.5 = DECIPHERING THE REGULATORY MECHANISMS BEHIND THE IMPROVED RESPONSE TO WATER DEFICIT AND RECOVERY OF *MEDICAGO TRUNCATULA* EXPRESSING the *At*TPS1 GENE

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Drought has a major impact on crop growth, survival and productivity (1). The manipulation of trehalose metabolism was shown to be a promising approach to improve abiotic stress tolerance in plants (2). We proved that transgenic *Medicago truncatula* plants ectopically expressing trehalose-6-phosphate synthase 1 (*At*TPS1) from *Arabidopsis thaliana* have improved responses to water deficit (WD) and WD recovery (WDR) (3). Our aim is to understand the transcriptional regulatory mechanisms underlying the differential response to WD and WDR of plants expressing *At*TPS1.
Using Illumina-based Massive Analysis of cDNA Ends (MACE), we were able to assess the transcriptome changes in the roots and leaves of transgenic and wild type plants under WD and WDR conditions. Differentially expressed (DE) transcripts encoding for transcription factors (TFs) were extracted from MACE data. Treatment induced changes were studied by comparing WW, WDR and WDR in Wt and in transgenic plants; the genotype effect was analyzed by comparing transgenic and Wt plants exposed to the same water condition. DE TFs were defined as those with a FDR ≤ 0.05 and with a foldchange ≥ 2 or $\leq 0{,}5$. A total of 216 TFs were found to be DE in at least one of the comparisons.
In general, the comparisons WW vs WD and WD vs WDR showed the higher amount of DE TFs. Transgenic plants (leaves and roots) showed a total of 143 DE TFs while Wt plants showed 199 DE TFs. Across the two genotypes 127 TFs were commonly DE, while 16 and 71 TFs showed an exclusive differential expression in the transgenic and Wt plants, respectively. Also, independently on the genotype, roots showed a lower number of DE TFs (118) than leaves (155).
Hierarchical clustering evidences a group of TFs with an opposite expression profile between transgenic and Wt plants. In leaves, after the imposition of water deficit, there was a group of 13 TFs up-regulated in the transgenic and down-regulated in the Wt. After the rehydratation period, a group of 3 TFs is up-regulated in the Wt and down-regulated in the transgenic plants. In the roots of plants under WD, 3 TFs were down-regulated in the transgenic plants and up-regulated in the Wt ones, while the expression profile of TFs in the WDR condition was found to be similar to the WW. In these cases, TFs with altered transcript abundance included members of the TFs families ethylene-responsive, MYB and MYB-like, Hsf, GRAS family, bHLH, GATA-type, WRKY and others.
The overall results will provide a list of TFs with possible role in the improved WD tolerance shown in *M. truncatula* plants overexpressing the AtTPS1 gene to be validated by RT-qPCR in furthers studies.

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2.5 = STUDY OF REEDS-BEDS DECLINE IN DIFFERENT FRESHWATER ECOSYSTEMS OF CENTRAL ITALY: FIRST INSIGHTS

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In the last two decades, evident signs of decline, or die-back, were detected on common reed [*Phragmites australis* (Cav.) Steud. subsp. *australis*] populations at first in central Europe, and more recently also in the Mediterranean area. Reed die-back can be caused by a wide range of factors such as regulation of water table, eutrophication, reduced rhizome ventilation and low genetic variability. In Italy, the die-back was first detected in the Po delta and in the Trasimeno Lake. Studies in Trasimeno Lake were carried out by our group: we demonstrated the presence of the die-back syndrome but also the close correlation between some chemical and cytological parameters and the health status of *P. australis* stands and consequently of the whole ecosystem. A wide-scale screening in central Italy, currently in progress, indicates the presence of die-back symptoms in other palustrine areas. We analyzed the status of reed populations in different freshwater ecosystems in Central Italy (Chiusi Lake, Vico Lake, Trasimeno Lake, Colfiorito's marsh, Fucecchio's marsh), in order to detect any symptoms of die-back and to investigate any possible correlations with the environmental features. Field surveys were carried out at the end (August - September 2014) and at the beginning (March 2015) of the vegetative season. Macromorphological, cyto-histological, genetic and vegetation data were collected. In order to quantify the status of regression/advancement of the *P. australis* populations in the studied areas, aerial photo were also used for diachronic analysis. The preliminary cross results outlined the presence of the reed retreat in some of the analyzed freshwater ecosystems and the presence of substantial differences between flooded and dry plots. Starch storage in rhizomes and roots turned out to be a useful parameter to investigate, as we found out the existence of a correlation with some die-back symptoms, such as the clumping habit. This study will extend knowledge on reed die-back in the Mediterranean basin and will help us to check the health status of important wet ecosystems, included in Natura 2000 areas.

2.5 = AUXIN ACCUMULATION AND TRANSPORT IN *ARABIDOPSIS THALIANA* (L.) HEYNH ADVENTITIOUS ROOTS ARE MODIFIED BY CADMIUM AND ARSENIC

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Cadmium (Cd) and arsenic (As) are very toxic elements for all organisms and the environment. They are not essential elements for plants but can be easily absorbed by the plant root system provoking damages in tissues of sensitive plants, e.g. *Arabidopsis thaliana*. Damages to the root system have negative consequences on plant growth and productivity. It is known that the quiescent centre (CQ) of the root apical meristem controls the root development in the primary root (PR), lateral roots and adventitious roots (ARs) (1,2). The QC definition and maintenance depend on polar auxin transport and accumulation in the stem cell niche surrounding the QC. The membrane carriers LAX3 and PIN1 are involved in root polar auxin influx and efflux, respectively (3,4). *DR5::GUS* line is a useful system for monitoring auxin localization at cell and tissue levels. Our investigations, based on morphological analyses of Columbia (Col) plantlets and histochemical GUS analyses on ARs of *QC25::GUS* (i.e., QC identity marker) transgenic plantlets, grown in the same experimental conditions of this research (5), revealed that these pollutants affected AR development, either increasing AR formation (60 µM $CdSO_4$, combined or not with 100 µM $Na_2HAsO_4{\cdot}7H_2O$) or inhibiting formation and growth (400 µM $Na_2HAsO_4{\cdot}7H_2O$). In all cases both pollutants altered the QC definition and maintenance. These alterations prompted us to investigate whether the two pollutants may cause anomalous IAA levels/distribution during AR development.
To the aim, we investigated auxin levels in the wild type, and auxin localization in the *DR5::GUS* transgenic line, under *in vitro* growth in the presence/absence of 60 µM $CdSO_4$ or 400 µM $Na_2HAsO_4{\cdot}7H_2O$ or in the presence of both salts. The plantlets were cultured in continuous darkness for nine days and then transferred to the light (long-day exposure) for additional seven days. The effects of Cd and/or As on auxin transport were also investigated in the ARs by analyses on *PIN1::GUS* and *LAX3::GUS* lines.
Results show that no signal was detected in the QC and surrounding stem cell niche of the ARs in *DR5::GUS* plantlets treated with 400 µM As, indicating that the semimetal compromised the regular establishment of the auxin maximum necessary for the correct definition and maintenance of the stem cell niche (2). Cadmium, combined or not with As, either caused a significant reduction in the number of ARs with a normally localized *DR5::GUS* signal with respect to control treatment, or induced a weak signal often dislocated to the columella cells. These results indicated that also the metal altered the auxin maximum necessary to the correct stem cell niche organization and functioning. Cd and As also interfered with the expression patterns of *PIN1::GUS* and *LAX3::GUS*, especially in AR primordia. Cadmium alone negatively affected auxin efflux by PIN1 causing its unexpression in the most of the primordia. Arsenic, alone or combined with Cd, exhibited minor effects in inhibiting *PIN1* expression. The auxin influx by LAX3 was reduced in AR primordia in the same way by both pollutants, alone or combined. In conclusion, Cd and As alter auxin apical accumulation by affecting the influx and efflux auxin carriers, thus negatively disturbing stem cell niche and QC functioning in the ARs.

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2.5 = TREE LONGEVITY IN TEMPERATE FORESTS: USING TREE-RINGS TO ASSESS THE FACTORS AFFECTING THE MAXIMUM LIFESPAN OF BROADLEAF DECIDUOUS TREES IN DIFFERENT ENVIRONMENTS

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Understanding which factors control the expression of longevity in trees is still an outstanding challenge for tree biologists and forest ecologists. Long-lived trees cope with severe growth suppression episodes and recurrent disturbance events along their multi-century life, and their longevity reflects the ability to withstand with different environmental stresses to gain and maintain a dominant position within the forest canopy.
We used the maximum age of dominant trees in old-growth forests (i.e. those with dominant trees dying naturally, 1) as a surrogate of tree realized longevity. In fact, frequent wood rot and fast decay rates makes it impossible to count tree-rings in declining and dead trees (i.e., the actual measure of tree realized lifespan). We performed a pan-continental analysis of original and literature tree-ring data from 25 species and 12 genera of broadleaf deciduous temperate trees growing in closed-canopy old-growth (OG) forests in the Northern Hemisphere (2). We wanted to explore how genetics, life-history traits, geographic patterns and environmental variability could affect tree longevity. Several broadleaf deciduous tree species growing in different sites of the temperate biome of the Northern Hemisphere showed longevities of 500–600+ years. In general, 300–400 years can be considered a baseline threshold for maximum tree lifespan in many temperate deciduous forests. Amazingly, species from evolutionary distant families and genera converge to similar age limits. These species do not show outstanding commonalities in life history traits, suggesting that functional traits (e.g. wood density or chemistry) may not always be the dominant determinants of longevity. We found that slow growth was the only life-history trait strongly associated to tree longevity. Thus, when site factors reduce growth rates tree maximum lifespan tends to increase. As a consequence, maximum age can vary greatly in relation to environmental features, even within the same species. At the hemispheric level, the meta-analysis conducted for the temperate deciduous forest biome highlighted convergent geographic patterns in maximum lifespan and temperature variability. The dependence of maximum lifespan on temperature is stronger in mesic forests (i.e. *Fagus*-dominated forests in Europe or Japan), with temperature affecting positively growth primarily through the growing season length. This relationship appears instead weaker in deciduous *Quercus* spp., maybe in relation to oaks' capacity to adapt to extremely dry, wet, or unfertile site conditions, and warrants further research in disturbance ecology.
Slower growth rates, and the associated smaller size, provide trees with an advantage against biotic and abiotic disturbance agents, supporting the idea that size, not age, is the main constraint to tree longevity. The oldest trees we found were living most of their life in subordinate canopy conditions and/or within primary forests in cool temperate environments and outside major storm tracks. Very old trees are thus characterized by slow growth and often live in forests with harsh site conditions and infrequent disturbance events that kill much of the trees. Biological, ecological, and historical drivers must all be considered to understand the constraints imposed to longevity within different forest landscapes.

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2.5 = CADMIUM AND ARSENIC AFFECT ADVENTITIOUS ROOT FORMATION AND THE DEFINITION OF THE QUIESCENT CENTRE IN *ARABIDOPSIS THALIANA* (L.) HEYNH PLANTLETS

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The heavy metal cadmium (Cd) and the semimetal arsenic (As) are the most toxic elements for animals and plants. Both are known to alter the architecture of the whole root system in numerous plants. In particular, in *Arabidopsis thaliana*, they reduce primary root (PR) growth, induce damages to the root anatomy and affect the root differentiation pattern (1-3). Adventitious roots (ARs) are essential for the post-embryonic development of the root apparatus in a wide range of plant species, contributing to anchorage, water-use efficiency, and extraction of nutrients from the soil. Moreover, their presence may enhance the plant ability to extract toxic elements from the soil. In *A. thaliana*, the quiescent centre (QC) of PR, lateral roots (LRs) and ARs controls the apical growth through an involvement of auxin and cytokinin (4,5). Therefore, the correct definition, maintenance and activity of the QC are essential for the organization of the roots. However, the effects of Cd and As on *A. thaliana* AR formation, and on QC definition, are unknown. The aim of this research was to investigate whether Cd and As affect AR formation in *A. thaliana,* and to determine their effects on QC definition in the ARs. To the aim, seeds of *A. thaliana* Columbia (Col) ecotype and of the transgenic line *QC25::GUS* (QC identity marker for PR, LRs and ARs; 5,6), were sown *in vitro* in the absence (control treatment) or in the presence of 60 µM $CdSO_4$ (Cd), or 400 µM $Na_2HAsO_4{\cdot}7H_2O$, (As) or 60 µM $CdSO_4$ plus 100 µM $Na_2HAsO_4{\cdot}7H_2O$ (Cd+As). In order to favour AR formation, the exposure to light (long-days) was preceded by nine days under continuous darkness (16 days of total growth period). Mean length of PR, hypocotyl length and AR density were evaluated in the absence and presence of the pollutants in both wild type and *QC25::GUS* plantlets. Moreover, in *QC25::GUS* line, AR QC definition and maintenance were investigated through histochemical GUS assays. The presence of the GUS signal and its localization in the apical root meristem were investigated starting from the stage VII of AR development (i.e. the stage of QC definition, 5). The results show that both pollutants, alone or together, significantly reduced the PR and the hypocotyl growth, with no significant differences among treatments. The presence of 400 µM As reduced significantly the percentage of plantlets with ARs, compared to the control treatment, whereas Cd alone did not cause any reduction. However, the plantlets treated with Cd, alone or combined with As, showed a greater density of ARs, unlike those treated with only As, in which the AR density decreased significantly. Moreover, in As alone treatment, the most of the roots were primordia at early stages. In addition, the percentage of ARs without GUS-signal in the QC significantly increased in the presence of the pollutants, and especially with 400 µM As and the combined treatment, with respect to the control treatment. Cadmium alone also provoked a shift of the GUS-signal to the columella cells. All together, these results suggest that Cd and As differently affect the QC, resulting into different AR development.

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2.5 = LOCAL ADAPTATION AND GENE FLOW IN THE SERPENTINE *DIANTHUS SYLVESTRIS*.

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Patchy distribution and stressful conditions of environment can induce the emergence of locally adapted phenotype among populations. Evolutionary theory supports that local adaptation is drown by strength of divergent selection to favour the genotype that better performs in a specific habitat. Nevertheless, adaptation could occur also via phenotypic plasticity that allows individuals to rapidly change their phenotypic response to environment and this ability may even slow down the effect of adaptive genetic divergence.
Plants from serpentine represent a typical model for studying local adaptation to soil type (Linhart & Grant, 1996) as selection in this environment is very intense and leads to the evolution of locally adapted populations, a phenomenon known as "serpentine syndrome" (Brooks, 1987).
Dianthus sylvestris Wulfen (Caryophyllaceae) is frequently found both on serpentine and limestone bedrocks. A previous genetic analysis with microsatellite molecular markers has showed high level of gene flow among serpentine and limestone populations in central Italy. To determine the contribution of selective factors and/or phenotypic plasticity to local adaptation of *D. sylvestris* to serpentine, from populations already examined in the previous genetic analysis, we have estimated the metal content in plant aerial parts, collected data on morphological traits, and performed field reciprocal transplantations.
High metal content (Ni, Cr) in plants aerial part confirmed, as in previous studies on other carnations, the bioaccumulation of heavy metals in *D. sylvestris* plants from serpentine soils. In these plants, several morphological traits were found statistically decreased when compared to plants from limestone so highlighting that serpentine is a less permissive habitat than limestone. However, most of the morphological differences disappeared in transplanted individuals suggesting a large contribution of phenotypic plasticity in determining the observed morphological divergences. Nevertheless, in transplanted plants from serpentine soil to limestone, a two-way ANOVA resulted in a significant difference in biomass with an effect of the original soil on the transplanting soil. Significant differences were also found in flowering time, as plants from serpentine, when transplanted on limestone, flowered before the resident limestone plants. These differences, persisting independently from the original soil type, should have genetic bases. Thus genetic differentiation of populations of *D. sylvestris* is occurred at least in few selected loci determining different affinity for the two habitats. This divergence is maintained among populations from different soil types even in the face of extensive gene flow as observed at neutral loci.

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2.5 = DYNAMICS OF PHOTOSYSTEM II SUPERCOMPLEX IN PEA PLANT LEAF TISSUES UPON EXPOSURE TO DIFFERENT INCIDENT LIGHTS

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At the heart of the photosynthetic process there is the Photosystem II (PSII), a multisubunit thylakoid membrane pigment–protein complex, responsible for light-driven oxidation of water and reduction of plastoquinone. In plants and green algae, PSII has an associated light antenna system (LHCII), responsible for increasing its light harvesting efficiency, forming the PSII-LHCII supercomplex (PSII-LHCIIsc) (1).
The main objectives of the FIRB 2013 research program entitled "Structure and structural dynamics of Photosystem II supercomplex in higher plants upon exposure to different incident lights" are the elucidation of the three-dimensional structure of PSII-LHCIIsc from garden pea (*Pisum sativum* L.) plants and the achievement of a better understanding of PSII-LHCIIsc assembly/disassembly. PSII-LHCIIsc is known to be a dynamic structure but there is still a lack of information on many functional aspects of its assembly/disassembly in relation to the optimization of the photosynthetic efficiency in different tissues during plant morphogenesis (2) and in response to different light conditions. Through biochemical/biophysical studies of their re-arrangement under different light conditions, we expect to provide more information about the molecular details of how plants react to the continuous changes in light quality and quantity that challenge the performance of the photosynthetic apparatus.
The first part of this research program focuses on the structural organization of PSII-LHCIIsc in different photosynthetic tissues of pea plant: the leaf is pinnately compound and consists of basal, foliaceous stipules, proximal leaflets and distal tendrils (Figure 1). Besides leaflets, stipules are known to be highly effective photosynthetic leaf parts, responsible for up to 30% of leaf photosynthesis and essential for high harvest index and grain yield (3). In this study, differences between leaflets and stipules have been characterized in terms of photosynthetic yields, by means of PAM fluorimetry (Figure 2), and chlorophyll-protein complexes association, by means of blue-native-PAGE, including stroma-exposed domains (Figure 3).
A better understanding of the regulation of this dynamic system will give valuable information to develop a comprehensive structural/functional model of PSII-LHCIIsc in higher plants, particularly in terms of regulation to different light conditions.

Fig. 1. Leaf structure in *Pisum sativum* plants

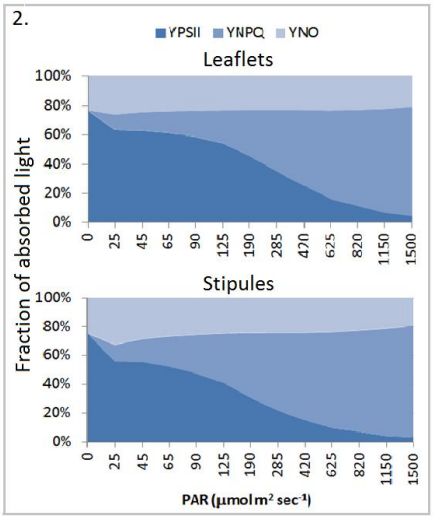

Fig. 2. PAM fluorimetry: photosynthetic yields

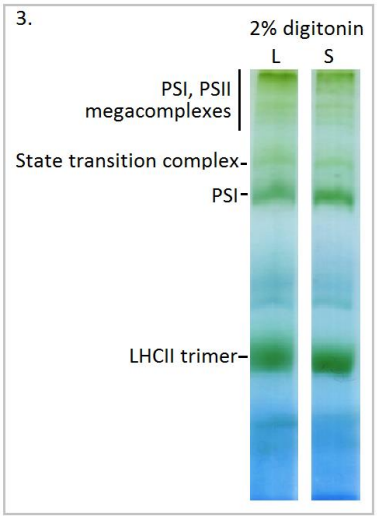

Fig. 3. BN-PAGE of thylakoids from leaflets (L) and stipules (S)

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2.5 = TOWARDS THE IDENTIFICATION OF BIOMARKERS OF GENERAL STRESS CONDITIONS IN HORTICULTURAL SPECIES

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Increasing number of studies have demonstrated that the response of plants to adverse environmental conditions is associated to changes in the expression of several genes, most of which encode enzymatic activities or transcription factors. Although most of these genes appear to be specifically modulated by different types of stress, transcriptomic studies conducted mainly in *Arabidopsis thaliana* have revealed that some of them are modulated in response to many types of stress and could be involved in what has been proposed to be a "general stress response" (1, 2). Nevertheless, recent studies suggest a complex scenario because the patterns of gene expression observed in plant subjected to multiple simultaneous stresses appear remarkably different compared to those seen under single stress conditions (3, 4, 5). This evidence makes more difficult to define common responses and hampers the identification of genes that could be actually involved in a possible general stress response in plants. The identification of such genes in horticultural species would strongly improve our knowledge of their response to environmental stresses and, most importantly, could set the basis for the implementation of molecular analyses that could allow a qualitative evaluation of the horticultural products, even without morphological evidences, and the optimization of their cultivation. In this study we report an attempt to identify genes that, on the basis of their strong homology to hortologs from *Arabidopsis thaliana*, could be possibly involved in a general stress response in tomato, a model horticultural species for which the complete genome sequence has been reported. More specifically, comparative analyses allowed us to identify putative tomato hortologs of the ZAT10, GBF3, LBD1, DIN11, AKR4C8, BGLU11 and BGLU8 genes of *Arabidopsis thaliana* and analyses of their expression confirmed their involvement in the stress response of tomato plants. These results could open the way to the definition and the optimization of protocols for the conduction of molecular analyses of quality control in important horticultural species.

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2.5 = *PTERIS VITTATA* L. IS ABLE TO COUNTERACT THE TOXICITY INDUCED BY CADMIUM COMBINED WITH ARSENIC

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Pteris vittata L. (Chinese brake fern) is the first fern to be identified as a hyperaccumulator of the semimetal arsenic (As). The fern can accumulate more than 27000 mg As kg^{-1} dry weight in its fronds and for this reason it has been proposed to remove As from contaminated soil and water (1). Due to excessive agricultural and industrial practices, frequently As is released into the environment with other toxic metals, such as Cadmium (Cd). Both Cd and As are a threat for human health and ecosystems due to their accumulation and long permanence in the soil, in the food chain and locally in drinking water. The toxicity of Cd and As, for plants and animals, is due to their high reactivity with biological molecules and to their similarity with essential ions that they can replace altering cellular homeostasis. The plants easily adsorb these elements through their root apparatus. In plant cells, Cd and As cause oxidative stress reducing glutathione (GSH) level with severe consequences at all organism level. For this reason there is a need to find a natural system able to remove As and Cd efficiently (2). Lately, it was demonstrated that the fern is able to absorb and accumulate higher levels of Cd and As when exposed to both elements in comparison with when exposed to single pollutants (3). The interaction between Cd and As in the soil may have synergic or antagonistic effects on uptake and accumulation of the two ions in the plant organs (4). In the plant cells, Cd and As induce the synthesis of small peptides called phytochelatins (PCs), with a general structure (g-Glu-Cys)n-Gly (n = 2–11). These peptides are able to form complexes with metals and semimetals (such as Cd and As) in order to reduce their toxicity by sequestering them in the vacuole (5). The fern also produces PCs after As exposure (6). Today the response of *Pteris vittata* to Cd and As combined toxicity requires further investigation because it is unknown whether the fern uses the PCs as detoxification system when exposed to both pollutants and if other strategies are implemented. Thus,the aim of this research was to investigate if and how the fern is able to counteract metal and semimetal toxicity by analyzing the formation of phytochelatin complexes and the morphogenic responses when exposed simultaneously to Cd and As. The plant response was evaluated by analyzing Cd and As accumulation in roots and fronds, by measuring the plant biomass, by analyzing histologically the fronds and evaluating the formation of PC-As complexes. Furthermore the exudates extruded from the fronds were chemically analyzed to assess the contribution of the extrusion mechanism to the metals detoxification in the fern. The results show that plant morphology is strongly affected by Cd presence combined with As. In addition their simultaneous exposure increases the semimetal uptake while reduces the metal one. The exudates extruded from fronds are composed by both Cd and As. Furthermore in our results, the fern exposed to As and Cd together forms different As-PC complexes in the roots in comparison with the fronds. In conclusion, we can state that *Pteris vittata* activates strategies that helps it respond to the additive toxicity of Cd combined with As.

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2.5. = MORPHO-PHYSIOLOGICAL AND MOLECULAR TRAITS THAT CONTRIBUTE TO EXCEPTIONAL SALINITY TOLERANCE IN *CHENOPODIUM QUINOA* WILLD.

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In Chile, there are two ecotypes of quinoa (*Chenopodium quinoa* Willd.), *salares* (salt flats) and lowlands/coastal, exhibiting significant differences in adaptation to altitude, water availability and soil salinity. Within these ecotypes there is a high genetic diversity, allowing landraces to adapt to contrasting agro-ecological gradients (Bazile et al. 2015) [1]. Being a halophyte, quinoa can complete its life cycle at very high salinity levels, although growth may be retarded (Hariadi et al. 2011) [2]. Many comparative studies have been conducted to evaluate the potential of quinoa genotypes to cope with salt stress and to attempt to understand the morphological and physiological mechanisms of salt tolerance (Ruiz et al. 2015) [3]. Few studies have dealt with an analysis of genes related to salinity tolerance (Maughan et al. 2009 Ruiz et al. 2011) [4, 5]. In the present work, the expression of genes involved in abiotic stress responses, and in particular those involved in polyamine and abscisic acid metabolism, were investigated during short-term exposure to salt stress in two Chilean landraces, one belonging to the *salares* ecotype (R49) and one to the lowlands/coastal ecotype (VR). Seeds were germinated *in vitro* on MS medium. After one week, seedlings were transferred to MS medium supplemented or not (controls) with 300 mM NaCl. Roots and shoots were collected separately at different times (0.5, 2, 24, 144 h after transfer) for polyamine quantification by HPLC and RealTime-qPCR analysis of transcript levels. Root architecture was modified in response to 300 mM NaCl relative to controls. Polyamine content and the expression of several stress-related genes (NHX, NCED, AOX, CYCD3) and transcription factors that regulate these genes, such as DREB1A and a putative bZIP24, were upregulated in salt-treated seedlings as compared with control ones, and differentially expressed in R49 *vs.* VR.

Acknowledgements: CONICYT-FONDECYT/PostDoc grant Nº 3130624 to KBR.

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2.5 = ADVENTITIOUS ROOTING: WHAT HAPPENS BETWEEN ETHYLENE AND AUXINS?

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Adventitious roots (ARs) are roots that develop from non-root tissues, mostly from aerial plant parts (1). It is an important adaptive response to stress. Several protocols for AR induction have been developed in *Arabidopsis thaliana*, *in planta* and in *in vitro* systems, e.g., stem thin cell layers (TCLs). The latter system improves the knowledge of the process because it allows the study of AR formation in a limited cell context and starting in tissues different from those usually involved *in planta.* ARs are controlled by multiple endogenous and environmental factors, and auxin, described as the rooting hormone, is one of the major control factors *in planta*, and is essential for ARs in TCLs (2). The auxin indole-3-acetic acid (IAA) is a potent growth regulator, but it is known that different auxins have a differential root-inducing ability. In accordance, recent studies have demonstrated the importance of the natural auxin-precursor indole-3-butyric acid (IBA), because IBA-derived IAA is a significant part of the auxin necessary for a lot of the processes related to seedling development (3). Moreover, when applied exogenously, IBA exhibits a greater ability to promote ARs compared with IAA (4). Ethylene (ET) could be another hormone involved in AR formation, because it affects a variety of processes during the plant lifetime, including adaptive stress responses. In particular, ET influences many features of auxin-dependent plant growth by altering auxin signaling, synthesis and/or transport (5). However, in *A.thaliana,* there is still limited information about ET roles on AR formation, and data, about other species are in contrast (6). Instead, there is information about lateral roots (LRs), i.e., the other post-embryonic rooting of the plant, showing developmental stages similar to ARs (2). In fact, in *A.thaliana*, studies *in planta* show an inhibitory effect of 1-aminocyclopropane-1-carboxylic acid (ACC), the direct ET precursor, on LR formation (7). However, this effect seems concentration dependent, because concentrations lower than 10^{-7}M stimulate the process (8).
The present research studied ET effects on AR formation in seedlings and TCLs of *A. thaliana* investigating the possible relationship of ET with IBA and IAA. For this reason, after a preliminary screening of ACC concentrations, AR density was evaluated with/without ACC (0.1µM), and with/without IBA or IAA (10 µM) in both systems, by the use of wild type (wt) and ET/auxin mutants. The presence of both auxins was detected in wt seedlings grown without exogenous hormones (HF). Contrariwise the TCLs showed no significant level of these hormones under HF treatment (9). For this reason, TCLs were cultured in the presence of either IAA or IBA. Only the latter auxin induced a high AR response. As consequence, the IBA treatment was chosen as the AR control treatment for TCLs, to compare with the HF treatment of the seedlings *in planta*. In both systems, the presence of ACC caused a significant reduction in AR density. Because IBA acts only through its conversion to IAA (10), AR production, with/without ACC, was evaluated in knockout mutants of ET and IAA, i.e. the ET insensitive mutants *ein2-1* and *ein3eil1*, the IAA biosynthetic double mutant *wei2wei7* and the IAA partially insensitive double mutant *tir1afb2*. The AR response showed that no change in AR density per seedling/AR number per TCL occurred in these mutants with/without ACC, suggesting that ET uses for AR formation the same reception pathways of all the other ET-dependent processes (11). ET exhibited an indirect action, i.e. modulated IAA biosynthesis and reception, in accordance with the endogenous auxin levels detected. All together, results show that in *A. thaliana* ET affects AR formation *in planta* and TCLs, through a regulation of IAA biosynthesis and reception. The possible ET role on the conversion of IBA into IAA is under study.

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2.6 = THE ROLE OF POPULATION BIOLOGY AND GENETICS IN PLANT TRANSLOCATION: REINFORCEMENT OF *LEUCOJUM AESTIVUM* AND DE-EXTINCTION OF *STRATIOTES ALOIDES*

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The release of individual plants into their native habitat or in new sites, namely translocation, is one of the methods conservationists employ to reduce the risk of losing threatened species, as also suggested by the CBD, GSPC and IUCN. So far, Several types of translocation from population reinforcements to more extreme assisted colonizations and de-extinctions have substantially contributed to the conservation of many threatened species, but much more effort is required as translocation are often ineffective and with high risk of failures (1). Reasons of failure are manifold, but a poor knowledge of the species ecological requirements, species population and genetic structure and dynamics along with the selection of ecologically unsuitable translocation sites are key constraints (2,3). In this context, the future success of translocation will depend on whether we will be able to create a more active multidisciplinary dialogue between related disciplines in order to reduce uncertainties and improve the successes of rare plant translocation. Here two case studies are presented in order to highlight the importance of preliminary studies on species biology and genetics in the establishment of viable population dynamics and genetic structures in translocation. Both cases highlight how the success of a translocation, whatever the type, is highly dependent on preliminary *ad hoc* studies on the target species and on the ability of the operators to reproduce the natural biological, ecological and/or genetic dynamics in the translocated populations.
Case studies are:
- *Leucojum aestivum* (Amaryllidaceae), a perennial wetland-dependent plant threatened by habitat loss and fragmentation in its Euri-Mediterranean range. The study of 26 populations of the species, highlighted density-dependent dynamics related to the reproduction performance (4). High-density wild populations show a greater fruit set and seed set as a consequence of their greater attractivity for pollinators. According to this finding we reintroduced two populations with different density (12 and 24 flowering plants/m^2) near Parma. After 4-years monitoring the high-density population produced ca. 30% more fruits than the low-density population, with a fruit set in high-density population 12% greater than in low-density population.
- *Stratiotes aloides* (Hydrocharitaceae) a dioecious widely distributed aquatic plant, extinct in Italy since 1990, due to decreasing water quality (5). Considered lost forever, the Italian female genotype of *S. aloides* has been rediscovered in an *ex situ* private collection. With the aim to promote the de-extinction of *S. aloides* in Italy through the reintroduction of a new population, we investigated the genetic structure of 9 European and Asian populations of the species, with the specific objective to identify a source population for male individuals. Surprisingly, AFLP of ca. 190 individuals suggested that the populations more closely related to the Italian one are those occurring in the Rhine Basin in the Netherlands and not the closer Bavarian population. Our results suggest that long-distance dispersal mediated by waterfowls may complicate the genetic structure of plant populations, with implication on the choice of source populations for translocation.

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2.6 = DESIGNING PLANT TRANSLOCATIONS: *HYPERICUM ELODES* IN MIGLIARINO-SAN ROSSORE-MASSACIUCCOLI REGIONAL PARK AS A CASE STUDY

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Plant conservation urgently needs a unifying vision of plant life cycle to support conservation planning. The lack of such a vision is glaring especially in the use of ex situ seed bank collections. Indeed, despite the growing use of seed bank facilities (1), and the deeper understanding of seed storage longevity and of dormancy-breaking and germination requirements of wild species, seed banks samples arguably suffer from low genetic diversity (2, 3). Consequently, the use of seed bank collections in translocation projects could result in negative outcomes in the face of future environmental stresses (4). Effectively using seeds in translocation activities also depends on post-germination plant propagation handling practices: protocols for germination developed in seed banks allow radicle emergence but often overlook or completely fail to address cotyledon emergence and thus the growth of fully developed seedlings. Furthermore, the survival of seedlings obtained in benign environments (i.e. laboratories), is often negatively affected during the earliest steps of cultivation: this stage is crucial because it dictates the number of available transplants and ultimately the success of translocation trials.

Based on a case study from translocation project funded by the Migliarino-San Rossore-Massaciuccoli Regional Park in Tuscany (Italy), we focus on the methodology for selecting and generating suitable plant material and programming its release in the translocation sites. We also report results from the installation settings and monitoring over one vegetative season. The target species is *Hypericum elodes* L. a species threatened by climate change (5) living in swallow soft-waters pools, a notable category of freshwater habitat because of their biodiversity value and ecosystem services function (6). This species is able to spread both by seeds and vegetative propagation (7); hence, the plant material was generated from seeds gathered in the wild but also by clonal propagation. Based upon the knowledge of the species' breeding system, seeds were obtained through different hand-pollination techniques as well as through open pollination, to increase the number of genetic lineages in the offspring (8). Each lineage has been followed within the whole line of seed management from fruit set to the reintroduction step. Propagated plants were cultivated under near-natural conditions in pots of various size and shape.

We highlight that laboratory seed germination guarantees the successful control of all sown seeds, especially in the case of seeds with special requirements for germination (e.g. light). We also discuss seedling development and transplanting by testing various methods of seedling cultivation to obtain adult plants. Specifically, different genetic lineages obtained from sexual reproduction have been included within plant assemblages to recreate a heterogeneous population genetic structure.

In conclusion, we propose a methodological approach finalized to produce different types of suitable material with different strategies of colonization and genetic diversity degrees, to be used in appropriate combination to maximize success of the designed translocation.

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2.6 = INTER INDIVIDUAL VARIABILITY IN THE INFLUENCE OF CONE PRODUCTION ON RADIAL GROWTH IN NORWAY SPRUCE (*PICEA ABIES* (L.) H.KARST.)

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Mast-seeding, i.e. the production of occasional large seed crops followed by years of low seed production with a high level of synchronization among neighbouring individuals (1), is a reproductive strategy common of several perennial plants, among which Norway spruce (*Picea abies*).
Although the biomass of reproductive structures usually represents a rather limited fraction of the net annual production by trees (2), it has been frequently hypothesized that since primary production is usually limited by the pool of available resources, a trade-off should exist between growth and reproduction, especially for mast-seeding trees (3), with a consequent constraint in vegetative growth during mast years. With regard to xylem, several studies reported reduced radial growth during mast years both in hardwoods (4) and softwoods (5). Association between large cone production and reduced ring growth has been observed in Norway spruce in several studies (6,7). However, positive correlation between xylem growth and cone production has been observed too (8), thus suggesting that mast associated ring width reduction could vary depending on trees access to resources. In absence of constraints trees with access to resources would be able to overcompensate the effects of a large cone production and would not show any trade-off between ring growth and reproduction.
In this work we expand a study (9) conducted on 13spruce trees in the Valbona Forest Reserve located at 1700 m a.s.l. in the Natural Reserve of Paneveggio (TN, Italy), aimed at investigating the presence of a trade-off between growth and reproduction for trees living at forest edges. In the original study year-to-year variability in individual cone production for the 13 trees was recorded from 1983 to 1990 and related to annual ring increments. In such time span individual trees showed large differences in cone production, which appeared to be consistent among years, and three good seed years (1983, 1985 and 1988) were recorded, but no mast ones. Given the absence of mast years, no effect of the cone number on radial increment could be found, thus supporting the thesis that trade-off between ring growth and reproduction can be overcompensated by trees with access to resources.
In the present study we expand the records time span to 30 years (1983-2013), and test if high cone production trees and low cone production ones show a different trade-off between ring growth and reproduction.

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2.8 = THE INVASION OF *AMBROSIA ARTEMISIIFOLIA* L. IN ITALY: GENETIC VARIABILITY, POPULATION STRUCTURE AND COLONISATION ROUTES

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The study of genetic structure and colonisation sources of alien species populations is essential to identify invasion patterns and the reasons for their success (1). Our study aimed to investigate the genetic pattern of *Ambrosia artemisiifolia* Italian populations in comparison with French and Canadian ones in order to ascertain their origin and to characterize the colonisation areas of *A. artemisiifolia* in Italy by using herbarium specimens.
Molecular analyses were based on 6 SSR markers (2) by which we analyzed a total of 18 populations from Italy, Canada and France. The spatio-temporal spread of *A. artemisiifolia* through Italy was then reconstructed mapping the distributional pathway of 193 herbarium specimens.
Ambrosia artemisiifolia Italian populations scored higher values of genetic diversity than Canadian and French (H_E = 0.683, 0.639 and 0.650, respectively). Across Italian populations their time of residence was positively correlated with observed heterozygosity (H_O) and negatively correlated with the coefficient of inbreeding (F_{IS}). STRUCTURE analysis suggested high level of admixture of populations. Distributional data from herbarium records showed that *A. artemisiifolia* at first colonised the Po plain and later, in different steps, spread out to some Mediterranean areas.
This study links genetic and historical-distributional data on *A. artemisiifolia* population in Italy, emphasizing that several invasion events has occurred through Italian peninsula in different times, determining high levels of gene flow between populations of different origin.

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2.8 = GERMINATION ECOPHYSIOLOGY OF THREE INVASIVE SOLANACEAE SPECIES IN SARDINIA (ITALY)

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Biological invasions by alien plant species have become a major agronomic threat and a focus for weed scientists (1). Most alien species are found in artificial and agricultural areas and they cause losses in production and even control-related costs in arable lands, pastures, nurseries, greenhouses, plantations and in the water management (2). During the first year project "Germination ecophysiology and reproductive biology of invasive species that threaten environment, economic activity and human health", funded by "Regione Autonoma della Sardegna" (Sardinian Local Government), preliminary results on germination ecophysiology of three invasive species of the *Solanaceae* family were obtained. *Solanaceae*, or nightshades, is a family of considerable economical importance which includes species that are either food and drug plants. They also include an high number of poisonous members many of whom are listed as invasive plants in the world (3) and in Sardinia (4). For instance, *Solanum elaeagnifolium* Cav. and *Lycium ferocissimum* Miers, are invasive in the agroecosystems, while *Nicotiana glauca* Graham is invasive in both natural and semi-natural habitats, roadsides, monuments and archaeological remains. All three species are toxic to humans and livestock and for these reason were considered in the present study. Seeds were collected in Sardinia during dispersal season in winter. Germination requirements were evaluated at constant (5-25°C) and alternating temperatures (25/10°C), both in light (12/12 hours photoperiod) and in darkness (0/24 h), salt stress (0, 125, 250, 500 mM NaCl) and its recovery on seed germination were considered for the three species. *S. elaeagnifolium* germinated only under the alternating temperature regime of 25/10°C (ca. 80%) and its seeds were germinated well in both light and darkness, indicating that they are neutral photoblastic and therefore not photo-inhibited. *N. glauca* seeds germinated at all the tested temperatures (ca. 90%), although at 5°C germination decreased (ca. 45%) and light significantly increased seed germination respect to total darkness. *L. ferocissimum*, as the previous species, showed the capability to germinate at all the temperatures (ca. 70%) and germination was higher in light conditions, but only when seeds were previously manually scarified with scalpel. The limit of tolerance to NaCl varied among the species and seed mortality in salt conditions was very high only for *N. glauca*. *S. elaeagnifolium* seeds germinated up to 125 mM and no seed mortality occurred, while *L. ferocissimum* seeds showed their capability to germinate also at the highest NaCl concentration (500 mM) without seed mortality. On the contrary, *N. glauca* seeds did not germinate in any of the NaCl concentrations. *N. glauca* and *L. ferocissimum* showed a low capability of germination recovery after salt exposure. In particular, the seeds of the first species did not recover at the lowest temperature of 10°C, while *L. ferocissimum* seeds did not recover at the highest NaCl concentration. *S. elaeagnifolium* showed a moderate recover capability at all the tested temperatures.
Our results are an important contribution in developing a wide management strategy of these alien species and for the control of the biological invasions in agricultural ecosystems.

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Acknowledgements
This communication has been produced as part of the research project "Studio dell'ecofisiologia della germinazione e della biologia riproduttiva di piante invasive che causano impatti di tipo sanitario, socio-economico e ambientale in Sardegna" developed at the "AGRI AMBIENTE srl, Pula (CA)", by a research grant financed from the resources P.O.R. SARDEGNA F.S.E. 2007-2013 - Obiettivo competitività regionale e occupazione, Asse IV Capitale umano, Linee di Attività I.1.1. e I.3.1.

2.8 = PRELIMINARY ECOLOGICAL DATA FROM A WETLAND AREA CHARACTERIZED BY MASSIVE INVASION OF MYRIOPHYLLUM AQUATICUM (VELL.) VERDC.

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Biological invasions are causing extensive damages to the ecosystems and the economy in many countries around the world (1)(2) becoming a phenomenon of overriding concern. *Myriophyllum aquaticum* (Vell.) Verdc. (Fig.1) is an invasive macrophyte coming from South America, commonly used as an ornamental plant (3), and naturalized worldwide thanks to its phenotypic plasticity(3)(4). In Italy *M. aquaticum* has been reported in several regions (5). In Tuscany it was found in the wetlands of Lake Porta, between Lucca and Massa-Carrara (Fig. 2). This study aimed to provide a preliminary ecological survey of the invaded habitats around the Lake Porta and an assessment of *M. aquaticum* influence on these areas.
The study involved 16 plots: 8 invaded by *M. aquaticum* and 8 non-invaded. In each of the 16 sampled plots, temperature, pH, salinity, dissolved oxygen and global and underwater irradiance were measured. Water samples were filtered and used for nutrient (NO_2^-, NO^{3-}, PO_4^{3-}, P_{tot}) and photosynthetic pigments (chlorophyll *a* and 8 diagnostic pigments) analysis.
The study area is characterized by channels of variable size, but the values of salinity and pH were similar in each channels and conform to freshwater environments. The water temperature and the percentage of dissolved oxygen, however, were highly variable in relation to the type and the size of the channels and their tree coverage. The average concentrations of P_{tot} and chlorophyll *a* (the autotrophic biomass indicator) have shown a mesotrophic condition in the tributary and an eutrophic condition in the other channels, characterized by stagnant and sediments-rich waters. The wetlands are spread between urban and rural areas and, probably for this reason, the whole area showed an average nitrate concentration that was within the attention threshold established by Directive 91/676 / EEC (Nitrates Directive). Nitrites, instead, were present with a lower average concentration. The accessory pigments with higher relative abundances were chlorophyll *b*, fucoxanthin and zeaxanthin, diagnostic of Chlorophyta/Euglenoidea, Diatoms, and Cyanobacteria, respectively. Analyzing the impact of *M. aquaticum* on the physical and chemical parameters, not significant differences between invaded and not-invaded areas were founded, with the exception of underwater irradiance that, as expected, was markedly attenuated by *M. aquaticum* coverage. These preliminary data suggest that variations in the microalgae communities are more related to the variability of the trophic status in the different channels rather than to the invasion status of the waters. However, in order to minimize the effect of the high environmental variability of the studied water courses and identify the possibile influences of *M. aquaticum* on trophic status and microalgae communities, further samples in new channels and in different season will be carried on in the future.


Fig. 1 *M. aquaticum*


Fig. 2 Invasion of *M. aquaticum* in the tributary of Lake Porta

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2.9 = VEGETATION LANDSCAPE MANAGEMENT IN THE NATURAL RESERVE OF CHIARONE, MASSACIUCCOLI LAKE BASIN (TUSCANY, IT)

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The Massaciuccoli Lake is located in NW of Tuscany (Italy) (43°50'N 10°19'E) and it is integral part of the Regional Park of San Rossore, Migliarino and Massaciuccoli. This endorheic lake and the surrounding marshlands, with over 2,000 hectares of surface, form the largest retrodunal wetland of Tuscany. Over the last century this ecological system has undergone profound changes both because of land drainage, both because of the industrial and agricultural development of the surrounding areas that have heavily polluted lake, whose waters are still affected by serious eutrophication and ecological degradation.
In order to preserve the great natural value of the lake in 1979, with the birth of the regional protected area, was established in the marshy area around the village of Massaciuccoli, the "Natural Reserve of Chiarone"(100 ha). Since 1985 the association LIPU (Italian League for Bird Protection) has obtained the management of the reserve, and its activity is directed to purposes of environmental education, environmental restoration, monitoring and conservation of specific habitats.
This report shows the results of several years of investigation on the main vegetational types and their diachronic tranformations, the presence and distribution of endemic/rare/protected species, the changes on the structure of floating islands of peats and the main actions of conservation and management of these wetlands. The vegetation landscape is mainly formed by a mosaic of *Phragmitetum australis* Gams 1927, *Cladietum marisci* (Allorge 1922) Zobrist 1935, *Typhetum angustifoliae* Pignatti 1953, *Myriophyllo-Nupharetum* Koch 1926, and a large spread of microwoods of hygrophilous phanerophytes as *Alnus glutinosa*, *Frangula alnus*, *Salix* sp.pl. A particular aspect of this lake environment are the "aggallati", floating islands of peat incurred by intertwining rhizomes of straws. Above these peatlands often develop communities of *Sphagnum* sp.pl, *Osmunda regalis* L. and *Thelypteris palustris* Schott (Fig.1).
The monitoring of these environments has highlighted the importance of ongoing management, in relation to the maintenance of certain habitats, through periodic cuttings, such as *Sphagnum* bogs, the control of exotic fauna such as *Myocastor coypus*, particularly damaging for *Cladietum* meadows and the special maintenance of the islands, whose structural peculiarity has lately been severely damaged catastrophic weather events.

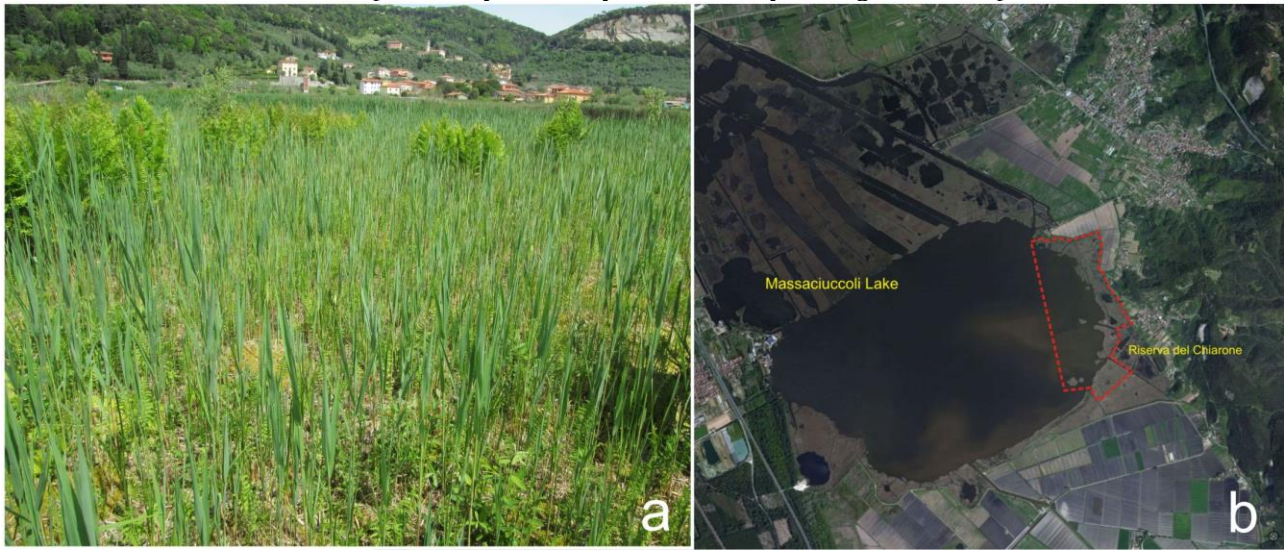

Fig.1 - (a) Peatlands communities of *Sphagnum*, *Osmunda regalis*, *Thelypteris palustris* and *Phragmites australis* (b) The Massaciuccoli lake basin and the location of " Natural Reserve of Chiarone"

2.9 = EFFECTS OF ENVIRONMENT AND DIFFERENT MANAGEMENT ON MORPHOLOGICAL TRAITS OF A CENTRAL-ITALIAN POPULATION OF *KLASEA LYCOPIFOLIA* (VILL.) À.LÖVE & D.LÖVE, A NEAR THREATENED ANNEX II PLANT SPECIES

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Klasea lycopifolia is a hemicryptophytic species with a SE-European (Pontic) distribution (1); its range extends from the southern French Alps to eastern Russia, including the Balkan Peninsula, centered on the border between Eastern Europe and Russia. The species is considered endemic to the European mountain ranges (2) and is listed in Habitat Directive Annex II (Dir. 92/43/CE and subsequent amendments) including the animal and plant species of Community interest whose conservation requires the designation of special areas of conservation. The latest Red List of the Italian Flora refers this taxon to the risk category NT, including those species next to be considered at risk and that in the absence of adequate measures may become endangered in the foreseeable future (3). In Italy, the species has a very fragmented distribution and has been recorded in Umbria, Emilia-Romagna, Abruzzo and Marche (4). This study refers to the local populations of Umbria, located in two Apenninic sites (M. Faeto and M. Pennino) that are currently excluded from the regional network of protected natural areas. The analysis takes into account some population parameters (density of individuals and flowering stems per m^2) and some macromorphologic traits (stem height, peduncle length, head height and diameter, number of stem leaves, number of green and dried basal leaves at the time of flowering). Additional macromorphologic parameters of the head are still under study. Since this species can develop rhizomes, we considered as sampling unit the functional unit represented by a ramet (5). The density parameters were measured within 40x40 cm^2 squares, located at intervals of 1 m along the major axis of each colony, having start and end in correspondence with the first and last square occupied by the species. The macromorphological parameters were measured on three randomly chosen flowering stems in each colony. In total, 33 colonies were sampled, within which the parameters of density were measured in 329 squares, while the macromorphological parameters could be measured on 93 individuals, since some colonies presented less than 3 flowering stem. For each colony, geographic coordinates and some ecological features (altitude, slope, aspect, land use) were also recorded. Statistical analysis of data was performed using the software Analyst Soft Stat Plus v2009 mac in order to assess the variability and to point out any significant different distribution of the considered traits, in relation to environmental characteristics and land use. For data with normal distribution, parametric statistical analysis was based on the application of ANOVA using Fisher's F-test; for nonparametric data, we used Mann-Whitney's U test. The first results point out some interesting differences mainly related to the different land use. In particular, the different agro-pastoral practices seem to induce significant changes in the reproductive gamic rate vs. vegetative propagation. Since this is a species whose conservation is closely linked to mountain farming (6), the present study provides useful insights for a proper planning for the preservation of viable populations of *K. lycopifolia* in a context of particular biogeographic relevance, being represented by fragments on the edge of its main area of distribution.

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2.9 = UNMANNED AERIAL VEHICLE APPLICATION FOR MAPPING INDIVIDUAL PLANTS: THE CASE OF THE NEBRODI BEECH FOREST

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The UAVs (Unmanned Aerial Vehicles) are small unmanned aircraft remote controlled electronically, commonly called "drones", which can serve as a vector to carry one or more sensors for georeferenced digital image capture. The development of these platforms is begun in the 50s for military purposes and is today successfully applied in the civil sphere with levels of technology and higher performance.
It is a remote sensing technique to the scale of detail that allows for monitoring, measuring and mapping of several environmental data in a short time.
The use of NDIR sensors (Non Dispersive Infrared sensor) for the detection of plant cover, allows discriminating plant species, mapping individual plants and evaluating the vegetative state.
This technology has been tested to monitoring the state of health of the beech forests of Monte Soro, in the Nebrodi Natural Park (Sicily), which in recent years are show signs of desiccation of numerous stumps. It was drawn up a map of the stumps of beech distinct classes of health: l) healthy plants; 2) diseased plants; 3) dead plants. The goal is to develop a useful document to manage the evolution of the process of spreading the disease.
For the case study, the apparatus used was a mini-UAV planing belonging to the "Tactic", subcategory mini UAVs (classification dell'Unmanned Vehicle Systems International Association, 2008), which are more suitable for wide-ranging surveys with predominantly horizontal flight.
It is driven by an electric engine and equipped with a navigation system with control of flight altitude, barometric pressure sensor, GPS receiver with sub-meter positioning accuracy and signal transmission, navigation system and autopilot module. The drone is connected with the ground-station for real-time viewing of flight conditions and makes all adjustments necessary for the proper configuration of the system during the flight.
The data collected are comparable in terms of spectral resolution to high-resolution satellite images (IKONOS, QuickBird, GeoEye), while ensuring a geometric resolution incomparably greater. Therefore, this technology has allowed execution speed greater than detection methods in the field, such as GPS, and costs much lower than the common methods of remote sensing (aerial or satellite images).
The application of this technology of ecological analysis in protected areas confirmed to be efficient and proved to have lower environmental impacts.

2.9 = BRYOPHYTES IN COASTAL SAND DUNES AND THEIR RELATIONSHIP WITH MAIN ENVIRONMENTAL VARIABLES

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Bryophyte vegetation in Mediterranean coastal dunes systems has been rarely studied (1, 2, 3). Bryophytes are important in arid ecosystems due to their ability to colonize different substrates, which are not suitable for vascular plants (4). They are able to stabilize dune surfaces and activate fixation process (5): in dunes and near seaside, mosses take an active part consolidating the incoherent and unstable soil, starting humification processes and contributing to the development of more complex types of soil (6). Hence, their presence has a direct influence on vegetation succession (7).
Aims of the study were i) to compile a list of bryophyte species present in sand dunes environments ii) to analyze the relationships of presence and richness of bryophytes with environmental variables such as distance from the sea, presence and type of litterfall, soil micro-morphology and habitat type.
The study was conducted in the south-west coastal area in Sardinia, in a sand dunes site included in Rete Natura 2000 (Porto Campana ITB042230). The area selected for the surveys includes several habitats listed in the Habitats Directive 43/92/CEE, such as the priority habitat "coastal dunes with *Juniperus* spp." (code 2250*). In the site, restoration activities have been made to preserve and restore dune habitats (8).
To survey the area, we adopted a simple random sampling, generating 111 random points in a GIS environment. In each point, from January to May 2015, we recorded the presence and the relative cover of bryophytes, total vascular plant cover, bare soil and/or litterfall cover in a square plot of 60 cm length.
We surveyed a total of 13 bryophyte species, with *Tortella flavovirens* being the most common species. Environmental variables influence the composition and richness of species: for example, the "2250* coastal dunes with *Juniperus* spp." hosts the greatest number of species, while the habitat "2120 fixed beach dunes *Crucianellion maritimae"* has the lowest presence of bryophytes.

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2.9 = WINNERS AND LOSERS ON ITALIAN COASTAL DUNES IN THE FACE OF RECENT GLOBAL CHANGES: THE ROLE OF THE PROTECTED AREAS NETWORK

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Coastal dune ecosystems are among the most dynamic environments on earth. Different plant communities can be found in relatively small areas resulting in a high ecological value in terms of biodiversity and landscape heterogeneity. In central Italy, starting from the postwar period up to the present, coastal dunes have experienced a loss of natural vegetation cover and a considerable modification in composition and structure in favor of the expansion of artificial, agricultural and afforested areas (1). However, there is no comprehensive picture at national scale, primarily because the kind of long-term monitoring programs needed to assess the success (or lack thereof) of conservation measures are critically missing. Despite numerous habitats are considered priorities for international conservation goals, it remains unclear whether the current network of Italian protected areas provides an adequate safeguard for these fragile environments. In this study we aim to (i) assess how the major psammophilous habitats and species in Italy have changed in cover between the late 60's and the present day, (ii) identify which ecological species strategies have been most/least successful in dealing with global change, and (iii) evaluate the efficacy of the Italian protected areas network as a conservation tool.
From the VegDunes database of Italian coastal dune vegetation (2) (part of the national database VegItaly (3)) 2,583 georeferenced phytosociological relevés from 1967 to 2011 distributed across the six most representative dune habitats were selected: beach, embryo dune, mobile dune, fixed dune, dune grasslands and wooded dune. To determine how key individual species have changed in cover over time, we selected 42 species to conduct a species-level analysis, in particular those considered diagnostic and characteristic of each habitat of the typical Italian dune zonation, and those with widespread and often locally abundant distribution. We modelled changes in cover over time using sampling year as a temporal predictor. To determine whether species' responses are influenced by their ecological strategy, we related the slope values of previous models to specific ecological indicators (plant traits, life forms and Ellenberg values). Finally, in order to establish the relationship between changes in cover of habitats and species and the state of protection of coastal dune systems, we performed a gap analysis overlapping all the relevés with the National Protected Areas and the Natura 2000 sites in a coastal buffer of 2 km. We assigned each relevés to one of three time periods (1967 to 1980, 1981 to 1994 and 1995 to 2011) and then we tested how vegetation cover has responded following the expansion of the protected areas network through a two-way ANOVA.
We observed a higher proportion of relevés in protected areas in more recent years and plant species' cover within protected relevés was actually much higher than the cover within unprotected relevés, and both protection state, time and their interaction have affected this cover increase. Between 1967 and 2011 the average cover of five of the six dune habitats significantly increased. The strongest increase in cover occurred in the wooded dune habitat and its diagnostic species, mostly phanerophytes with high values of plant height and seed mass, and species with preferences of low values of light and high values of soil moisture. On the contrary, species with high leaf density values decreased in cover (especially grasses and therophytes), mostly localized in the dune grasslands, the only habitat that significantly decreased in cover. Our results suggest that a low degree of disturbance within protected areas has facilitated the natural succession towards the more mature vegetation of inland dunes, at the expense of dune grasslands. This positive trend appears to be linked particularly to the implementation of the Natura 2000 network, which extends over large areas and contains a very high number of sites, being spread in a more capillary way and thus being more representative than the system of National Protected Areas (4).

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3 = THE ROLE OF MIRRI RESEARCH INFRASTRUCTURE IN THE CONSERVATION AND DISTRIBUTION OF MICROBIAL DIVERSITY

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The aim of the pan-European Microbial Resource Research Infrastructure (MIRRI – www.mirri.org) is to improve access to enhanced quality microorganisms in an appropriate legal framework and to resource-associated data in a more interoperable way.
MIRRI will connect resource holders with researchers and policy makers to deliver the resources and services more effectively and efficiently in order to meet the needs of microbial biodiversity conservation and innovation in biotechnology in accordance with the international legislation.
MIRRI involves more than 40 public collections and research institutes from 19 European countries (Fig 1), and all kind of "microorganisms" and derivates are included in MIRRI (Fig 2). MIRRI will provide the specific resources to facilitate discovery of solutions to the grand challenges of climate change, improved agriculture and animal husbandry for food security, bioremediation, alternative energy sources and accelerated discovery in healthcare (Fig 3). MIRRI addresses the five fundamental challenges that Europe faces regarding the proper preservation use of microbial resources and their impact on the bio-economy:
1) Fundamental uncertainty in knowledge about where the latent value in microbial resources lies. MIRRI aims to pool existing information on microbial systems, sort them and make them more accessible to users. The idea is to create a user- and quality-driven virtual center for microbial raw material, expertise and legal advice.
2) There are significant gaps in the available resources as well as gaps in the data and in taxonomic expertise that underpins resources. MIRRI will focus on the expertise and capacities of participating microbial resource centres (mBRCs). To fight the loss of expertise well-structured training in different fields of modern microbiology will be offered.
3) Microbial diversity services need to be trusted and sustained. MIRRI will provide the services, including training to access new microbial diversity to support the bio-economy, based on a quality management system that increases the reliability of available organisms and associated data.
4) Places of origin need to be able to capture value and benefit from their microbial resources. MIRRI will create a legal operational framework for access to microbial resources in full compliance with the Nagoya Protocol. This provides legal certainty in using microbial resources for the bio-economy.
5) The quality and reproducibility of microbial science need significant improvement. MIRRI supports the mandatory deposition of key resources included in the scientific literature and publicly funded research to safeguard valuable biological material for improving the credibility of science. MIRRI's aim is to establish an intensive communication with the European bio-industry, thereby identifying their needs and expectations from this pan-European infrastructure delivering microbial resources and expertise to support research and development.

Fig. 1. Partners and collaborating parties of MIRRI.

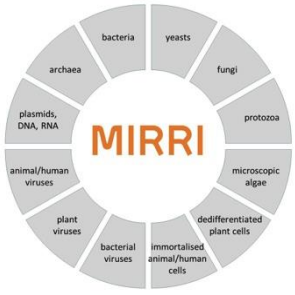

Fig. 2. Holdings covered by MIRRI.

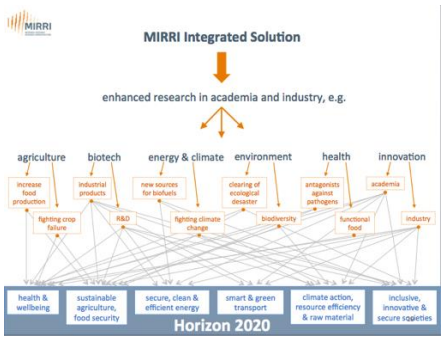

Fig. 3. Expected impact of MIRRI in different fields.

3.1 = A FIRST APPROACH TO THE KNOWLEDGE AND CONSERVATION OF CROP WILD RELATIVES IN ITALY

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The National Biodiversity Strategy, adopted in Italy in 2010 as the national instrument for the implementation of the Convention on Biological Diversity (CBD)(1), within the Work Area "Genetic Resources" has identified priority measures as the promotion of knowledge of the genetic resources and their sustainable use as well as conservation of crop wild relatives which are threatened and/or subjected to genetic pollution and/or genetic erosion.
The implementation at national level of the International Treaty on Plant Genetic Resources for Food and Agriculture (ITPGRFA – FAO), ratified by Italy with Law 6 April 2004, n. 101, has to be considered as an additional instrument to support national actions in achieving CBD objectives for conservation and sustainable use of biological diversity and particularly of plant genetic resources for food and agriculture. Furthermore the European Strategy for Plant Conservation (2), implementing the Global Strategy for Plant Conservation (3) at regional level, has identified several targets relating to crop wild relatives (CWR). In this framework the Directorate for Nature and Sea of the Ministry for the Environment and Land and Sea Protection, in cooperation with the Department of Nature Protection of ISPRA and the Department of Agricultural, Food and Environmental Sciences of the University of Perugia, has carried out a study with the aim to enlarge knowledge and conservation of CWR of crops listed in ITPGRFA Annex I and grown in Italy.
In general terms a CWR can be defined as a *taxon* with relatively close phylogenetic relationship to a given crop (4).
CWR collectively constitute an enormous reservoir of genetic variation that can be used in plant breeding (to obtain pests resistance, to adapt to climatic change, to enhance productivity) and are a vital resource in meeting the challenge of providing food security. CWR are also useful to counteract genetic erosion caused by agricultural models that employ, for a given crop, only a few commercial varieties usually genetically uniform, which are widely grown and generally characterized by high yields. Such commercial varieties gradually replace landraces and traditional varieties that show high genetic variability.
Identifying CWR at national level is a priority action in support of the intrinsic importance of the flora characterizing specific environments, recognizing both their ecological and economic value.
Taking as a basis the work developed by the University of Perugia in the framework of the European project PGR Secure (5), that has provided preliminary lists of Italian CWR (6), a survey was carried out with the aim of identifying the CWR of those crops listed in ITPGRFA Annex I and grown in Italy.
625 infrageneric *taxa* have been identified related to ITPGRFA Annex I; 209 of these *taxa* are cultivated while 416 grow only wildly. Information regarding their occurrence in national protected areas, botanic gardens and germplasm banks was collected and ordered in an *ad hoc* database that, as soon as possible, will be available in the National Network of Biodiversity (NNB) (7).

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3.1 = CROP WILD RELATIVES OF *FESTUCA* S.L. (POACEAE) IN ITALY: A NATIONAL CHECKLIST

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Festuca L. s.l. is one of the most widely studied crop genera among non-cereal grasses (1), additionally included in the European Red List (2). Hundreds of cultivars of different taxa are worldwide cultivated for forage [*Schedonorus arundinaceus* (Schreb.) Dumort., *S. pratensis* (Huds.) P.Beauv.], as turf [e.g., *F. rubra* L. s.l., *F. heteromalla* Pourr., *F. stricta* subsp. *trachyphylla* (Hack.) Patzke ex Pils, *F. filiformis* Pourr.] and ornamental grasses (e.g., *F. ovina* L. s.l. 'Glauca') (3, 4, 5, 6, 7). On taxonomic ground, *Festuca* s.l. represents a problematic critical group within Euro-Mediterranean and Italian flora (8); this aspect seemingly affected a clear definition of the crop wild relatives (CWR) of the cultivated fescues, resulting in incomplete and approximate CWR lists.
Among the different approaches available for the definition of a CWR (9, 10, 11), the lack of specific studies evidencing the level of fertility in crosses between *Festuca* s.l. crops and the Italian wild taxa, led us to adopt the Taxon Group (TG) concept introduced by (10). A consolidated infrageneric treatment of *Festuca* s.l., traditionally employed since (12), facilitated a circumscribed definition of the CWRs occurring in Italy, through the selection of members from TG2 (taxa belonging to the same series or section as crop) for *Festuca* s.s. and from TG4 (taxa belonging to the same genus as crop) for *Schedonorus*. The resulting Italian CWRs of *Festuca* s.l. are the following (number of subspecies between brackets), amounting to 48 taxa: CWRs of *F. rubra*: *Festuca cyrnea* (Litard. & St.-Yves) Signorini, Foggi & E.Nardi, *F. heteromalla* Pourr., *F. heterophylla* Lam. (1 subsp.), *F. nigricans* (Hack.) K.Richt, *F. nitida* Kit. ex Schult., *F. norica* (Hack.) K.Richt., *F. picturata* Pils, *F. rivularis* Boiss. (1 subsp.), *F. rubra* (4 subsp.), *F. trichophylla* (Ducros ex Gaudin) K.Richt. (2 subsp.), *F. violacea* Schleich. ex Gaudin (3 subsp.); CWRs of *F. ovina*: *F. apuanica* Markgr.-Dann., *F. arvernensis* Auquier (1 subsp.), *F. billyi* Kerguén & Plonka, *F. centro-apenninica* (Markgr.-Dann.) Foggi, F.Conti & Pignatti, *F. cinerea* Vill., *F. filiformis*, *F. gamisansii* Kerguélen (1 subsp.), *F. imperatrix* Catonica, *F. inops* De Not., *F. laevigata* Gaudin (2 subsp.), *F. ovina* L. (2 subsp.), *F. pignattiorum* Markgr.-Dann., *F. riccerii* Foggi & Gr.Rossi, *F. robustifolia* Markgr.-Dann., *F. ticinensis* (Markgr.-Dann.) Markgr.-Dann., *F. veneris* Gr.Rossi, Foggi & Signorini; CWRs of *F. stricta*: *F. bauzanina* (Pils) S.Arndt. (2 sspp.), *F. guinochetii* (Bidault) S.Arndt, *F. stricta* Host (2 subsp.), *F. valesiaca* Schleich. ex Gaudin (1 subsp.); CWRs of *Schedonorus arundinaceus* and *S. pratensis*: *S. arundinaceus*, *S. giganteus* (L.) Holub, *S. interruptus* (Desf.) Tzvel. (2 subsp.), *S. pratensis* (3 subsp.).
This is the first scientifically based priority list of Italian CWR of *Festuca* that are more likely to serve in breeding programs or for direct use in fodder seed mixtures. The list can therefore be used to find gaps in existing *ex situ* collections and to identify the most valuable taxa to be included in preservation mixtures, as per Commission Directive 2010/60/EU.

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3.2 = STUDY OF AN ANCIENT MAIZE FROM VALCAMONICA (NORTHERN ITALY) RICH IN CAROTENOIDS AND PHLOBAPHENES

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After the discovery of the Americas, maize cultivation became widespread in Europe and elsewhere in the world where hundreds of different landraces have been used to feed local populations. However, in the second half of the last century, American dent hybrids began to be widely grown, leading to the disappearance or marginalization of less productive traditional varieties. Nowadays, the characterization of traditional landraces can help growers to discover precious alleles that could be useful for modern genetic improvement. Furthermore, the detailed characterization of these ancient cultivars will allow the correct conservation of these open pollinated varieties (opvs), necessary for assessing their value and for further research. This work characterized the ancient pigmented "mais nero spinoso di Vallecamonica". A preliminary spectrophotometric analysis revealed the presence of flavonols, phenolic acids and in particular phlobaphenes, probably responsible for ear coloration: further investigations by HPLC to further characterize this pigment are in progress. Phlobaphenes are reddish insoluble substances synthesized in maize through the flavonoids pathway by the polymerization of flavan-4-ols. In maize this pigment is due to the presence of the pericarp1 gene (p1), a transcription factor belonging to the MYB gene family driving the accumulation of the pigment in the pericarp layer. Genetic and molecular analysis confirmed the presence of the *P1* gene in this landrace and histological analysis confirmed that the pigment is accumulated in the pericarp. In the meantime, HPLC analyses also showed the presence of a large amount of carotenoids and, probably thanks to the high amount of pigments, this maize variety showed very high antioxidant ability. Given the chronic disease prevention properties of antioxidant molecules, this variety could be of great interest also from a nutritional point of view. Characterization will allow an accurate description of this landrace with the aim of studying and preserving maize biodiversity in Europe and in particular in Italy. We are collaborating with the municipalities of Esine and Piancogno in the application process needed to register this cultivar on the National Register of conservation varieties with the aim of valorizing this ancient landrace.

3.2 = SEED PHENOTYPIC IDENTIFICATION OF ITALIAN BEAN LANDRACES (*PHASEOLUS VULGARIS* L.) BY BIOMETRIC AND TEXTURE DESCRIPTORS

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Common bean (*Phaseolus vulgaris* L.) is one of the most important grain legume for direct human consumption, since it represents a cheap source of dietary proteins. After its introduction from the Americas in the 16th century, it promptly reached a wide diffusion in Europe (1) where farmers selected for morpho-productive traits and maintained a large amount of landraces. The result of a different selective pressure, due to an adaptive evolutionary process in addition to the microclimate of cultivation areas as well as local constraints to production and different consumer preferences, is the wide differentiation of types that can be observed within the European common bean germplasm.
Several techniques, involving the analysis of morphological, biochemical and molecular markers, can be used to evaluate the germplasm collections (2). Phenotyping strategies by computer vision is a least expensive method compared to the molecular approach, and equally efficient in term of distinctiveness, with high potentialities for the characterization and taxonomic identification (3, 4, 5). Seed morphological traits were used to identify 67 Italian bean accessions, belonging to 58 Italian landraces. On each seed, an overall of 138 size, shape and texture descriptors were measured by means of image analysis techniques. The achieved data, analysed applying the stepwise Linear Discriminant Analysis, allowed to discriminate among bean landraces, also identifying the harvest year and the cropping areas. Comparative analyses were carried out to verify the possibility to distinguish seeds belonging to the same landrace but grown applying different agricultural practices (6).
Preliminarily, it was possible to discriminate three main color categories of bean seeds, with an overall performance of 99.1%. Moreover, for each of these three categories, the belonging bean landraces were identified, with overall correct identification percentages included between 94.3% and 99.7%. Following the same procedure, it was possible to assess the possibility to identify the bean landraces origin, reaching overall correct identification percentage higher than 88%. Also considering the effect of the cropping year, the cultivation region and the agricultural practices, high identification performances were recorded.
The obtained results, support the application of the computer vision system not only for the identification, classification or grading purpose, but also to define the product traceability, in order to get a “market card” for landrace beans.

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3.2 = THE 'LUCERA ARTICHOKE': A NEGLECTED LANDRACE FROM CAPITANATA (SOUTHERN ITALY)

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The 'Lucera artichoke' is a landrace of *Cynara cardunculus* L. subsp. *scolymus* Hayek, which has been cultivated for decades by local smallholder farmers in Capitanata (Puglia, Italy) around the town of Lucera (Foggia). Its flower heads have an ovoid shape, while the colour of the external bracts is green with purple nuance (Fig. 1). The length is of about 10 cm, the diameter between 7.5 to 8.5 cm and the weight ranges from 150 to 180 grams. At the beginning of 1900s, this landrace of artichoke was cultivated in many family gardens surrounding urban areas of Lucera (1). Unfortunately, these areas have been affected by intense building activity that has irreversibly subtracted from agriculture areas traditionally used for horticulture. This has determined a considerable reduction of the 'Lucera artichoke' production as well as a loss of knowledge and historical memory, since today this landrace is grown on only a few home gardens by some old farmers. On the other hand, it is possible that also for this last reason the 'Lucera artichoke' is currently grown using environmentally-friendly farming techniques and a low input of farm machinery.
The historical importance of the 'Lucera artichoke' is due to the presence of the Saracens in Puglia during the 12th century. In effect, Egidi (2) reports as a colony of Saracens was transferred into Lucera city between 1224 and 1225 a.C. by the king 'Federico II of Svevia'. Furthermore, investigations on historical texts have been carried out about Saracens customs during ancient times in Puglia. Their findings indicate that the cultivation of the *kinaria* (artichoke) was practised by the Saracens in Southern Italy at least since the 12th century. Thus, it is possible that the cultivation of the artichoke began at least two century before respect to what reported in some texts of horticulture. In this context, further study could be carried out with the aim to verify if the *kinaria* cultivated by Saracens in Capitanata during ancient times represents a possible ancestor of the 'Lucera artichoke' landrace.

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Fig. 1. A typical flower head of the 'Lucera artichoke'

3.2 = GENETIC IDENTITY OF COMMON BUCKWHEAT (*FAGOPYRUM ESCULENTUM* MOENCH) LANDRACES CULTIVATED IN VALTELLINA (CENTRAL ALPS, ITALY)

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Landraces are dynamic populations of cultivated plants lacking of formal improvement, that have historical origin and distinct identity. They are adapted to the local environmental conditions and are associated with traditional farming systems, as well as often being genetically diverse (1). The conservation of landraces is challenging because very often they have already disappeared or cannot be properly identified, but are currently of great interest for the market, thanks to their nutritional characteristics (e.g.: gluten-free).
Collection of molecular and historical data on common buckwheat (*Fagopyrum esculentum* Moench) locally cultivated in N-Italy (Alps) since XVI century (2, 3) and to date surviving among other commercial cultivars, was carried out, to identify and study the original landraces. In the Valtellina area (Teglio and surroundings in Sondrio Province) the most ancient germplasm is called "*Nustran*", whereas a French variety was introduced later and called "*Curunin*". The "*Nustran*" and "*Curunin*" selected accessions are easily distinguished on the basis of seed morphology and colour: "*Nustran*" shows brown bigger seeds and is cultivated up to 850 m a.s.l,. whilst "*Curunin*" that has grey and smaller seeds is cultivated up to 1200 m a.s.l. As plant materials of common buckwheat, we analyzed nine Italian accessions (Sondrio and Trento Provinces) along with two foreign accessions from Poland and Nepal, for a total of 174 individuals. Molecular investigations were based on a set of eight nuclear SSR marker loci.
The mean observed heterozygosity over all accessions was equal to Ho = 0.466, being significantly lower than the expected heterozygosity (He = 0.764). Recognition of a marked inbreeding rate (Fit = 0.387) and a reduced fixation index (Fst = 0.061), indicate that most genetic variation is found within populations. A significant overall gene flow among accessions was found (Nm = 3.846). Results indicated that only two of the examined accessions could be considered authentic Valtellina landraces.
On the basis of our results, we successfully developed a multi-locus marker system and identified a number of co-dominant marker alleles suitable for genetic traceability and authenticity certification of a "*Nustran*" and a "*Curunin*" autochthonous landraces of Valtellina and its food derivatives (i.e. Pasta named Pizzoccheri and Polenta taragna). This results may contribute to include this cultivar as Italian registered landraces ("Varietà da Conservazione"), by the Ministry of Agriculture (MIPAAF), as planned at EU level within the Directive 2008/62 CE. Seeds are now stored at the Pavia University Plant Germplasm Bank.

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3.2 = THE POLIGNANO CARROT: A MULTIDISCIPLINARY APPROACH FOR PRESERVING AN INTERESTING LANDRACE AT RISK OF GENETIC EROSION

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In order to conserve genetic resources in agriculture, the Puglia Region Administration (Southern Italy) planned two specific actions under its 2007–2013 Rural Development Programme. This led to several activities being carried out for some local varieties, including the so-called "yellow–purple Polignano carrot" (PC), a multi-coloured landrace grown by a handful of old farmers who act as seed savers. The PC is greatly appreciated by local people for its special taste, tenderness, crispness, flavour, fragrance and great variety of colours which ranges from yellow or deep orange to dark purple (Fig. 1), while the inner core colour ranges from pale yellow to light green (1). The roots of this landrace, which is grown near the sea, have also interesting nutritional traits as well as great potential for culinary applications and promising uses as raw material for new food products(1, 2). PC thus represents a genetic resource which could meet not only the requirements of breeding programs but also the needs of specific markets characterized by the demand for local and functional foods obtained with environmentally-friendly farming techniques. However, this interesting landrace is grown on only a few hectares by a small number of local old farmers and is regarded as a vegetable at risk of genetic erosion (3). In this context, the research activities that have integrated the Puglia Region Administration's programs may boost consumer demand for new products based on this landrace. Therefore, the possibility to obtain a real income from the PC could favour the diffusion of this crop and reduce the risk of genetic erosion or extinction. Moreover, this strategy which integrates institutional programs with research and development projects, could thus be a model to apply to other plant resources for *in situ* biodiversity conservation. On the other hand, there may be a need to implement further measures aimed at its genetic characterization, *ex situ* conservation in gene banks as well as its acceptance into the common catalogue of varieties of vegetable species as a "conservation variety". Thus, a collection of several accessions of the PC from local old farmers is actually in progress for these objectives.

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Fig. 1. A typical wooden box with Polignano carrots ready for market: it is interesting to note the great variety of the root colours which is highly appreciated by consumers.

3.2 = CONSERVATION AND CHARACTERIZATION OF LANDRACES IN NORTHERN ITALY

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Humans have started to improve crops (from their wild relatives) since the beginning of agriculture and nowadays hundreds of cultivars are selected every year to satisfy the need of higher productivity and stress tolerance (1). Indeed, while new cultivars are selected, others are at risk of extinction and progressively abandoned, especially because of minor productivity.
Landraces are local cultivars of domesticated species, which sometimes present unique and rare characteristics (2, 3) and have developed over time by adaptation to the natural and cultural environment in which they originated. They are usually more genetically and morphologically diverse than formal plant breeds because they retained many characteristics of their wild relatives and for that reason they are potential source of genetic variation for crop improvement, but also potential products for more sensitive consumers, as well as source of economic gains for farmers. Their unique characteristics, such as resistance to pests and environmental stress, and their organoleptic and beneficial qualities can be used to improve the quality of crops. Moreover, they contribute to maintain the biodiversity in agriculture. For these reasons the interest in landraces have increased at different levels, including the EU political level, with Directives aimed at revaluating the role of landraces and assuring their conservation *in situ* (on-farm) or *ex situ* in gene banks (i.e. Commission Directive 2008/62/EC; 2009/145/EC).
The main problem to be afforded with landraces is their actual availability. In many cases they have already disappeared or are very rare or cannot be properly identified due to lack of knowledge on their original characteristics, thus preventing their conservation and valorization.
So, the first step for a revaluation process is the identification of landraces in order to conserve them at least *ex situ*, in gene banks (seed banking) and the study of their distinctive traits. In fact, *ex situ* conservation in seed banks represents a very effective long term conservation form, since it requires reduced space for storage; in addition it allows to use germplasm for research and ensure the protection of threatened species and cultivars from extinction (4).
The University of Pavia has started a program for the long-term conservation of the seeds of landraces and their CWR from N-Italy in order to collect these materials on-farm (*in situ*), to study and then store them *ex situ* to prevent their loss. The germplasm is stored (dehydration at 15°C and 15% RH and then freezing in sealed containers at -18°C), according to the standards set by FAO (5), at the new "Plant Germplasm Bank of the University of Pavia" (www.labecove.it) operating in the Department of Earth and Environmental Sciences. The University of Pavia also collaborates with other germplasm banks like the Svalbard Global Seed Vault (Longyearbyen, Svalbard, Norway), the Millennium Seed Bank of the Royal Botanic Gardens, Kew (UK) and some Italian Agronomic Research Centres (CRA), to which duplicates of the collected samples will be sent to ensure the cultivars greater possibilities of preservation.

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3.3 = THE POTENTIAL OF TRUFFLE ALLELOPATHY ON MANAGEMENT OF PLANT-PATHOGENIC FUNGI IN CROP FIELDS

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Plant-pathogenic fungi in creating a variety of plant diseases stands as one of the greatest factors negatively impacting crop yield and produce quality due to the toxic chemicals of the fungicides applied to control them (1). A seemingly limitless variety and complexity of these diseases have pushed towards the development of a correspondingly large number of fungicides. Their constant application will lead to a lasting negative change in the ecosystem and provoke outbreaks of new, more resistant, strains of fungi. In recent years the study and utilization of the allelopathic activity of fungi and plant extracts have become the focus of research in an attempt to minimize the effects of the application of fungicides (2).
The promising research subject truffles *T. aestivum* Vittad. and *T. uncinatum* Chatin species complexes are naturally effective producers of phytotoxins. They are widely recognised for their burns or brûlés which are direct products of their allelopathic activity attempting to clear all competitive vegetation from the vicinity of their host plants. Secreting phytotoxic allelochemicals as exudates or leachates from their mycelia and ascocarps effectively creates the 'burnt' area of dead vegetation for which they are known (3, 4). Despite this marked ability their potential as biocontrol agents of plant parasitic fungi has yet to be revealed.
The present study was carried out to investigate the antifungal potential of methanolic extracts of *T. uncinatum* and *T. aestivum* against plant-pathogenic fungi as *Fusarium oxysporum* Schltdl., *Sclerotinia sclerotiorum* (Lib.) de Bary, *Pythium aphanidermatum* (Edson) Fitzp., and *Botrytis cinerea* Pers.
The results have shown that *T. aestivum* and *T. uncinatum* methanolic extracts, completely inhibited mycelial growth of the test fungal species at 200 µg/ml. The MGI (mycelial growth inhibition) of *T. aestivum* methanolic extract was almost always lower than that of *T. uncinatum*.
Among the plant-pathogenic fungi tested, susceptibility to *T. aestivum* and *T. uncinatum* methanolic extracts was higher in *Botrytis cinerea*, while *Sclerotinia sclerotiorum* was the least sensitive to the extracts. These observations establish in principle that *T. aestivum* and *T. uncinatum* methanolic extracts (and their allelochemicals) could be important in improving pest and diseases control, and thereby increase crop production. This research is supported by Fondazione Cassa di Risparmio di Perugia (Perugia, Italy), project code 2014.0094.021.

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3.3 = BIOACTIVE AGRO-MATRIX. EFFECTS OF ITS USE ON VEGETATIVE GROWTH, PHOTOSYNTHETIC ACTIVITY AND CROP PRODUCTION

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Sustainable agricultural production has become an important achievement in a globalized world with growing demand for food. A new integrated system of plant and animal production practices becomes a must having application that will, over the long term, satisfy human food needs, enhance environmental quality and the natural resource base upon which the agriculture depends, make the most efficient use of nonrenewable resources, sustain the economic viability of farm operations and enhance the quality of life for farmers and society as a whole. The present research aims at evaluating the effects of using agromatrices as new experimental agronomical treatment on vegetative growth, photosynthetic activity and productivity of some crops. The "Bioactive Agromatrices" are matrices derived from the agricultural cycle (rice straws, corn stalks etc.) integrated with selected viable micro-organisms (bacteria and fungi). The microorganisms were selected for their bio-stimulating effects on the plant growth and defense. Among them, species of *Trichoderma* have a direct antagonistic activity on many species of important phythopathogenic fungi and act as biofertilizer due to their nature of strong decomposers. The application of "Bioactive Agromatrices" could result in better yields, with benefits such as reducing the use of chemical fertilizers and fungicides and speeding the degradation of plant residuals. Among the various crops surveyed, we focused on corn (*Zea mays* L.), one of the most common agrarian species in the Po valley. The agromatrix composed by vegetable oil containing selected fungal strains of *Trichoderma asperellum, T. harzianum, T. atroviride* was used during the corn seed treatment in the pre-sowing phase (10 g of agromatrix per 1 kg of seeds). In order to test the bio-agromatrix effectiveness this innovative treatment was compared to a direct seeding devoid of treatment. During surveys 20 plants treated with the agromatrix and 20 control plants were sampled randomly in field. Some auxometrical parameters (plant height, length and weight of cobs, number and length of leaves per stalk) of the same samples were measured in a standardized way. In addition it was measured the photosynthetic efficiency of the plants using a fluorometer and a portable analyzer of gas exchange. In order to evaluate the seed germination, the density of the plants was estimated by linear transects of 5 meter and sample areas of 1 square meter. The results showed that the seed agromatrix treatment had a significantly positive effect on the corn production. A positive effect on vegetation growth was recorded as shown by the significant difference between the treated samples and the control samples on the auxometrical parameters. No effect was observed on plant germination and photosynthetic activity. Although further studies are needed, this research can be a starting point for future studies on "Bioactive Agromatrices" that represent innovative tools in agronomical field and contribute to the achievement of a sustainable and productive agriculture. In addition the methodology used for data acquisition is innovative in the agronomical field.

Work supported by the MIUR and Lombardy Region - "Agromatrici Bioattive" Project, (ID 30176165).

3.3 = *UPTAKE* AND INTERNALIZATION OF POLY(LACTIC-CO-GLYCOLIC) ACID NANOPARTICLES IN *VITIS VINIFERA* AND PHYTOPATHOGENIC FUNGI (*BOTRYTIS CINEREA*; *ASPERGILLUS* SP. PL.)

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In recent years, nanoparticles (NPs) have been applied in several fields of biomedicine. Only recently researchers have begun to explore the potential of nanocarriers in plant biology (1). In the near future, the development of NPs for plant research and agriculture will allow several new applications, including treatments with pesticides and fertilizers. Poly(lactic-co-glycolic) acid-based nanoparticles (PLGA NPs) are currently considered among the most promising drug carriers (2). For the first time, in this work it has been evaluated the ability of cells and plants of *Vitis vinifera* to internalize, transport and accumulate PLGA NPs, with and without an outer shell of chitosan. To visualize the cellular uptake, we synthesized PLGA NPs tagged with the high fluorescent probe coumarin 6. The fluorescence-microscopy analysis has shown the ability of PLGA NPs to cross the cell wall and the membrane of *in vitro V. vinifera* cells. It has been observed a relatively weak and diffuse fluorescence in the cytoplasm and in the nucleus, while a very intense fluorescence signal in large spherical bodies (generally 1 or 2 per cell), whose nature at present remains unknown. Cell viability test has shown that PLGA NPs were not cytotoxic in grapevine cultured cells. Through the use of fluorescent probes and inhibitors of specific endocytic pathways, it has been demonstrated that the internalization involves both the clathrin-dependent and clathrin-independent pathways. TEM analysis on cultured cells showed that PLGA NPs with a diameter ≤ 50 nm were able to enter in grapevine cells, while the larger ones remained adherent to the cell wall. Furthermore, it was demonstrated that PLGA NPs can enter in leaf tissues of *V. vinifera* through the stomata openings and that they can be absorbed even by the root and transported to aerial organs via the xylem. The cellular *uptake* of PLGA NPs has been also studied in grapevine pathogenic fungi (*Botrytis cinerea*, *Aspergillus carbonarius*, *A. niger*), suggesting that PLGA NPs could be used as vectors to deliver antifungal compounds. In contrast to what observed for *V. vinifera* cells, the PLGA NPs coated with chitosan enter quickly into fungal cells as well as those without chitosan coat. These results suggest a possible role of PLGA NPs surface charge: NPs without chitosan coat, with negative surface charge, are able to enter both in plant and fungal cells; NPs with chitosan coat that made positive the surface charge, selectively enter into fungal cells; NPs with a thin chitosan coat, showing neutral charge, are internalized only in the *in vitro* grapevine cell suspensions. PLGA NPs could provide targeted delivery systems, using the developed electrostatic reactions between residues of the wall and the surface charge of NPs. These results suggest that PLGA NPs might play a crucial role in the future development of crop management techniques, offering the possibility to deliver chemicals to specific targets in a controlled manner (3).

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3.3 = THE (IN)COMPATIBLE STORY OF *VITIS* GRAFTING: THE TRANSCRIPTION FACTORS ROUTE

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We analysed the expression of transcription factors (TFs) in two clones (TN21 and TN112) of the cultivar Touriga Nacional with different degrees of compatibility with a specific rootstock Richter-110 (R110). Our aim was to identify potential molecular markers able to identify incompatible pairs of scion/rootstock.
To do that we collected samples from two time points during the production of material to be sold by a nursery. The two time points studied were after the formation of a callus in the junction point (21 days after grafting at 30ºC) and after rooting in the field (3 month after grafting) (Fig.1 to 5). At the end of the season the TN21/R110 pair was shown to be the most compatible.
The extracted RNA from the living tissue was send for next generation sequencing using MACE (Massive Analysis of cDNA Ends) technology (from GenXPro) and the results were scrutinized to identify differentially expressed transcription factors between the two grafting pairs and between the two time points. Differential expressed TFs were selected based on False Discovery Rate (FDR<0.5) and on the ratio of the normalized expression means between groups (Fold Change), shown as the logarithm to the basis 2 of the ratio of Fold Change. An arbitrary Log2FoldChange >|1.9| was considered.
The following transcription factors meeting the considered criteria were selected for further analysis using digital PCR: Ethylene responsive factor protein 3 – ERF3; Ethylene responsive factor – ERF1A; Auxin responsive regulator protein 9 – ARR9; Auxin response factor protein 4 – ARF4; Auxin responsive protein 16 - IAA16; Lateral organ boundaries protein 4 – LBD4; Wuschel homeobox protein 4 – WOX4; and Homeobox-leucine zipper protein 6 – ATHB6.
Six of the eight tested TFs were significantly more expressed in the most compatible pair (TN21/R110) at the second time point (WOX4, LBD4, ERF3, ERF1A, ARF4 and ARF6) in relation to the less compatible pair (TN112/R110). In contrast WOX4, LBD4 and ERF3 was more or similarly expressed in the more incompatible pair in the first time point.
WOX4 (WUSCHEL-related HOMEOBOX) promotes differentiation and/or maintenance of the vascular procambium, the initial cells of the developing vasculature (1), LBD4 (LOB domain containing protein 4) is predominantly expressed in stems undergoing secondary (woody) growth (2) and ERFs (Ethylene Response Factors) are important for ethylene signalling in vascular tissues (3).
We will discuss our results in terms of the possible identification of markers for graft compatibility in grapevine.


Fig. 1 - Canes 21 days after grafting at 30 ºC.

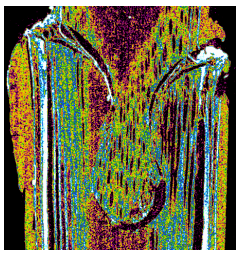
Fig. 2 - X-ray computer microtomography, 21 days after grafting.


Fig 3 - Canes in the field.


Fig. 4 - Canes in the field 3 month after grafting.


Fig. 5 - Rooted cane 3 month after grafting.

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3.3 = CANOPY CARBON ASSIMILATION RATE AND RELATED NUT YIELD IN *CORYLUS AVELLANA* ORCHARDS UNDER DIFFERENT IRRIGATION SYSTEMS

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Corylus avellana L. is one of the world's major nut crops (1) selected for its large kernels of high quality (2) with a world production totaling 888,328 Mt in 2010 (FAOSTAT 2012). To date, Turkey is the major world *C. avellana* producer (430,000 tons of dry in-shell nuts), accounting for about 71% of the world production, followed by Italy (128,940 tons), USA (34,927 tons), Azerbaijan (32,922 tons) and Georgia (31,100tons) (FAO Production Yearbook 2011). *Corylus avellana* production is limited to those areas where rainfall is enough to fully satisfy crop water requirement (3). In areas characterized by a lower annual rainfall (in the range 550-750 mm), *C. avellana* orchards require supplemental irrigation (4). In particular, water availability during the growing season affects the quality and quantity of its production (5), resulting in an increased frequency of blank nuts and a decreased kernel percentage (6). Thus, it is important to optimize irrigation management (7). Drip irrigation is the main system used to supply water to *C. avellana* orchards (8). In particular, the surface drip irrigation is the most common used technique where a supplemental irrigation is required. However, in recent years the sub-surface drip irrigation system is widely spreading, since it improves *C. avellana* water use efficiency by delivering water and nutrients directly to the root zone (9). The *C. avellana* production is in relationship with the daily net CO_2 exchange rate at the canopy level ($NCER_{canopy}$) (10).

The main objective of this research was to investigate the effects of the different drip irrigation systems on *C. avellana* nut yield. Two areas in the Central Italy (Nepi, 42° 15´ N; 12°17´ E, 251 m a.s.l.) were considered. In particular, orchards in the area A are subjected to surface drip irrigation while orchards in the area B to sub-surface drip irrigation. $NCER_{canopy}$ was estimated in the period May-September 2014 (i.e. from the end of shoot growth to nut fall). Nut size and yield were also analyzed in the two considered areas.

The results show that the two irrigation systems determine differences in $NCER_{canopy}$ being 45% higher in B than in A in May, which is the most relevant period affecting yield due to the highest carbohydrate demand (11). The 22% higher total leaf surface area of the canopy (SLT) in B results in an increased biomass accumulation as attested by the 15% higher daily photosynthetic radiation use efficiency (PhRUE). Moreover, the 13% higher daily water use efficiency (dWUE) in B than in A confirms that the sub-surface drip irrigation increases stomatal opening, thus improving the net photosynthetic rates. The higher $NCER_{canopy}$ in the B leads to a significantly greater nut yield (10602 ± 2547 g nut $bush^{-1}$) compared to 7810 ± 1847 g nut $bush^{-1}$ in A. The greater efficiency of the sub-surface drip irrigation in B is also attested by a 7% higher kernel percentage (K_p) due to a greater investment of photosynthates to carry out fruit filling. Moreover, a lower frequency of blank nuts is justified by a higher water availability, particularly during the kernel development (4).

On the whole, the results highlight that different irrigation systems determine significant variations in *C. avellana* nut yield. The sub-surface drip irrigation system improves *C. avellana* performance and might result advantageous in those areas where supplemental irrigation is required.

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3.3 = MOLECULAR TOOLS TO IMPROVE SEED PRIMING: THE 'PRIMTECH' APPROACH

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'Priming' is a pre-sowing technique widely applied by seed operators to improve seed germination and vigour, leading to high quality seedling establishment and crop yield (1). The beneficial effects of priming are due to the activation of the seed pre-germinative metabolism which triggers the seed repair responses. Therefore, primed seeds show a faster and more efficient germination, being also better prepared to overcome stress factors and adverse climatic conditions (2, 3). Based on these premises, it is clear that a deeper understanding of the molecular mechanisms involved in the pre-germinative metabolism, can result in valuable tools for priming improvement. Molecular biology techniques can be applied to seeds in order to monitor, throughout the priming treatment, their ability to repair DNA damage and withstand oxidative injury (2). This analysis produces specific molecular profiles, used to establish hallmarks of the seed response to priming, therefore supporting the development of improved protocols to be used in the Seed Industry. These are the main goals of the PRIMTECH project ('Advanced PRIMing TECHnologies for the Lombardy Agro-Seed Industry', www.unipv.it/primtech), funded by Lombardy Region and Cariplo Foundation. Research activity within the PRIMTECH project focuses on horticultural crops and bread wheat with the common goal of establishing reliable molecular indicators of the seed response to oxidative stress during early imbibition.

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3.3 = ALLELOPATHIC EFFECTS OF *CYNARA CARDUNCULUS* VAR. *SYLVESTRIS* LEAF EXTRACTS ON WEED SEED GERMINATION

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It is known that the presence of weeds causes serious losses to the agricultural production, both in quantitative and qualitative terms. Putnam and Duke (1) were the first ones to assess the possibility of using allelopathic crops for weed management in agriculture, minimising the serious problems of environmental impact, improving the toxicological profile of the food plants and ensuring the preservation of biodiversity in cropping systems. Furthermore, there is an increase in the worldwide consumption of products from 'organic agriculture', which does not make use of any synthetic herbicide. As weeds are very good colonizers, reproduce themselves faster, produce a large number of small seeds with very prolonged viability in soil and survive in the most adverse situations (2, 3) becoming part of the persistent soil seed bank (4), they are in general more competitive than other plant species.
Synthetic herbicides are generally used in low concentrations to control weeds. Their indiscriminate use can have a negative impact on the biological communities as well as on the human population. For an integrated weed control approach, the potential use of natural, biodegradable, water soluble and halogen free molecules belonging to the allelochemicals family can have several advantages over synthetic herbicides, i.e. higher activity at low concentrations and reduced negative effects on the environment (5, 6).
The aim of the study was to investigate the effect of allelochemicals of *Cynara cardunculus* var. *sylvestris* (wild cardoon) plants on the seed germination of two common weeds (*Amaranthus retroflexus* L. and *Portulaca oleracea* L.), to simulate some of the factors that could occur in the environment. Germination bioassays were performed using 5 mL of aqueous leaf extract at 40 and 80% (v/v) and distilled water (0% control) to humidify a double layer of sterilized filter papers (Whatman No. 2). Petri dishes were stored in incubators at the optimal temperature of 35°C in continuous darkness. During the counting process, seeds were manipulated under a green safelight (490–560 nm); seeds with a 2-mm radicle elongation were considered as germinated and removed from the Petri dishes. Analysis of variance was performed using an Anova software. Percentage data, before Anova analysis, were square root transformed to increase homogeneity of error variances.
The germination rate dropped by increasing aqueous leaf extract concentration of *C. cardunculus* var. *sylvestris* which also induced significantly greater reductions in seed germination than the control for *A. retroflexus* (37% *vs* 94%) and *P. oleracea* (17% *vs* 61%). Reductions in mean germination time (MGT) and substantial improvements in the seed germination of *A. retroflexus* and *P. oleracea* were observed for seeds treated with control and allelochemical solutions (3 and 4 d *vs* 5 and 8 d). The interaction between the analysed weeds and the aqueous leaf solutions used proved to be less significant. There was no significant difference between the different concentrations (40 and 80%) utilized during biological assays.
On the basis of such results, allelopathy should be further evaluated and used to screen allelopathic plant species. Our research demonstrated that wild cardoon contains substances with inhibitory effect, which could reduce the percentage of germination of two common weed species in laboratory conditions, providing novel and biologically active compounds to control weeds in different agro-ecosystems. Further studies on the phytotoxic activity of natural compounds could lead to the discovery of new herbicidal active ingredients and to the implementation of environmental friendly weed control strategies.

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3.3 = NOVEL MOLECULAR HALLMARKS OF SEED IMBIBITION

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The need for increasing seed quality is currently regarded as a priority, necessary to face the demand for high standards in the seed market (1, 2). Within this context, cutting-edge research will help improve knowledge on the complex molecular aspects of germination (3). Dehydration and rehydration during seed development and germination result in enhanced oxidative stress and DNA damage accumulation, however during seed imbibition DNA repair pathways are activated. Such a response requires chromatin remodelling and local changes of nucleosome structure. Studies concerning chromatin remodelling in seeds are strictly related to nucleosomes condensation occurring during seed maturation (4). The present investigation, carried out in collaboration with the Petunia Platform (http://flower.ens-lyon.fr/PetuniaPlatform/PetuniaPlatform.html) focuses on the *Petunia hybrida PhTRRAP* (TRansformation/tRanscription domain-Associated Protein) gene. The TRRAP protein, which plays an intriguing role as chromatin remodeller in animals, is poorly investigated in plants. TRRAP is a subunit of Histone Acetyltransferases (HATs) but it also associates with DNA damage response proteins, such as MRN complex and several transcriptional regulators (5). TRRAP relaxes the chromatin allowing to reprogram gene transcription and to make the DNA damaged sites more accessible to the DNA repair machinery. In order to find out more information about this function and the putative correlation to seed viability, we are currently exploring the *PhTRRAP* gene expression profile in the early phases of seed germination in *Petunia hybrida* in presence/absence of abiotic stress.

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3.3 = PLANT RED LISTS IN ITALY: ADDRESSING THE THREATS TO ENDEMIC PLANTS FROM AGRICULTURE

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"The New Red List of Italian Flora" project, promoted by the Ministry of Environment (MATTM), in collaboration with the Italian Botanical Society, started in 2013 with the aim to update the conservation status of Italian plant species (1, 2). So far, more than 700 vascular plant species were evaluated according to IUCN Red List criteria (3). All the species listed in the Directive 92/43/CEE and Bern Convention (4) and more than one third (484) of the about 1400 endemic taxa (5) were assessed. Data were organized in a database including distribution, population trends, and main threats categorized according to the IUCN threats classification scheme (6) and identified by expert-based observations.
Threats in the major category of "natural system modifications" and "human intrusion and disturbance" are the most frequent, however, an important threat is also represented by "agriculture", with more than a quarter of endemic species (129, 26.6 %) threatened by agricultural expansion and intensification, including farming, silviculture and aquaculture practices. In particular, 66 out of 129 (51 %) endemic species resulted affected by farming and grazing that are the main threats between those connected to agriculture; 20 (15.5 %) species are threatened by annual and perennial non-timber crops and 16 (12.5 %) species are threatened by timber crops. For the remaining 27 species only generic threats linked to agricultural practices could be identified. Intensified livestock farming, and especially intensive grazing activities is the main threat also for Policy species at European level (7). However, it is worth noting that intensification of agriculture especially in lowland areas as a threat for the endemic flora of Italy is only one face of the coin. In fact, change in land use and land abandonment, especially in mountain areas, is also a threat for biodiversity, highlighting the positive effect of sustainable agricultural practices.
A general trend is that the least-intensive systems support greater species richness. Consequently to improve conservation of plant diversity, sustainable-farm systems (e.g. extensive grazing, organic and integrated farming, conservation agriculture) must be rewarded for the ecosystem services performed by their good practices, using instruments embedded by agricultural policies like Common Agricultural Policy (CAP), possibly in strict connection with Natura 2000 network, which is largely represented in EU also in agricultural areas.

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3.3 = PROTOPLAST LARGE-SCALE ISOLATION FROM POTATO: AGRONOMIC AND BIOTECHNOLOGICAL APPLICATIONS

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Optimization of protocols for the large-scale isolation of viable protoplasts, the related *in vitro* culture and plant regeneration procedures represent the starting point for research programs that aim at the genetic improvement of crops worldwide. Protoplasts are used to carry out somatic hybridization and genetic engineering techniques (1). Besides this, the protoplast-based technology has recently become a valuable tool in cell-based experiments for validating the output of innovative 'omics' approaches and genome editing (1).
The tuber crop potato (*Solanum tuberosum* L.) has high economic, agricultural and nutritional relevance. It can adapt to different climates, although extremely sensitive to freezing and drought, while pests and fungal diseases can severely affect yields.
In the present work, two potato varieties (*Solanum tuberosum* cv. 'Mozart' and *Solanum tuberosum* cv. 'Desirée') were tested for their ability to produce protoplasts having regeneration ability. To this purpose, tuber-derived explants were cultured *in vitro* on different growth media until the optimal combination of nutrients (P1 medium) for effective plant regeneration, and rooting was sorted out. A further optimization of the growth substrate in terms of phytohormone composition allowed us to obtain the P2 medium. Potato shoot explants transferred to P2 medium resulted into consistent expansion of the aerial parts and production of appropriate amount of leaf biomass for protoplastization. Viability of potato protoplasts from the *S. tuberosum* varieties was assessed with histochemical staining. The protocols developed from this study will be used for the production of pre-breeding material having increased tolerance to biotic and abiotic stress, suitable to implement potato yields in the Kurdistan Region-Iraq.

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3.6 = MORPHOLOGICAL AND AGRONOMIC TRAITS OF A NATURALIZED POPULATION OF PARSLEY [*PETROSELINUM CRISPUM* (MILL.) NYMAN] COMPARED WITH THREE COMMERCIAL CULTIVARS

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Parsley [*Petroselinum crispum* (Mill.) Nyman, Apiaceae] is a biennial herbaceous plant, naturalized in much of Europe and cultivated worldwide as an aromatic and edible plant. The origin of the species is uncertain, but perhaps South Est Europe or West Asia (1); in Italy it is present as cultivated or naturalized (2). A naturalized population of parsley is reported in the province of Trento, Italy, in the locality Cronil within the municipality of Ala (3). The presence of this parsley population is known since long times by the people living in the neighboring localities, who traditionally use to collect its seeds and to cultivate it in their homegardens for personal use, appreciating it for its strong aroma, distinguishable by the one of the commercial varieties. In this study the morphological characteristics and the main agronomic traits of the naturalized population of parsley were compared in a field trial with that of three commercial cultivars, two smooth leaf and one curly leaf type: information about the distinctness, productive capacity and composition of essential oil of the naturalized population could provide the necessary cognitive basis for its improvement by the starting of a breeding program. Morphological and agronomic traits were recorded twice, one day before starting the first and second harvest, on ten randomly chosen single plants per replicate. A modified CPVO technical protocol for performing distinctness, uniformity and stability tests of parsley (4) was used for the morphological characterization.
The naturalized population belongs to the smooth leaf type for the absence of curling, but differed from the other two smooth leaf type accessions for a weaker undulation of the leaflet margin and for the attitude of leaves, prostrate instead of erect. Highly significant differences were detected among accessions for all the quantitative traits. The naturalized population was statistically different from the other two smooth leaf accessions for the lower height of plant and the smaller length of petiole. These characters, together with the prostrate attitude of plant, are undesirable for making more difficult the harvest of plants. With respect to the differences between harvests, some of the quantitative characters were higher during the second harvest, due to a greater development of plants. No interaction between accessions and dates of harvest was observed for any of the morphological traits. Highly significant differences were detected among investigated accessions for the yields and the other agronomic traits recorded, with exception of the percentage of the missing plants, which was very low and not different among the accessions. The naturalized population had the lowest yields both of aerial parts and leaves, and the highest essential oil yield, resulting statistically different from the other smooth leaf type accessions for the aerial part yield and from all the other accessions for the yields of leaves and essential oil. The naturalized population resulted statistically different from the other accessions also for the essential oil content of the leaves calculated as a percentage of fresh weight, which was more than the double than the cultivars, and higher than those reported by the majority of previous authors. The naturalized population had the highest content of dry matter too, both of entire plant and leaves. This result suggests its adaptation to the dry conditions of its place of origin and is evidently related to the higher content of essential oil.
As a conclusion, the naturalized population differed from the smooth leaf type varieties evaluated in this study for some morphological characters that easily permit its identification. The small dimensions of the plant and especially its prostrate attitude are however two undesirable characters which could make more difficult its harvest. These characters, together with the low yields of leaves and aerial parts, could be improved in an eventual breeding program, which could be justified by another interesting feature of the naturalized population, that is the high yield of essential oil extracted from its leaves.

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3.6 = MORPHOLOGICAL VARIABILITY AMONG NATURAL POPULATIONS OF *CHAMAEROPS HUMILIS* L. GROWN EX-SITU

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The European fan palm (*Chamaerops humilis* L.) is a pivotal species of the thermo Mediterranean vegetation belt of southern Europe, where it plays an important ecological role. Nonetheless, its distribution in natural stands is reducing due to anthropic disturbance. Moreover, gene flow from cultivated plants to natural stands is likely occurring, with potential drawback of genetic pollution. This has induced the European community to call for its protection in Special Areas of Conservation. However, information about the morphological variability of natural stands is still fragmented and this hampers the outline of the morphological traits important to discriminate such species at an intra-specific level.
In a previous experiment (1), we showed that natural Sicilian *Chamaerops humilis* populations likely arose from secondary adaptation to the environment of a common ancestor. In those populations, many morphological traits were related to environmental characteristics. Aim of the present work was studying the morphological variability among *Chamaerops humilis* var. *humilis* plants coming from 6 natural Sicilian populations and grown under ex-situ conditions. In addition, an Algerian *Chamaerops humilis* var. *argentea* André population was added as a outgroup. Seeds from the natural stands were germinated and allowed to grow until adult stage (5 years). At the beginning of the adult age, 15 morphological descriptors important to discriminate among populations as described in Giovino et al. (1) were measured. Correlations among ex-situ morphological traits and between these and climatic conditions of the collection sites were studied. Finally, standardised data of mutually uncorrelated morphological traits (2) were used to plot populations after a canonical discriminant analysis (CDA) in which population was designated as class and plants from the same population as interval variable. Many ex-situ morphological variations strongly correlated with climatic conditions of the collection sites. In particular, trunk height and diameter increased at reducing summer and fall water availability and hair density increased at reducing aridity and maximum winter temperature of the collection sites. Only 5 morphological descriptors were retained for the further CDA, including trunk height, crown height, depth of leaflet split to the total lamina length, thorn density, and hair density. Trunk and crown height mostly discriminated populations in the CDA. CDA discrimination and relationships among morphological vectors were similar to those observed in (1).
These results suggest that such morphological traits are efficient descriptors of the European fan palm at an intra-specific level and can be used in structuring efficient conservation programmes under both in-situ and ex-situ conditions.

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3.6 = ECTOMYCORRHIZAL FUNGAL DIVERSITY IN A FOREST PLANTATION OF EXOTIC CONIFERS IN SARDINIA (ITALY)

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Cedrus atlantica (Endl.) G.Manetti ex Carrière, *Pinus nigra* J.F.Arnold and *Abies alba* Mill. are widespread in many southern and/or Mediterranean mountain forests but are exotic in Sardinia. These conifers were introduced in Sardinia through the last century for reforestation purposes but no specific data on their ectomycorrhizal (ECM) fungal community are available in literature. The present study aimed to assess the diversity of ECM fungal community in a mixed plantation of *C. atlantica*, *P. nigra* and *A. alba* where natural regeneration of *A. alba* and *Quercus ilex* L., the unique indigenous tree species, was only present. The study site is located on the Limbara mountain complex in northern Sardinia (Tempio Pausania, Italy) at 1262 m a.s.l. Five plots homogeneous for host plant composition were selected for root sampling. ECM morphotypes from each host plant species were morphologically characterized and fungal symbionts were identified by comparing their ITS sequences with those present in public sequences repositories (GenBank, UNITE). The whole ECM fungal community on the roots was composed by 46 species of which more than 86% were Basidiomycetes. Thelephorales accounted for 44% of the total observed ectomycorrhizas, distributed among 5 genera and 14 species. Six and 5 ECM fungal species resulted specialist for conifers and *Q. ilex*, respectively. *Quercus ilex* showed the highest species richness, Pielou and Shannon index values whereas *P. pinea* had the lowest values. Multivariate analysis (CAPC) showed that ECM fungal community observed on *Q. ilex* was separated from those of other host plants. However, most of the dominant ECM fungal species colonized both conifer and *Q. ilex*. Probably these fungi could play an important role in the natural process of reversion of this forest exotic plantation to Mediterranean evergreen oak wood.

The increasing studies on ECM fungal communities in different forest ecosystems could be useful to unravel the ecology and biology of the ECM fungal species as well as to improve the understanding of forest ecosystem functioning, evolution and management.

3.6 = DOES SEED PRIMING INCREASE THE SEED LONGEVITY OF WILD ALPINE PLANTS?

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The longevity of seeds is crucial for their *ex situ* conservation and future uses. Different seed priming techniques have been developed to increase the seed viability, uniformity and longevity in crop plants. However, the effects of priming on seed longevity of wild species remain poorly investigated (1). In this regard, seeds of alpine plants are known to be short-lived in storage (2), indicating that long term conservation might be difficult for these species. Thus, the aim of this study was to investigate different techniques to restore viability in storage (i.e. osmopriming) of 10 alpine species representing different plant families from the Italian Alps.
Mature ripened seeds were collected randomly from at least 20 individuals for each species in September 2014. Seeds were cleaned and stored under dry room condition (i.e. 15 °C, 15% RH) until use. Seeds of each species were exposed to controlled ageing test (CAT, 45^{o}C and 60 % RH) and regularly sampled for germination testing. At the 15th day of ageing, a further subsample of seeds was removed from CAT and osmoprimed at 0, -2.5 and -10.0 MPa for 48 hours in dark at 25^{o}C. Primed seeds were washed with tap water, air dried and then returned to CAT. Hence, one sample of 50 osmoprimed seeds was regularly sampled for germination testing. Probit analysis was carried out on the data by using GenStat 11.1 to estimate the viability to fall to 50% and seed survival curves fitted to the data using the equation of Ellis and Roberts (3).
Across all species, seed viability declined with increasing duration of the ageing treatment, with a wide variation in the time taken for viability to fall to 50 % across the species (i.e. ranged between 3 to 15 days). Consistently, the effect of priming also varied across the species. Primed seeds (all treatments) improved the survival/longevity of six species, with -10 MPa providing the greatest benefit for subsequent longevity/viability. Our results confirmed that alpine plants produce short lived seeds in storage, but showed that osmopriming may have an important rejuvenating effect on aged seeds in several of them. Such technique may therefore have important implications to maintain high quality *ex situ* collections of alpine species. This experimental activity is a part of the research carried out in the project NASSTEC (ITN Marie Curie Action- grant no 607785).

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3.7 = ARCHEOBOTANICAL STUDY OF TRADITIONAL AGROECOSYSTEMS BASED ON SEM-EDX ANALYSIS OF BURIED PHYTOLITHS

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The FP7 Project "MEditerranean MOntainous LAndscapes: a historical approach to cultural heritage based on traditional agrosystems (MEMOLA)" (http://www.memolaproject.eu/it) is studying past landscapes using ancient soil horizons as archaeological records. The project aims at evaluating the biodiversity of no longer existing environments, in order to reconstruct the past agroecosystems. One of the approach used in the study consisted in the selective sampling of buried paleo-soil horizons for the search of "testimonials" or useful pedo-archaeological "indicators" that can tell something about past environments and peculiar ecosystems no longer present. To this purpose it was decided to start extracting phytoliths from soil samples analysing them. Phytoliths are produced in and between the cells of living plants (1). They consist of biogenic silica, also referred to as opal, namely a hydrated amorphous form of silica (SiO_2 nH_2O) formed by complex inorganic polymerization processes. Once the portion of the plant containing phytoliths dies, they are released in the environment due to the decomposition of the organic matter. In general, phytoliths are not transported over long distances because they are relatively "heavy" particles (as opposed to pollen, for instance). Depending on the type of activities carried out by the human group or individual, the natural input of phytoliths can be more or less important in the constitution of the sedimentary assemblage. All human activities leave behind plant organic matter and phytoliths. Phytoliths have different shapes and size and can often have taxonomical significance. They can therefore be good indicators of past vegetation cover and environmental conditions, providing evidence of vegetation change if studied in stratigraphical records. They could represent a first step to explore the biodiversity of recent and past- environments (2). Phytoliths were studied from buried and charred plant micro-particles, fragments of epidermal tissues, that were recovered during the soil sieving procedures (Figs 1, 2, 3). Samples were observed under a scanning electron microscope (SEM). SEM analyses were performed using, in Variable Pressure mode, at 20 keV, an EVO 50 Scanning Electron Microscope (Carl-Zeiss, Oxford, UK) fitted with detectors for both Electron Backscattered Diffraction (BSD) and Variable Pressure Secondary Electron Scanning (VPSE). VP-SEM permitted the observation of micro-sections of soil and sediments and powdery samples extracted and selected to contain phytoliths without metallisation. It therefore represented a non-destructive technique for MEMOLA pedo-archaeological studies, which allowed to preserve small samples for their further characterisation.

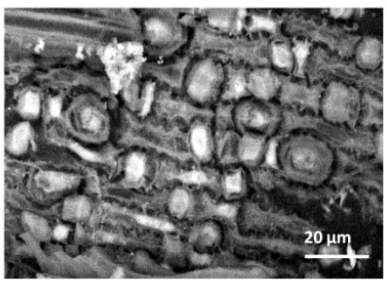

Fig. 1. SEM VP, BSD. Buried charred plant leaf fragment with phytoliths.

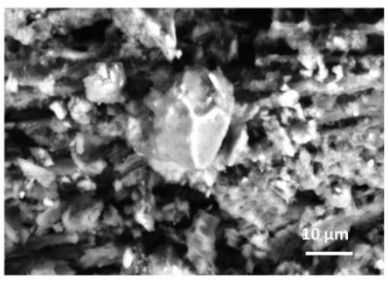

Fig. 2. A parallepipedal bulliform cell phytolith.

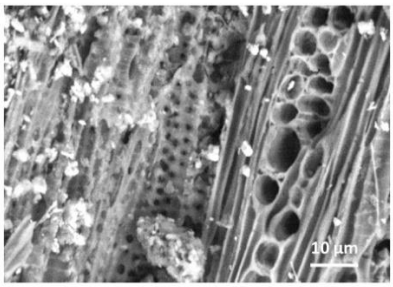

Fig. 3. SEM VP, BSD. Charred wood.

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3.7 = ANCIENT DNA EXTRACTION FROM MEDIEVAL GRAPE SEEDS

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The study of plant archaeological remains is fundamental for understanding and reconstructing the origin and the diffusion of agronomic species. The ancient remains of grape (*Vitis vinifera* L.) mainly consist in seeds, whose DNA usually is not well preserved. Morphological and morphometric approaches, recently based on image analysis techniques (1), were used to distinguish cultivated from wild grapes, and to recognize and discriminate different morphotypes (2). Any doubts should be clarified by ancient DNA (aDNA) extraction and sequencing, a growing field of research that gives reliable answers in different study of paleontology.
The recovery of genomic DNA from archaeological remains represents a problem for the diagenesis of relic DNA molecules, that often consist of small fragments.
Ancient medieval grape seeds found in an urban excavation in Palermo were morphologically and morphometrically analyzed (3, 4). In order to perform a molecular investigation the attention was also focalized to set up a suitable DNA extraction and on to define the most appropriate molecular markers.
Ancient DNA extracted using both phenol-chloroform method and CTAB protocol resulted in very small concentration quantified in a range of 8 to 20 ng/µl by Bio-spec nano (Shimadzu). PCR products of *rbc*L gene were of about 100 bp in length and the sequences are under analysis.
The study is in progress, focusing on other target sequences to amplify and subsequently to compare with the old and historic cultivated grapes of Sicily.

Acknowledgments: The biological samples analysis was attributed to L. Sineo with protocol n. 2861- S16.5 (Soprintendenza per i Beni culturali e ambientali – Palermo).

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3.7 = EFFECT OF PLANT AGE ON SEED ASPECT OF NATIVE SARDINIAN GRAPE CULTIVARS

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During the first year of the project "Origin, characterization and conservation of autochthonous grapevines of Sardinia", funded by "Regione Autonoma della Sardegna", interesting preliminary results were obtained.
The main aim of the project is to use some of seed biometric features, measured with computer-aided image analysis techniques, to characterize the studied *Vitis vinifera* L. cultivars (1, 2, 3). The recorded data were used to implement statistical classifiers able to discriminate among the cultivars, and comparing them with archaeological seed lots founded in Sardinia, in order to improve the knowledge about the origins of viticulture in Sardinia (4, 5, 6). During the first year, some sample bunches from old grapevines of different ages were collected in the North-Central Sardinian territory. In particular, they are from Bono (NU), Oliena (NU), Atzara (NU) and Pattada (SS). Seeds were extracted from the bunches and cleaned. Digital images of samples, consisting of 100 seeds (when available) for each seed lot, were randomly disposed on a flatbed scanner, acquired and processed for the morpho-colorimetric analysis. A total of 137 phenotypical descriptors and the mean seed weight was measured in each seed.
The data were statistical analyzed, applying the stepwise Linear Discriminant Analysis method (SPSS Inc. 2006). Studying plants from 10 to 80 years old, the analysis showed that the seeds of the cultivar "Muristellu", reduced their dimensions, increasing their luminosity and minimum diameter, during the time. Moreover, the system allowed to correctly identify the compared seed lots with percentages of ca. 96%.
Analysing the seed lots of the cultivar "Arvesiniadu", collected from plants from 20 to 120 years old, it was possible to underline a high level of similarity. Nevertheless, the studied seed lots showed a significant variation in seed colour intensity and seed roundness, both directly proportional with the age of the grapevines.
These preliminary results prove that the age of the plant can affect the seed aspect. This is probably due to the modified agronomical practices during the last century and in particular in the recent decades. Anyway, these achievements could be useful in order to better understand the origin of viticulture in Sardinia.
This contribute has been carried out thanks to the research project "Origine, caratterizzazione e conservazione dei vitigni autoctoni della Sardegna" developed at the enterprise "Azienda Gostolai S.A.S. di G.A. Arcadu and C., Oliena (NU)", by a research grant financed from the resources of P.O.R. SARDEGNA F.S.E. 2007–2013 – Obiettivo competitività regionale e occupazione, Asse IV Capitale umano, Linee di Attività l.1.1. e l.3.1.

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3.9 THE CHILD GARDENERS: SOWING SEEDS, HARVESTING KNOWLEGE

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One gardener and 10 m^2 of land in the Botanical Garden of Florence; 5 classes and 5 teachers of the primary school of the "Scuole Pie Fiorentine"; 68 child gardeners: these are the numbers of "The Child Gardeners" project that offers gardening and observation activities which take place during the entire school year and has been going on for several years.
Aims of the project:
• Find out what lays behind a vegetable: to enhance the effort behind cultivation so to appreciate more the results;
• Observe the seasons and the weather patterns (every cultivation practice at the right time!);
• Observe the diversity (different varieties of the same vegetable or the same aromatic plant);
• Promote organic farming (not only is it possible, but you will get great results!);
• Talk about food (what feeds the world, what do we eat);
• Learn how to recognize what we put on our tables (seeds or fruits? bulbs or tubers? Which plants are flour or sugar from?);
• Make the children promoters of knowledge in their families (eg. the little known *Stevia* plant);
• Actively involve all children in gardening operations, although at different times of the year or in subsequent years.
How the project is carried out:
After the soil processing, which is done by the gardener on the dedicated 10 m^2, the children carry out the spudding, preparation for sowing, transplanting or direct planting, weed removal, irrigation and harvesting activities.
The choice of plants to be cultivated in the Botanic Garden follows certain criteria:
• Innovative plants (eg. perilla, stevia);
• Ancient plants (eg. marzuolo wheat, ancient variety of the Pistoia mountains);
• Aromatic (at least 3 varieties for each one, eg. large-leaved parsley, curly and normal);
• Little known plants but widely used in other cultures (eg. coriander):
• Vegetables (eg. beans, salad, carrots, potatoes).
The gardening activities are alternated with observation done in the Botanic Garden which covers different topics:
• Morphology: roots, stems, flowers, leaves, fruits, seeds;
• Distinction between herbs, shrubs, trees;
• Weeds used in cooking;
• Poisonous plants, medicinal plants;
• Plants and geography;
• Plants and history;
• The evolution of plants.
The observation activities in the Botanic Garden is followed by classroom sessions on various topics with practical examples: spontaneous salads; seeds: forms and scattering; soils: which are the most suitable for different crops.
The "Scuole Pie Fiorentine" believe that this project is a highpoint experience to be pursued in coming years: in fact all the children have the opportunity to follow all stages of cultivation, alternated with the knowledge of plants and food, and they can repeat this experience for 5 consecutive years. The school remits the Botanical Garden for the purchase of seeds, plants and the material needed for this activity.
Conclusion: "The Child Gardeners" project, which has been held for several years in the Botanic Garden, has allowed us to: interlace the wisdom of cultivation with the theoretical knowledge in order to establish a more intense relationship with the environment; increase children's awareness on the great themes of agroecology, climate change, uneven distribution of food resources; spread the culture of botany through very concrete examples; encourage a critical approach in future consumers; highlight the connections between different disciplines, from history to biology, from horticulture to ecology of nutrition.

3.9 = THE HERBARIUM OF MICHELE PADULA: AN INVENTORY OF THE MATERIALS FOR A "FLORA OF CASENTINO" NOW CONSERVED IN THE HERBARIUM CENTRALE ITALICUM (FI)

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In the years between 1973 and 1997, the forests of the Casentino area in Tuscany were under the administration of Michele Padula, head of the office of the "Azienda di Stato per le Foreste Demaniali di Pratovecchio" and already professor of Forest Botany at the University of Firenze. Along with his long commitment in the State Forest Body, Padula was as an enthusiastic botanist who conducted several floristic and vegetation studies. During his years in Casentino, he started to collect plants to build an Herbarium dedicated to this area, a work that was then intensified after his retirement in 1997. The collection was aimed to serve as the main basis for a "Flora of Casentino". Due to his sudden death in June 2011, the herbarium (now including 3500-4000 specimens) could not be completed but still represents a very important resource for the botanical knowledge of the Casentino area. It was therefore donated to the Natural History Museum of the Firenze University, for appropriate conservation and to facilitate further botanical studies on the area. Aware of the scientific value of this collection, the "Ufficio Territoriale per la Biodiversità di Pratovecchio" and the Central Italian Herbarium have recently promoted the inventory and musealization of this collection. The first step consisted in the selection of the plant material from Casentino, separating it from the general collection, which included specimens from other Italian regions. The specimens from Casentino had already been partly catalogued by Padula himself in a database, and intercalated in a central collection together with other specimens of different geographical provenances. Another part of the collection was instead temporarily conserved in homogeneous folders, waiting for a more accurate study and final accomodation. By checking the descriptive label of the herbarium sheets and the notes given on the overall content of the folders, the specimens from Casentino were placed together to form a homogeneous collection. Afterwards, the Casentino specimens that were outside the collection "*Flora* del Casentino", and still not catalogued in a database, were analyzed for their temporal and geographical distribution. Using the geographic database of Tuscany and the "Geoscopio" portal, it was possible to make the georeferentiation of all the ca. 1100 localities reported on the herbarium labels by means of Google Earth. This provided a general view of the territorial coverage of the Padula collections, and consequently of the location of the potentially interesting areas for future field researches because still poorly (or even not) explored from a botanical point of view. From a first analysis of these geographical data, however, the Casentino area appeared to be fairly well covered, except for small areas that had been possibly explored by Padula in previous times. The specimens from these areas may in fact be part of the bulk collection of Casentino that was not considered in this first step of the work. The areas of Pratovecchio and Chiusi della Verna (Arezzo) were the most collected. The temporal range of the collections is well distributed across the months of the year, and in line with the seasonal cycles of the vegetative activity. It is interesting to note that about 80% of the material was collected in the time span 1997-2011, although the first specimens date back to the year 1966. After the first sorting phase, the work consisted in the taxonomic and nomenclatural analysis of the collection, in order to facilitate the management and accommodation of the herbarium material. Waiting for further examination of the correspondence between the names adopted by Padula and those currently in use, all recent sheets have been sorted and distributed on taxonomical grounds. Cataloguing the specimens was a further step of this work. As mentioned before, Padula created a "Flora del Casentino" database where he wrote all essential information for each taxon in his collection. In order to facilitate accessibility and use of these data, this database file was converted into an Excel sheet and rearranged in a way to have a single row for each *record*. To sum up, this work is still in his initial phase as also the analysis of the temporal and geographical range of the collections is still to be completed. Nevertheless, it already allows the access and much easier use of the plant material in the "Erbario del Casentino" and the associated valuable information in the database. To complete the inventory and musealization of this collection, as well as to enhance its scientific value, it will be important to examine the names adopted and to make it consistent with the nomenclature currently in use, besides the taxonomic revision of some "critical" specimens. The last step will be the mounting of the specimens on the standard herbarium sheets, their labelling and arrangement in a sound taxonomic order.

3.9 = THE CITRUS COLLECTION AT GIARDINI BOTANICI HANBURY: HISTORICAL INVESTIGATION AND SUSTAINABLE CONSERVATION

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In 1867, Sir Thomas Hanbury bought a large property on Cape Mortola, Riviera, at the border between Italy and France, to create a botanical garden that soon became world famous. The land was cultivated and within the property, there were crops of *Citrus* (1, 2) and olive trees, which he added to, along with a variety of exotic fruit and ornamental plants.
In the following years other Citrus were acquired from France, Liguria and Sicily to create a rich collection of old, ornamental and edible varieties that became one of the most complete in existence (3).
Box of lemons were sent in England for household consumption and for sale "…Lemons arrived in England in a good order…" (L. Winter, in litteris).
Historical investigation on catalogues of the plants cultivated in the garden [(4), 22 records; (5), 27 records; (3), 59 taxa; (6) 82 taxa; (7) 18 records], articles (2) and ancient documents of the Hanbury Archives (Quietanze di piante e semi e di lavori per il giardino/*Accounts for plants and seeds and for garden works*, 1877-1906; Istruzioni ai giardinieri /*Orders to gardeners* 1869-1884; letters), compared with some more recent records, enabled us to reconstruct, to a certain degree, history and dynamics of the original collection.
Today the Hanbury collection comprises about 270 *Citrus* trees, including ancient ornamental and fruiting varieties; there are nearly 60 different entities of sweet oranges, bitter oranges, lemons, limes, *citrons*, *chinotto oranges*, grapefruit, shaddocks, bergamots (and others). Many are important for food, cosmetics, pharmaceuticals, as well as ornamental and grow in various parts of the garden. These plants are all sustainably grown; we control pests organically subject to scientific criteria. Some activities are of an experimental nature and are conducted in collaboration with other countries' research institutions. Many *Citrus* produce fruit with abundant seeds and, therefore, do not have good commercial qualities, however, the conservation of these varieties is very important for the maintenance of the biodiversity in terms of genetic, historical and agronomic profile, also in line with any future processing data**.**
Among the *Citrus* genus is the extremely showy shaddock (sciadocco or pomelo) that produces fruit with very thick scented peel that can weigh more than 2 kg and are 25 cm in diameter. The largest number of plants, however, are those of the bitter orange (or Seville Orange), orange "Vanilla" and lemon.
It is important to note a fine example of *Microcitrus australis* thought to be the oldest specimen in Europe.


Fig. 1. *Citrus medica* L. 'Sarcodactylis'.


Fig. 2. *Citrus aurantium* L. 'Corniculata'.


Fig. 3. *Citrus maxima* Merr.

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3.9 = PLANT LIST OF VILLA ROQUEBRUNE GARDEN (ROQUEBRUNE CAP-MARTIN - FRANCE)

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The garden of Villa Roquebrune currently extends to about three hectares on the Cap Martin promontory.
It is at the centre of an important piece of land that Mr Emerson Muschamp Bainbridge (1845-1911) purchased in 1902. He bought it as a gift to his young bride Norah Mosson Merryweather (1880-1979) (1).
In 1912 Norah became Warre after a second marriage with George Francis Warre. She cared passionately about this garden introducing plants from all sources until the last days of her life.
She had a close relationship with the owners of many villas and gardens on either side of the Italian-French border: Lawrence Johnston and Mme Stern, the Hanburys and Waterfields, Edith Wharton, Miss Willmott, and, later, Charles de Noailles.
At the end of the seventies the Villa was converted into apartments, mostly second homes. The garden suffered only minor changes. A part near the house became a car park and a part further down the garden was used for the construction of a swimming pool. Fortunately the garden now is much as it was with almost no interference to the land. The planting that required minimal care mostly survived.
Villa Roquebrune today is back under a single ownership.
Although the Villa is only a few kilometres across the border between Italy and France, the plant list of the garden is of particular interest, because it counts taxa which were once common in many gardens of both the French and Italian Riviera, but now disappeared almost everywhere, still remaining at Villa Roquebrune.
We have now listed the plants, though some of them were very difficult to identify and remained still unresolved, such as the 200 bush roses which are mostly classic roses dated back to the late nineteenth and the first half of the twentieth century (Tea, tea-noisettes, hybrids of *R. gigantea*, hybrid tea).
The plants found and identified relate to more than 600 taxa belonging to 101 families. There are a significant number of units characterizing the ancient garden of Villa Roquebrune, this proves how some taxa have a remarkable ability to survive in places that are marginally affected by great environmental changes, with partial or no horticultural assistance. Therefore, the units found in the garden not only are surviving witnesses of the early twentieth century collection on the Riviera, but also give us a remarkable opportunity to further studies in order to understand the plant ability to acclimatize and response to global climate change.



Figg. 1, 2. *Xanthoceras sorbifolium* Bunge



Figg. 3, 4. *Purshia plicata* (D.Don) Henrickson

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4.1 = *JUNIPERUS OXYCEDRUS* SUBSP. *MACROCARPA* (SIBTH. & SM.) NEIRL (CUPRESSACEAE): A PROMISING RESOURCE FOR INTEGRATED POST-HARVEST CROP INSECT PESTS MANAGEMENT

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Juniperus oxycedrus subsp. *macrocarpa* (Sibth. & Sm.) Neirl (Cupressaceae) is a dioecious evergreen shrub species, native of the Mediterranean region whose extracts, from different parts of the plant, have been used since ancient times for fragrance, flavoring, medicinal, antibacterial, insecticidal, and cosmetic purposes (1). *J. oxycedrus* subsp. *macrocarpa* is distributed from southwestern Spain to Middle East coasts (2). In Italy it is present almost in the entire peninsula including islands where it is characteristic of the consolidated coastal dunes (Habitat 2250*: Coastal dunes with *Juniperus spp*.).
The rust-red grain beetle, *Cryptolestes ferrugineus* (Steph.) (Coleoptera Laemophloeidae), is the smallest of the grain beetles (1.5-2 mm), reddish-brown, flat and slender in the in the appearance. It is a worldwide-diffused secondary pest that attacks grains, grain products and a multitude of dried vegetables and fruits. *Sitophilus oryzae* (L.) (Coleoptera Curculionidae) also called lesser rice weevil, is a small (2.5 - 4 mm), dark brown species with 4 distinct reddish patches on the elytra. *S. oryzae* is one of the most important pests of grains, pasta, flour, legumes, baked goods and even tobacco (3). Both species are therefore considered among the most harmful to the stored food causing huge losses wordwide. For these reasons, eco-friendly strategies to prevent such insect attacks to the stored and/or final packaged products are therefore highly foreseen.
Due to their repellent properties, essential oils, obtained from aromatic plants, could represent a valid, eco-friendly alternative to synthetic repellents. In this study, the *J. oxicedrus* subsp. *macrocarpa* essential oil (JEO) was prepared from leaves and seed cones collected, from individuals growing along the Northern coast of Tuscany (Italy) in the Migliarino, San Rossore, Massaciuccoli Natural Park. JEO chemical composition was assessed by gas chromatography (GC) and by gas chromatography/electron impact mass spectroscopy (GC-EIMS). The repellent activity of JEO, against *C. ferrugineus* and *S. oryzae*, was then evaluated, *in vitro*, by the Area Preference Method (4).
The results showed that JEO is constituted mainly by mono and sesqui-terpene hydrocarbons and that is rich in chemical components such as α-pinene that has been shown to be very effective as repellents against stored food insect pests (4). Area preference bioassay indicated that JEO exerts a good repellency against *S. oryzae* and *C. ferrugineus*. Interestingly, according to probit analysis, *C. ferrugineus* is about 15 times more susceptible to JEO than *S. oryzae*. Since aromatic plants EOs present very low toxicity to mammalians and no significant risks to the environment, JEO could represents an interesting, sustainable alternative to synthetic insects repellents that could contribute to fight the losses caused to stored food by insect pests.

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4.1 = CADMIUM IN TOMATO BERRIES

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Tomato (*Solanum lycopersicum* L.) is native of the American continent, in Italy it was introduced in 1596. Here it found favorable environmental conditions and the color fruit change from the original yellow gold (from which the name) to red. Nowadays the tomato is one of the main points of the Mediterranean diet. It is considered a real ally of the wellness for the low caloric content and the high content of vitamins and mineral salts. The consumption of tomato pro-capite is estimated at about 40 g/day. In almost all producer countries, tomato cultivation involves a considerable use of fertilizers and pesticides which contain, albeit in trace, heavy metals such as nickel, cadmium, lead, highly hazardous for all living organisms health (1). Cadmium is one of the most hazardous; it is ubiquitous and very mobile in the soil, so it enters the food chain easily (2). Cadmium is very toxic to humans, it can cause kidney and skeletal damage, and cancer (3). The goal of this study was to evaluate the ability of tomato plants, grown in soil containing high concentrations of cadmium (Cd), to uptake and accumulate the metal in its tissues. For this purpose, tomato seedlings (cv Ausonio) thirty days old, were grown in plastic pots (Ø 18 cm) containing three growth substrates (S1-S2-S3); each substrate was formed from by a mixture of agricultural soil and commercial soil in the ratio 1: 4.5 w/w. The physicochemical characteristics of the substrates were the same, they differed only for the total Cd content, which was zero in S1 (control), 11,73 mg/kg in S2 and 23.90 mg /kg in S3. Cd content in S2 and S3 was respectively two and four times higher than the maximum permissible in agricultural soils (4). The plants were grown in a cold greenhouse, from May to August 2014. At the end of experiment cadmium effect on growth and productivity of the plants was evaluated. Cadmium content of leaves and ripe berries was determined by means of acid mineralization in a MEGA FKV microwave oven and a Perkin Elmer Lambda 3 model atomic absorption spectrophotometer with AAS-graphite furnace HGA 700. The response of tomato to cadmium was also evaluated by the Bioconcentration Factor (BCF) and the Translocation Factor (TF) (5). The results obtained have shown that cadmium had no effect on growth and productivity of plants: dry weight, number, size and dry weight of ripe berries of plants treated with Cd did not differ significantly from control plants. Tomato plants showed a remarkable ability to absorb Cd from soil, indeed it was observed a significant and positive correlation between Cd content of soil and Cd content of plant tissues. The highest concentrations of metal were found in the leaves; however, it should be emphasized that the cadmium content of berries, although lower than the leaves, is much higher than the value considered acceptable in food (<0.51 mg / kg dw) (6). If we consider that plants grown under our experimental conditions never showed alterations in the growth and productivity, and that in the agricultural soils, heavy metals content never attains levels that cause phytotoxicity symptoms, we can say that most of vegetables that reach our tables may contain heavy metals in concentrations above the maximum allowed threshold. Given the impact that the consumption of these foods may have on our health, it is crucial to evaluate, in addition to the degree of contamination of agricultural soils, primarily the amount of heavy metals transferred to the edible parts of the plants grown in these soils.

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4.1 = OCHRATOXIGENIC MOULDS IN THE AIR OF A SALAMI MATURING ROOM IN OLTREPO PAVESE (PAVIA, IT)

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Ochratoxin A (OTA) is a mycotoxin synthesized by several *Aspergillus* and *Penicillium* species when certain condition of temperature, humidity and pH occur. Mycotoxins are produced and released in cereals and vegetal foodstuffs but also in fermented meat products.
Traditional salami are colonized by fungi present in the air of ripening room, because no fungal starter is usually added. Even if moulds play an important role in the correct ripening of fermented meat, the presence of OTA proved to be nephrotoxic, carcinogenic, teratogenic and immunotoxic (1) has to be monitored.
This work is part of a project funded by Ministry of Health aimed to search for ochratoxin A and ochratoxigenic moulds presence in salami manufactured in Lombardy and Emilia Romagna regions.
We found *Aspergillus westerdijkiae*, a strong OTA-producer and the OTA beyond the limits permitted by Italian law (1µg/kg of meat) (2) in two salami manufactured in Oltrepo' Pavese area - Pavia province from the same producer in two different periods of 2014. Subsequently we sampled the air and the surfaces of this family-run factory to identify the source of contamination.
Quantitative air sampling was conducted with SAS ® Super ISO 100 using PDA medium (Potato Dextrose Agar). The air volumes aspirated were 200 and 100 liters in each ripening room and 200 liters in the other environments (the laboratory, the bakery and the outdoor environment).The same medium was used for the qualitative analysis. Seven swabs from different points of the ripening rooms and two from salami belonging to different batch produced in 2015 were collected for surface sampling. Fungal loads were evaluated on PDA and DRBC (Dichloran Rose Bengala Cloramphenicol Agar) media after 5 days at 25 °C incubation (3). Moulds identification was performed observing macroscopic and microscopic features as suggested by dichotomic keys for *Aspergillus* and *Penicillium* species (4, 5).
All fungal loads from air sampling resulted high (> 3300 ufc/m^3) and *A. westerdijkiae* was the predominant species in all rooms, except for the outdoor environment in which it was anyway found.
A. westerdijkiae was also isolated from 6/7 of the surface points of the ripening rooms and from both the salami produced in 2015.
This massive contamination of ochratoxigenic strains is very dangerous because it was demonstrated that these strains are able to produce ochratoxin A. In our cases 691 µg/kg and 9,4 µg/kg respectively in two salami manufactured in 2014 were detected. For this reason is crucial to sanitize all rooms and implement the strategies to prevent this kind of contamination. If the source of contamination is in the outdoor environment, technological procedures to avoid the mycotoxin synthesis will be carry out.

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4.2 = USING PROTEOMICS TO UNDERSTAND SEED DEVELOPMENT IN *PHASEOLUS VULGARIS* L.: DEVELOPING RESOURCES TO MODULATE SEED QUALITY TRAITS

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Common bean (*Phaseolus vulgaris* L.) is the most consumed grain legume in developing countries in Latin America and Sub-Saharan Africa[1]. Like other grain legumes, common beans are rich in protein, carbohydrates, fibers and other health-promoting phenolic compounds thus being vital for food security and income source for local small farmers[2]. Seed quality depends on accumulation of various storage molecules during seed development (SD) and is influenced by genotype and by environmental adaptive changes[3]. In common beans, there is still a lack of a deeper knowledge on SD molecular processes which, in turn, hampers the development of new biotech approaches for the modulation of seed traits and timely address challenges of agriculture or industry.

Our present work aims to unravel the molecular mechanisms underlying SD using a proteomic approach. To achieve this goal, we characterized SD in terms biomass, seed length and weight in the genotype SER16, one of the most promissory drought-resistant release of the CIAT-CGIAR. Seed samples were collected at the 4 main SD stages: Late-Embryogenesis (10 days after anthesis, d.a.a.), Early (20 d.a.a.) and Late Maturation (30 d.a.a.) and Desiccation (40 d.a.a.).

The analysis of bean proteome was conducted using a gel-free proteomic analysis (LC-MS/MS) under the scope of EU-FP7-PRIME-XS project. A total of 410 unique proteins were differentially expressed throughout the 4 major seed development stages, in which most of the identified proteins belong in the 'protein metabolism' (31,98%) functional category that includes synthesis, regulation and folding. Other functional categories are represented such as carbohydrate and lipid metabolism (11,26%) and stress/defense and redox metabolism (11,04%). We identified 93 proteins unique to the first (10-20 d.a.a.), 22 to the second (20-30 d.a.a.) and 40 to the last (30-40 d.a.a.) phase transition, reflecting the major biological processes occurring at the specific seed developmental stages. This study will contribute to reveal key metabolic pathways and mechanisms with potential role in the modulation of the development and quality traits of common bean seeds.

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4.3 = EFFECTS OF *BETULA AETNENSIS* RAFIN. (BETULACEAE) EXTRACT ON A HUMAN COLON CANCER CELL LINE

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Oxidative stress was involved as a causative agent in many human degenerative diseases, including aging, cancer and neurodegenerative disorders such as Alzheimer's disease, Parkinson's disease (1). Recently, considerable effort has been directed towards identifying naturally occurring substances that can protect against oxidative stress. Natural antioxidants have a wide range of biochemical activities such as inhibition of ROS generation and alteration of intracellular redox potential (2). Much attention has been focused on phytoconstituents present in fruits, vegetables and medicinal herbs that may be helpful in preventing complications related to cancer. The possibility that fruit and vegetables might help to reduce the risk for various types of cancer raised great interest already in the 1970s, when studies conducted to assess differences in cancer rates and diet between countries, suggested that various dietary factors might have important effects on cancer risk (3). Many antioxidant compounds have been investigated for their potential usefulness as cancer chemopreventive agents (4, 5). Since there is an increasing interest in the *in vivo* protective effects of natural compounds contained in plants against oxidative damage involved in cancer, in this study we investigated the effects of *Betula aetnensis* Rafin. bark alcoholic extract on a human colon carcinoma cell line (CaCo2).
Betula aetnensis Rafin. (Birch Etna) is a medium-sized deciduous tree, typically reaching 5–20 m tall, which belongs to the family Betulaceae. It grows on the eastern slope of Etna, at an altitude between 1200 and 2000 m (6, 7). Many *Betula* species are used in folk medicine to treat skin diseases, infections, inflammations, rheumatism and urinary disorders (8). Nearly, all species contain flavonoids, tannins, saponins, sterols and pentacyclic triterpenoids, such as betulin, betulinic acid and ursolic acid which have shown multiple antioxidant, antitumor and antiviral activities. In particular, betulinic acid is considered a promising anticancer agent (9).
The present study investigated the effects of *Betula aetnensis* Rafin. bark alcoholic extract on the viability of CaCo2 cells. In addition, in order to elucidate mechanisms of action of this extract, LDH release, ROS levels and several markers of oxidative stress were also evaluated.
Results obtained in the present study showed that alcoholic extracts of *Betula aetnensis* Rafin. significantly reduced cell viability of CaCo-2, inducing apoptotic and/or necrotic cell death in a concentration-depending manner. These results confirm that *Betula aetnensis* Rafin. exhibits interesting health promoting properties suggesting that this extract could be both in the prevention of cancer, due to its antioxidant activities, both as adjuvant in antineoplastic chemotherapy (due to its antiprolifertive/cell death-inducing effect).

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4.3 = A NEW SESTERTERPENE FROM *SALVIA TINGITANA* ETL. (LAMIACEAE)

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Salvia tingitana Etl. (Syn. *Salvia foetida* Lam., *Salvia disermas* auct. non L., *Salvia sclarea* sensu Rosúa in Taxon 37: 188. 1988) is an aromatic woody-based perennial shrub, up to 100 cm, with erect tomentose stems, simple, ovate, subcordate, crenate leaves, with eglandular and glandular indumentum, and inflorescence arranged in broad branched panicles with white flowers showing a cream-yellow lower lip (1, 2). The species is original from the Arabian region (1) and cited by Gattefossé as the greek *Marum* (3). *S. tingitana* was used and cultivated as a medicinal plant in the area of Arabia, and during the expansion of the Arab Empire it probably diffused in Egypt, Syria, Aleppo, Tunis and Tanger. In the early 1600s Alpini described the species in the Padua Botanical Garden, identifying it as "*Marum Aegyptium*", and suggesting the use of the flowers and the seeds as rubefacient, solving, digestive, the decoction of the leaves and the shoot tips against colds and drunkenness, and the fresh juice of the leaves in oxymiel to remove facial blemishes (4). The species has also a long history of cultivation in gardens; it is reported as grown inside the Jardin du Roi, belonging to the Comte d'Artois (who became Charles X) in Paris in 1777. At present *S. tingitana* is cultivated as an ornamental in different parts of the world (1). The species could be extinct in North Africa (2), and the only known recent collection is from Saudi Arabia (5). Unless for a long time the taxonomic interpretation of the species was confused (2, 5, 6), the recent studies state *S. tingitana* as a distinct species and separate it from *S. sclarea* and other species that in the past have been considered as related with it (1).
We have started an extensive study on *S. tingitana* aerial parts since phytochemical data are still lacking. The hexane-soluble fraction of the surface exudate obtained by rinsing the fresh biomass with CH_2Cl_2 was subjected to repeated column chromatography on Sephadex LH-20 and silica gel, and to semi-preparative RP-HPLC. One flavonoid, salvigenin (7) as well three known sesterpenes (8, 9) and 8□-hydroxy-sandaracopimar-15-ene (10) have been isolated together with a new sesterterpene, identified by IR, NMR, including TOCSY, COSY, HSQC, HMBC and ROESY experiments, ESI-MS and HR-MS analysis.

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4.3 = PHARMACOGNOSTIC STUDY AND ANTIVIRAL PROPERTIES OF *COMBRETUM MICRANTHUM*

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The Combretaceae family is a widespread pantropical flowering group of plants, including more than 500 species and 14 genera of which 8 are located in West Africa (tropical and sub-Saharan regions). The genera *Combretum* and *Terminalia* are known to be widely used and play an important role in Africa's traditional medicines (1, 2). The genus *Combretum* includes almost 100 African species and is the most interesting due to the medicinal properties of its bark and leaves, which produce important compounds for traditional treatment of a wide range of diseases (3). *Combretum micranthum* G. Don is an undomesticated bushy shrub or creeper growing in the Tiger bush region of sub-Saharan western Africa (with higher concentrations in Senegal, Mali and Burkina Faso). In this region, it is so widely used as a general panacea that the African name of *C. micranthum*, kinkéliba, has become synonymous with "medicine" in some languages (4). The plant is considered one of the 50 most important African ethnomedicinal remedies by the Association for African Medicinal Plant Standards (www.aamps.org). *C. micranthum* leaves are used as a popular herbal infusion or tea, the roots are used in a decoction. These extracts are generally recognized to exert a therapeutic effects as: antibacterial (5), antiviral (6), anti-inflammatory (3) and antimalarian (7). Several leaf components have been identified, including flavonoids, catechins, organic acids, triterpenoids and polyphenols.

This study is focused on the pharmacognostic characterization of *C. micranthum* and the antiviral (DNA and RNA viruses) activity of extracts from dried leaves. Herbal samples were obtained from: (a) commercial source (herbal preparation, from Senegal-Burkina Faso); (b) plants collected in Benin by the Gruppo Solidarietà Africa (GSA Onlus). Pharmacognostic analysis was carried out by using DNA barcoding, optical and scanning electron microscopy and phytochemical studies. DNA barcoding analysis involved the use of four different markers: *matK, trnH-psbA, rbcL* and *ITS*. Comparison with the international database (NCBI) enabled the correct identification of the species analyzed, consistent with micro-morphological data. Microscopic observations revealed typical anatomical features of the *Combretum* genus (8). Secretory peltate brown scales were found scattered on leaf surfaces, with a higher density on the abaxial leaf (glandular trichomes containing a secretion consisting of lipids, terpenoids, phenolics, flavonoids and alkaloids, with possible anti-microbial effects), calcium oxalate druses within the mesophyll and marsupiform domatia like small pockets, observed also in other species of Combretaceae such as *Guiera senegalensis* (9). These latter were found at the point of branching of secondary vein from the main vein. Active antiviral fraction from *C. micranthum* was isolated by extraction of dry minced leaves with ethanol, subsequent dissolution of the solid material in a minimum amount of water, pH 7.0, removal of a precipitate, and freeze-drying of the aqueous solution, affording a pale yellow solid that was stored at -80 °C. Thereafter, the final solid material was further purified with organic solvents, yielding a highly hygroscopic whitish solid. The DPPH (2,2-diphenyl-1-picrylhydrazyl) assay (compared to ascorbic acid) confirmed the anti-oxidant power of all extracts tested, in particular those obtained from leaves previously exposed to sun or boiled. All fractions were also tested for antiviral properties against Polio Virus 3 and Herpes Simplex Virus 2 in Vero cells. Data confirmed previously observed inhibition of *C. micranthum* on Herpes Simplex Virus 2 infection (6) and also showed the inhibition of Polio Virus 3 infection, which had never been demonstrated before.

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4.3 = PHYTOCHEMICAL STUDY OF *TAMARIX AFRICANA* POIR. (TAMARICACEAE)

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Tamarix genus is a tree or shrub halophytes from coastal regions and desert, relatively long-living plants that can tolerate a wide range of environmental conditions and resist to abiotic stresses such as salt, high temperature, and drought stresses (1). *Tamarix* spp. are employed in traditional medicine as astringent, appetizer, stimulant of perspiration, diuretic, and spleen trouble and eye remedies (2). Younos et *al.* (2005) indicated the importance of this genus in some old civilizations and the actual uses of *Tamarix* species different organs (leaves, flowers, and galls) in the traditional Asian therapy as anti-inflammatory, anti-diarrhoic, healing and antiseptic agents (3). *T. africana* Poir. is an ornamental and folkloric medicinal halophyte particularly abundant in Mediterranean salt marshes (2); it's a perennial shrub with brownish branches, punctuated, opaque, oval and acuminate leaves; its bunches reaching 6 cm long, 5-6 mm wide and contained flowers with four to five petals and four stamens. The ability of halophytes to tolerate salt and to overcome oxidative stress is determined by multiple biochemical pathways that facilitate water retention and/or acquirement, protect chloroplast functions, and maintain ion homeostasis. Essential pathways include the synthesis of compatibles solutes, specific proteins and radical scavenging molecules, such as phenolic acids and flavonoids (4). In recent years, the interesting feature of halophytes as a potential source of natural antioxidants, has aroused the interest of many researchers. In fact, it's reported a beneficial preventive and therapeutic effects of phenolic compounds against diseases associated with oxidative stress. These constituents, and more particularly the flavonoids, are endowed with important biological activities including antioxidant, anti-inflammatory, anti-ulcer and anti-tumor. Previous studies on *Tamarix* species reported the isolation of hydrolyzable tannins (5), flavonoids (6) and phenolic compounds (7), while in a phytochemical work on *T. africana trans*-coniferyl alcohol 4-sulphate was purified and other cinnamyl alcohol sulphates were detected (8). Furthermore, several researches proved antioxidant, antimicrobial and anticancer activities of some *Tamarix* species such as *T. ramosissima* (9), *T. hispida* (10), and *T. gallica* (11).
In this context, the aim of the present work was to carry out a phytochemical study of the polar extract of *T. africana* aerial parts to isolate and identify bioactive compounds, including some sulphated derivatives, which could be responsible of its biological activities. The plant material was collected in Gabès (Tunisia) and dried at room temperature, then extracted with Soxhlet apparatus with *n*-hexane, dichloromethane and then at room temperature with methanol. The MeOH extract was partitioned between *n*-BuOH and H_2O and the *n*-BuOH soluble fraction was separated by Sephadex LH-20 followed by RP-HPLC. Sulphated flavonoids, including some new compounds, were purified and fully characterized by spectroscopic techniques such as 1D and 2D-NMR, and mass spectrometry.

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4.3. COMPOSITION OF ESSENTIAL OILS OF *ROSMARINUS OFFICINALIS* L. FROM ELBA ISLAND (TUSCANY, ITALY)

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The genus *Rosmarinus* L. (Lamiaceae) is represented by spontaneous shrubs growing all over the world. In the Mediterranean area, the genus *Rosmarinus* L. includes five species: *R. officinalis* L., *R. eriocalyx* Jourdan and Fourr, *R. laxiflorus* (De Noé) Batt., *R. lavadulaceous* Batt. and *R. tomentosus* Huber-Morath and Marie [1-3]. *Rosmarinus officinalis* L. is widely used in the folk medicine, cosmetics, herbal products, and food flavoring [4].
The composition of *R. officinalis* essential oil has been reported in several studies [5-7]. The major components were □-pinene, 1,8-cineole and camphor, associated with variable content of camphene, limonene and verbenone.
In a previous work, the composition of EOs obtained from *R. officinalis* growing in Tuscany and minor islands of Tuscan Archipelago (Gorgona, Capraia, Giannutri, Giglio, Montecristo and Pianosa) has been described [8].
The present work deals with the composition of eighteen samples of *Rosmarinus officinalis* L. essential oils from six locations of Elba Island (Tuscany, Italy) characterized by different type of soil and microclimate and collected in three different periods of the year (January, May and October 2010). The analysis was performed by Gas Chromatography (GC) and Gas Chromatography/Electron Impact Mass Spectrometry (GC/EIMS). Eighty-eight components were identified representing 96.8-99.6% of the whole chemical composition. These oils were characterized by a high content of oxygenated monoterpenes (49.2-80.3%) and hydrocarbon monoterpenes (18.7–48.3%). The main constituents were 1,8-cineole (26.4–49.1%), α-pinene (4.5-34.8%) and camphor (1.1–18.8%).
The results of this study have been compared with those from all the other islands of Tuscan Archipelago (Gorgona, Capraia, Pianosa, Giglio, Montecristo and Giannutri) previously studied [8]. Apart from Giglio and Giannutri Islands, the HCA analysis showed a clear separation between the Elba samples and those from the other minor Islands.

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4.3 = IN VITRO ACTIVITY OF *WALDHEIMIA GLABRA* (DECNE) REGEL EXTRACTS ON THE SOMATOSENSORY AND PAIN RECEPTOR TRPA1

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The Himalayan regions are reputed to be a rich storehouse of biodiversity and medicinal and aromatic plants (MAPs). Among Himalayan MAPs, *Waldheimia glabra* (Decne.) Regel (family: Asteraceae, tribe: Anthemideae) commonly known as Smooth Ground Daisy is an aromatic, high altitude perennial species commonly found on slopes among small stones near glaciers and in rock crevices in sandy-loam soils at 4000–5400 m above sea level (a.s.l.) in Himalayan alpine zones, particularly in Nepal, Bhutan, Tibet, Chitaland Gilgit-Baltistan districts of northern Pakistan, Kashmir and the great Himalayas of northern India (1). This plant is traditionally used by the local populations in religious rituals (incense) or in traditional herbal medicine to treat skin diseases, headache, join pain and fever (2). In literature few data are available on the investigation of this aromatic plant (3). Therefore, the present work aims at deepening the knowledge about the chemical composition of *W. glabra* essential oil, extracts and incense using HS-SPME GC/MS and GC/MS analysis. Preliminary tests on the activity of *W. glabra* extracts on TRP channels were also performed.
Five different polarity extracts of aerial part of *W. glabra* has been submitted to preliminary *in vitro* test with the cloned TRPA1 receptor. This ion channel is involved in several sensory mechanisms, including pungency but also environmental irritatation and pain perception. Some extracts showed an interesting activity in terms of efficacy and potency that could be related to some use of this plant in traditional folk medicine. Molecular analysis is undergoing to isolate the specific agonists and to elucidate their role in this perception pathway.

Fig. 1. *Waldheimia glabra* (Decne.) Regel.

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4.3 = THE COLLECTION OF MEDICINAL MACROFUNGI OF PAVIA UNIVERSITY

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In 2010 researchers of the University of Pavia began the collection of wood decay macrofungi strains, and among them various are medicinal mushrooms, able to develop their sporomata in the Italian environmental conditions. The culture collection, nowadays deposited at the Mycological Laboratory of DSTA of the same University, has been year after year implemented and enriched also thanks to the contribution of Bologna and Siena Universities.

This action was driven by the necessity to protect fungal species having officinal properties from the threat of extensive picking, as well as to conserve *ex situ* rare *taxa* of particular interest (1).

Indeed, culture collections play a key role in preservation and maintenance of fungal genetic resources and they are an important tool to get biological material for application purposes.

Wood-inhabiting fungi were collected from Italian Alps, Apennines, plain woods and Mediterranean areas. Particular attention has been paid towards "polyporoid and corticioid" fungi. Pure culture isolation was mostly carried out from sporomata and in a few cases from basidiospores; always by experimental sterile conditions.

During the last 5 years, 217 strains belonging to wood-decay macrofungi were obtained (Carolina Girometta, personal communication). Among these, about 50 are believed to have officinal properties, referred to scientific publications and observations from traditional medicines, mainly the Chinese one (MTC).

It must be underlined the isolation of *Fomitopsis officinalis* (Vill.) Bondartsev & Singer (syn. *Laricifomes officinalis*), well known since I Century A.D. for its medicinal properties (2). Due to the past extensive collection, it became so rare to be supposed almost extinct in Europe (3). Consequently eight European countries included it into the Red Lists of threatened species and in 2014 it was added to the Italian one too (Ministero dell'Ambiente e della Tutela del Territorio e del Mare). Thanks to programs of habitat protection together with a specific census, *F. officinalis* spread out again on the Alps (4). Noteworthy also is the recovering of *Ganoderma pfeifferi* Bres. from Teramo and *Hericium erinaceus* (Bull.) Pers. from Siena., Under assessment by the Global Fungal Red List Initiative of IUCN, this last is a fungus rich in physiologically important components and studied for its properties also at Pavia University (5).

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4.3 = MICROMORPHOLOGY AND ANATOMY OF BITTER MELON (*MOMORDICA CHARANTIA* L., CUCURBITACEAE) FRUITS AND SEEDS

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Momordica charantia L., known as bitter melon, is a climbing species, belonging to the Cucurbitaceae family growing in tropical areas of Asia, East Africa, Amazon, and the Caribbean (1). It is an important commercial vegetable crop in many countries with a raining tropical or subtropical climate, used also for medicinal purposes.
The plant presents characteristic leaves with serrated margins; probably this feature gave rise to the genus name deriving from the Latin word *momordere* that means "to bite". The flowers are yellow or yellow/orange. The fruit, resembling to a cucumber, have a tuberculate surface presenting numerous swellings; it is emerald green in colour when immature, turning orange-yellow during ripening. At the mature stage it splits from base into 3 valves exposing 5-15 red seeds. The fruit mesocarp is soft and spongy with a bitter taste. The nutritional value is scarce, the fruit containing low quantities of carbohydrates and very few proteins, but compared to other Cucurbitaceae has a relatively high nutritional value, due mainly to the iron, phosphorus and ascorbic acid content (1).
Several centuries ago, plants of the genus *Momordica* were cultivated also in Italy, as documented in a picture by an anonymous painter of the XV century representing the Virgin Mary with the Child and Saints under a pergola of *Momordica* bearing fruits at different ripening stages(2). Afterwards, the plant cultivation was abandoned, probably as a result of the introduction from the New World of other Cucurbitaceae, more pleasant to taste and more suitable to the Italian climate.
The plant was used in Africa and Asia not only for food, but also against several diseases, as anthelmintic, digestive, laxative, and many others (3). However the Western medicine paid attention to the plant only when the antidiabetic properties of fruits and seeds were experimentally demonstrated (4). Since then, the plant has been the subject of over a hundred scientific articles focusing on its phytochemistry and pharmacology (5).
In this work, micro-morphological observations on fruits and seeds are reported. The fruit presents a thin epicarp with a thick cuticle and several stomata, a multi-layered mesocarp showing numerous amyloplasts in the inner layers, and a ring of fibro-vascular bundles. The inner mesocarp layer bears stomata and delimits a cavity containing the seeds with the characteristic red slimy aril. The seeds are oblong with grooved margins and a sculptured surface. The seed coat shows four distinct layers: epidermis with a thick cuticle, hypodermis, sclerenchyma and chlorenchymatous parenchyma, as typically described in several members of Cucurbitaceae (6). Cotyledons are rich in proteins.

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4.3 = ULTRA-LOW PRESERVATION OF THE MEDICINAL MUSHROOM *GANODERMA LUCIDUM*

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Ganoderma lucidum (Curtis) P. Karst. is one of the most famous medicinal mushroom species, which has been used in traditional medicine of the Far East for more than 2000 years. Recent studies have confirmed the traditional knowledge and reported a list of medicinal properties of *G. lucidum* extracts, such as: immunomodulating, antitumor, antioxidant, antimicrobial, cardiovascular, antiallergenic, hepatoprotective, antidiabetic, etc. (1). This fungus is currently used in the formulation of nutraceuticals and as functional foods.
It is well known that medicinal mushroom bioactivities depend on growth conditions and genetic characteristics (2). With the name *G. lucidum* were included different species from Europe and Asia and recent phylogenetic studies asserted that *G. lucidum* used in Chinese traditional medicine is a different species from *G. lucidum* commonly found in Europe (3, 4). Different species of *Ganoderma,* but also different strains of *G. lucidum,* showed differences in antioxidant and antiproliferative activity (5). In order to preserve the genetic resources of *Ganoderma* spp. it is necessary to collect specimens from different geographical regions and maintain their pure cultures for long periods.
Cryopreservation in liquid nitrogen or ultra-refrigeration in deep freezer are considered as the most reliable methods for long-term storage of filamentous fungi (6, 7). These techniques are presumed to allow very long periods of storage (i.e. centuries or even millennia) and to prevent genetic, physiologic and phenotypic changes (8).
The aim of this work was to test the possibility to apply ultra-refrigeration to *G. lucidum* mycelium and to verify its ability to survive and fructify after storage at low temperatures (-120 °C). For the first time, cryoprotectans properties of wheat seeds, sucrose ($C_{12}H_{22}O_{11}$), glycerol ($C_3H_8O_3$), propan-2-olo (C_3H_8O) and sodium chloride (NaCl) were combined together to preserve vitality of three *G. lucidum* strains from Italy during freezing. Fruiting test was carried out inoculating plastic bags containing oak sawdust and straw with preserved and fresh pure cultures.
Our results showed that the mycelium of *G. lucidum* can be successfully preserved at – 120 °C. The morphology of the mycelia in pure cultures was not affected by low temperatures and all the *G. lucidum* strains investigated kept unchanged the ability to form fruiting bodies after freezing.
Our results demonstrated that ultra-refrigeration technique developed in this experience might be suitable also for the other species of mushrooms without use liquid nitrogen. The possibility to preserve fungal strains at -120°C opens up the possibility to collect fungal strains all over Europe for the creation of a germplasm bank, thus contributing to the maintenance of fungal biodiversity.

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4.3 = PRELIMINARY ESSENTIAL OILS STUDY IN THE GENUS *TEUCRIUM* FROM SARDINIA (ITALY)

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Teucrium L. is a genus belonging to the Lamiaceae family, which includes 300 species distributed in Europe, North Africa and temperate parts of Asia, but mainly concentrated in the Mediterranean region (1). It occurs in Sardinia with 11 taxa (2,3,4). *Teucrium* species are generally aromatic and they have been used in folk medicine as stimulants, tonics, stomachache remedies and also antidiabetic agents (5). In Sardinian traditional medicine some *Teucrium* species are used as antiseptic, cicatrizant and to treat skin diseases (6).
As part of our ongoing investigation, the aim of the present study was to characterize and compare the essential oils obtained from six wild *Teucrium* species growing in Sardinia (Italy): *T. scorodonia* L., *T. massiliense* L., *Teucrium capitatum* ssp. *capitatum* L., *T. marum* L., *Teucrium subspinosum* Pourr. ex Willd., *Teucrium flavum* ssp. *glaucum* (Jord. & Fourr.) Ronniger. The selected taxa belong to the chamaephytes life form, except *T. scorodonia* which is a hemicryptophyte.
Flowering aerial parts of all species have been collected from different locations, air-dried, and subjected to steam distillation. Their chemical composition was analyzed by GC/FID and GC/MS.
Our results showed that *T. scorodonia*, *T. marum*, *T. subspinosum* and *T. massiliense* essential oils were rich in sesquiterpene hydrocarbons (92.2%, 58.5%, 66.1% and 45.3% respectively), while in *T. capitatum* ssp. *capitatum* and *T. flavum* ssp. *glaucum* the monoterpene hydrocarbons predominate (87.6% and 76.8% respectively). Furthermore, *T. marum* reported the presence of diterpene hydrocarbons and *T. massiliense* of non-terpenic oxygenated compounds. The main compounds in *T. scorodonia* essential oil were (E)-Caryophyllene (19.0%), α-Cubebene (14.5%) and Germacrene B (14.1%); in *T. marum*, β-Bisabolene (23.0%), β-Sesquiphellandrene (17.8%) and 3*E*-Cembrene A (14.0%); in *T. subspinosum* (*E*)-Caryophyllene (22.6%), β-Bisabolene (19.9%) and β-Sesquiphellandrene (12.9%); in *T. massiliense* 6-Methyl-3-Heptyl Acetate (23.5%), γ-Muurolene (11.0%) and (E)-β-Farnesene (8.4%); in *T. capitatum* ssp. *capitatum* Limonene (30.4%), α-Pinene (29.8%) and β-Pinene (10.0%) and in *T. flavum* ssp. *glaucum* Limonene (27.0%), α-Pinene (25.1%) and β-Pinene (15.1%). Our outcomes on *T. scorodonia* and *T. flavum* ssp. *glaucum* confirmed the results previously obtained by other authors (7,8), whereas the comparison of *T. massiliense* essential oil with bibliographic data, showed quali-quantitative differences (8). This variability could be due to environmental conditions. Focusing on *T. marum* and *T. subspinosum*, two species often confused, it is possible to appreciate a similar pattern in the chemical composition, although quantitative differences in the main compounds have been highlighted. The comparison between the essential oil of Sardinian *T. capitatum* ssp. *capitatum* and the analysis carried out by other authors (8) showed some important differences, especially regarding the amount of Limonene (30.4%), which is higher in the sample collected in Sardinia.
This preliminary work highlighted the high variability of the essential oils of the genus *Teucrium* from Sardinia. This is a first step for a future broad-spectrum research on all the Sardinian *Teucrium* species. Moreover, the subsequent goal would be to collect other Sardinian *Teucrium* species and perform cluster analysis (CA; dendrograms) in order to identify statistical differences between the species under study.

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4.3 = ETHNOBOTANICAL SURVEY WITHIN SCI IT4050001 AREA (PARCO DEI GESSI BOLOGNESI, CALANCHI DELL'ABBADESSA), NEAR BOLOGNA, ITALY

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The aim of the Ethnobotanical survey carried out within the area of "Parco dei Gessi Bolognesi e Calanchi dell'Abbadessa" (SCI-SPA IT4050001) is to preserve in writing the information orally handed down and concerning the plant use by the local community; this work achieves the preservation of a knowledge heritage with practical and cultural importance. The park covers 4844 hectares in the municipalities of San Lazzaro di Savena, Ozzano dell'Emilia, Bologna and Pianoro (BO) and the buffer area to the Park area has an agricultural pattern. Inside the park area 17 communities coexist: Rastignano, Miserazzano, Croara, Monte Calvo, Farneto, Pizzocalvo, Settefonti, Ozzano, Ciagnano, Castel de' Britti, San Pietro di Ozzano, Cà de Mandorli, Idice and Campana, Uccellara, Monte Bello, Casola Canina, Villa Torre. Geographically, the investigated area is located in a hilly, between 40 and 400 m above sea level, between the streams Savena, Zena and Quaderna. The mixture of different geomorphological structures, dominated by selenitic outcrops, gives to the territory a great environmental heterogeneity, with considerable effects on biodiversity of the region. Because of the extreme variety of environments (such as gypsum outcrops, sinkholes, blind valleys), in a floristic perspective, this is the richest and most interesting site of all the SCI which are part of LIFE+Project 08NAT/IT/000369 "Gypsum". The ethnobotanical survey was carried out through anonymous demi-structured interviews, applied to residents in the area of interest, according to the modern operational schemes (1), supported by plant samples collected in advance or as suggested by the respondents, or by herbarium specimens, or by text images. The species mentioned by only the dialect were identified with the support of the Ungarelli dictionary (2), useful for a correct transcription of vernacular names. The interviewed has reported, for each species, information on the collection times and places, on the medical use, on the mode of preparation and administration. All mentioned species were classified according to Conti *et al.* (3). The total interview were 233, 64% of which was female; such an imbalance is explained considering that a lot of men declared to be ignorant about plant uses. Respondents were averaged of 76 years. Overall mentioned species were 143, each catalogued with binomial name, common name, dialect name (if known) and taxonomic family. Plant information amounted to 1001; if divided by uses, 17% are for food use; 14.1% are used against gastro-intestinal system diseases; 11.9% for epidermal diseases; 8.2% are used as anti-inflammatory or painkiller. Infusion (37.4%) and *in toto* use (21.6%) are the main administration ways, followed by decoction (12.6%) and maceration (8.9%). These results are in agreement with the Italian Popular Pharmacopoeia (4). Some therapeutic use are unique: in tradition, *Allium sativum* was used to made necklaces to be worn by children against mawworms; the infusion of *Santolina chamaecyparissus* was rub on skin bite by insects to relieve irritations. Some special cases testify how strong was the relationship with plants in the past: *Sambucus nigra* was used to make flutes, and according to tradition they should be realized before the crow, otherwise the tone would be hoarse. Furthermore, a habit was mentioned dating back to the Middle Ages: people used to gather under *Ulmus minor* to celebrate trials and to apply hangings. Relationship with plants became stronger during harsh times like famines and wars, as testifies the use in the past of *Cynodon dactylon* rhizomes in order to pad wheat flour. Among veterinary uses, fresh leaves of *Hedera helix*, added to fodder, calm goats when they are nervous because the captivity in narrow spaces; instead, feeding horses with peppermint contributes to make a shiny coat. Some information mostly pertains to the superstition; for example, diviners used to make their dowsers with *Salix* wood; or, according to tradition, cold and flu can be prevented keeping three seeds of *Aesculus* in pocket. The survey shows that the distribution of the plant uses across the study area is not homogeneous. Food uses constitute an important heritage of knowledge typical of the area. Moreover, despite the recent industrial development, it emerged the residue of a strong relationship between the human community and the park flora, and this research has contributed to maintain its knowledge.

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4.3 = METABOLOMICS APPROACH FOR SCREENING COLLAGENASE INHIBITORS FROM MEDICINAL PLANTS

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The extracellular matrix (ECM) constitutes the scaffold necessary for tissue architecture and integrity. Moreover, in addition to provide structural support, ECM was also demonstrated to be involved in several dynamic processes such as regulation of cell migration, adhesion, trafficking and signaling (1). Collagen is a crucial component of the ECM and it represents the most abundant protein found in mammalian tissues, indeed it is the main structural protein of skin, bone and tendon. Collagenase, the proteolytic enzyme responsible for the cleavage of native fibrillar collagen, is considered the founding member of the matrix metalloporoteinases (MMPs) family. A misregulation in the collagen turnover is characteristic of several diseases including arthritis, atherosclerosis, invasion and metastasis of malignant tumors, fibrosis and aneurysm, non-healing wounds (2). Moreover, the degradation of collagen is also involved in skin ageing processes leading to wrinkles formation. On these bases, collagenase and in general MMPs have recently become interesting targets for the discovery of novel types of pharmacological and cosmetic agents (3). Despite the first lead candidate targeting the MMPs was synthetized in the late 1970's, until now the antibiotic doxycycline remains the only FDA approved drug for its activity as MMP inhibitor used in the therapy of periodontal disease (4). Considering the severe side effects showed by the synthetic MMPs inhibitors, it results interesting to investigate the activity of medicinal plants on these biological targets. In order to found natural products endowed with collagenase inhibitory activity, we select 49 medicinal plants based on the potential correlation between their ethnobotanical uses and this biological activity. The collagenase inhibitory activity of these plants was tested *in vitro* and the ^{1}H-NMR-based metabolomic profiles was also investigated. A multivariate treatment of the data was performed (PLS and OPLS models) and a correlation between the presence of tannin-related compounds (signals at 7.08 ppm in the spectra) and the activity increase was highlighted. Based on this results, tannin-free extracts were prepared using polyamide columns and after this procedure the anti-collagenase activity was tested again. Only three out of the 15 active plants were found still able to strongly inhibit the enzyme, namely: *Alchemilla vulgaris, Tinospora cordiflia, Emblica officinalis*. In conclusion this study can provide an easy method to identify and threat plant material in order to found new collagenase inhibitors different from tannins. Further studies are in progress on the three above-mentioned plants to identify the active compounds.

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4.3 = *NASTURTIUM OFFICINALE* R.BR. SUBSP. *OFFICINALE*: NEW SOURCE OF NATURAL INHIBITORS OF PANCREATIC LIPASE

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Nasturtium officinale R. Br. (Brassicaceae) is a hardy perennial native to Europe. Normally, it is sold in fresh form and consumed as a vegetable in salads, soups and other recipes. The leaves of this plant are also widely used as a home remedy as depurative, diuretic, expectorant, hypoglycemic and antiodontalgic (1). Gill et al. reported that watercress supplementation in diet reduces lymphocyte DNA damage and alters blood antioxidant status in healthy adults (2). Also, antimitotic and anticancer effects of this plant have previously been reported (3). Yazdanparast et al. (4) concluded that *N. officinale* has a high hypolipidaemic activity and this may be attributed to its antioxidative potential.
Metabolic syndrome had become a global epidemic, defined as a cluster of three of five criteria: insulin resistance and glucose intolerance, abdominal obesity, hypertension, low high-density lipoprotein cholesterol and hypertriglyceridemia. The prevalence of metabolic syndrome in adults has been increasing rapidly in the past decades in most western countries.
Many plants are known to have not only nutritive and taste values but also physiological effects, as they are prescribed in various traditional preparations (5). Numerous trials have been conducted to find and develop new anti-obesity drugs through herbal sources, in order to minimize side effects associated with the present anti-obesity drugs. One most important strategy in the treatment of obesity includes the development of nutrient digestion and absorption inhibitors, in an attempt to reduce the energy intake through gastrointestinal mechanisms without altering any central mechanisms. Pancreatic lipase is a key enzyme for triglycerides absorption in the small intestine.
This study was designed to evaluate the hypolipidemic effects of *N. officinale* through inhibition of digestive enzyme pancreatic lipase. *In vitro* studies here reported were performed to evaluate the inhibitory activity of alcoholic extract and fractions of leaves of *N. officinale* from Calabria region (Italy) on lipase by monitoring the hydrolysis of *p*-NPC. Total phenolic content was determined using Folin-Ciocalteau methods. In order to identify phenolic constituents, HPTLC analysis of crude extract was performed. Crude extract of *N. officinale* leaves showed a total phenolic content of 10.34 ± 0.15 mg/g (expressed as chlorogenic acid equivalents *per* g of dried material). HPTLC analysis confirmed the presence of several phenols; in particular quercetin, kaempherol and chlorogenic acid were identified and confirmed by ^{1}H NMR, together with a relevant quantity of high molecular weight polyphenols.
Crude extract showed a significant anti-lipidemic activity with an IC_{50} value of 2.33 mg/ml, while among fractions AcOEt was the most active with an IC_{50} value of 1.01 mg/ml.

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4.3 = PHENOLIC CONTENT AND IN VITRO ANTI-INFLAMMATORY ACTIVITY OF ECHINOPHORA TENUIFOLIA L.

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The genus *Echinophora* (Apiaceae) comprises about ten species, distributed in Mediterranean and Middle East regions (1). In Turkey these plants were utilized in folk medicine to heal wounds and to treat gastric ulcers due to their antifungal, carminative and digestive properties (2).
In Europe, this genus is represented by two species (*E. spinosa* L. and *E. tenuifolia* L.), distributed from the Mediterranean region to Crete and Crimea. Two different subspecies of *E. tenuifolia* are known [subsp. *tenuifolia* and subsp. *sibthorpiana* (Guss.) Tutin]. However, according to more recent literature data, later subspecies should be treated as a separate species: *E. sibthorpiana* Guss. (3).
Some *Echinophora* species are also added to foods, such as soup, meat, cheese and yoghourt, to enhance their sensory properties. Moreover, the essential oil of *E. platyloba* DC has been recently used as a natural preservative in dairy products to prevent fungal growth (4, 5).
A number of studies dealt with the phytochemical composition of essential oils of different species, above all *E. platyloba* and *E. sibthorpiana*, but none of the previous works focused on the biological activity of alcoholic extract of *E. tenuifolia* to the best of our knowledge.
The purpose of this study was to evaluate the phenolic content and the *in vitro* anti-inflammatory properties of different aerial parts of *E. tenuifolia* L.: branches and inflorescences.
Samples were extracted through maceration and fractionated using solvents with increasing polarity. Total phenolic content of both branches and inflorescences was evaluated by means of Folin-Ciocalteau reagent. Total flavonoid content was also determined using a colorimetric method based on the formation of a complex flavonoid-aluminum. Inflorescences showed the highest amount of phenolic compounds (17.5 mg/g of dried plant material). Flavonoid content followed the same trend.
Several studies demonstrated that polyphenols are able to inhibit the inflammation process by regulating the production of pro-inflammatory molecules, such as cytokine (TNF-α) and nitric oxide (NO), which are known to cause host cell death and inflammatory tissue damage.
Nitric oxide reacts with free radicals, such as superoxides, causing the production of harmful peroxynitrates, which can oxidize low-density lipoproteins leading to cell membranes damage. Hence, inhibition of the production of such pro-inflammatory compounds is expected to have therapeutic significance against inflammatory diseases (6).
The ability of alcoholic extracts and their fractions to inhibit nitric oxide production was assessed using LPS-stimulated RAW 264.7 murine macrophage cell line. The crude extract of inflorescences was able to induce a significant inhibitory activity. A concentration 100 µg/ml resulted in 50.14% inhibition, and any cytotoxic effect on treated cells was observed. In contrast, for branches crude extract, cytotoxic activity has been highlighted at all tested concentrations.
The *n*-hexane fraction of inflorescence extract caused a significant inhibition of nitric oxide production (IC_{50} = 17.04 µg/ml), inducing significantly lower cytotoxic effects (IC_{50} = 94.45 µg/ml).
A relevant biological activity was observed for dichloromethane fraction of the same extract, with an IC_{50} value of 39.97 µg/ml. This fraction did not induce cytotoxic effects (IC_{50} > 200 µg/ml).

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4.3 = CHAMOMILE EXTRACTS AS POTENTIAL TREATMENT FOR INFLAMMATORY BOWEL DISEASES

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Inflammatory bowel diseases (IBDs) are chronic disorders characterized by disruption and ulceration of the colon mucosa (ulcerative colitis) or of any part of the digestive tract (Crohn's disease). Although IBDs etiology is still a matter of debate, oxidative stress seems to play a pivotal role (1). Increased oxidative stress has been revealed in multiple studies involving IBDs patients (2,3). On the other hand, antioxidant/anti-inflammatory herbal extract supplementation could represent an innovative approach to contrast IBDs (4,5).
Chamomile has long been used as a medicinal plant in the management of gastrointestinal disorders. Recently, a clinical trial demonstrated the efficacy of a natural formula, containing chamomile, in maintaining remission in patients with ulcerative colitis (6). This is consistent, albeit in part, with the antioxidant and anti-inflammatory properties of chamomile (7).
In this context, the aim of the present study was to explore the possible protective role of chamomile in an experimental model of acute ulcerative colitis in rats exposed to dextran sulfate sodium (DSS). Body weight gain and colon length were determined and the activities of different colonic biomarkers of inflammation and lipid peroxidation, such as myeloperoxidase (MPO), serotonin (5-HT), prostaglandin E2 (PGE2) and 8-iso-prostaglandin F2α (8-iso-PG F2α), were assessed. Our study showed that throughout the 8 days experimentation period, lyophilized chamomile extract (150 mg/kg) was able to reduce classical IBD signs, such as body weight loss (40.1%) and colon length shortening (-20.8%). Moreover, the biochemical assays performed on colon specimens showed that the extract was as able as sulfasalazine in reducing the production of several oxidative stress and inflammatory biomarkers, such as MPO (-26.3%), 5-HT (-84.6%), PGE2 (-54.4%) and 8-iso-PGF2α (-81.9%). In conclusion, in the present work we have investigated the possible efficacy of chamomile in the therapy of IBDs. The observed anti-oxidant and anti-inflammatory effects support a rationale use of chamomile supplementation as a promising pharmacological tool for the prevention and management of ulcerative colitis in humans. Further investigations, comparing different analytical methods and experimental paradigms for detection and quantification of oxidative stress are required to confirm the IBD beneficial activity of chamomile *in vivo*.

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4.3 = BIOLOGICAL PROPERTIES OF THE POLAR EXTRACTS FROM LEAVES AND FLOWERS OF *ISATIS TINCTORIA* L. (BRASSICACEAE) GROWING IN SICILY

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Isatis tinctoria L. (woad) is a herbaceous biennial species belonging to Brassicaceae family. It does grow wild mainly in Southern and North-western Italy, as well as on the major Italian islands. Rural people living around Vulcan Etna (Sicily, Italy) consume boiled the flower buds of this plant as ingredients for salads and omelets (1). Historically, *I. tinctoria* has been cultivated in Europe for the production of indigo dye (blue colour) and it has also been used as a medicinal plant. However, its use as a natural dye has declined, due to the import of cheaper indigo and the disappearance of woad cultures. Nowadays *I. tinctoria* and its closely related species *Isatis indigotica* Fort., are employed in traditional Chinese medicine (2). In the last decades, *in vivo* and *in vitro* studies highlighted the anti-inflammatory and anti-cancer properties of lipophilic extracts and isolated compounds from the plant. In particular some alkaloids, including tryptanthrin, indirubin and indolinone, have been studied for their beneficial effects. However, few investigations have been carried out for studying the potential biological properties of polar constituents. In continuation of our studies on *I. tinctoria*, this work was designed to define and compare some biological properties of the polar constituents of flowers and leaves of *I. tinctoria* grown wild around Acireale (Catania, Sicily, Italy). Moreover, the correlation between biological effects and phenolic compounds was investigated. Cauline leaves were collected in January (It-J) and April (It-A), basal leaves (It-B) in November and flowers (It-F) were harvested in April. All plant material was lyophilized and sequentially extracted with dichloromethane and methanol 70%. The total phenolic, total flavonoid and condensed tannin content of hydroalcoholic extracts was determined spectrophotometrically (3). The total phenolics ranged from 99.36 ± 0.17 (It-A) to 68.60 ± 0.50 mg GAE/g (It-B); the flavonoids ranged from 41.70 ± 0.46 (It-F) to 12.44 ± 0.14 mg QE/g (It-J); the condensed tannins ranged from 27.59 ± 0.21 (It-A) to 10.92 ± 0.47 mg CE/g (It-F). The antioxidant properties of the extracts were examined by different *in vitro* tests (3). In the DPPH test, It-F extract resulted the most active with an IC_{50} value of 0.437 ± 0.003 mg/mL, whilst It-A extract showed the best activity in reducing power (ASE/mL= 1.546 ± 0.006) and ferrous ions chelating activity assays (IC_{50}= 0.564 ± 0.011 mg/mL). Linear regression analysis revealed a positive relationship between total phenolic content and both reducing power and chelating activity, and between tannins and reducing power. Finally, a weak relationship with total flavonoids was highlighted for DPPH only. The potential cytotoxic activity of It-J, It-A, It-B and It-F extracts was tested using *Artemia salina* lethality bioassay (3); all extracts did not display any cytotoxicity against brine shrimp larvae (LC_{50}>1000 µg/mL). The anti-proliferative effect of *I. tinctoria* extracts was evaluated *in vitro* on three human anaplastic thyroid carcinoma (ATC) cell lines (CAL-62, C-643 and 8505C). By MTT assay, a reduced proliferation rate of all cancer cell lines in a concentration- and time-dependent way was observed. In particular, after 48 h of treatment, approximately 20% of growth inhibition ($p<0.05$ *vs* control) was observed in 8505C cells with 0.1-0.5-1 mg/mL of It-J, It-A and It-F, and a stronger inhibiting effect (about 50%) was obtained using It-B at the same concentrations. Similar results were observed in the C-643 cells. The strongest anti-proliferative effect was observed in CAL-62 cells; particularly after 48 h exposure to It-B at the concentration of 1 mg/mL, a decrease in cell growth of 80% was observed ($p<0.001$ *vs* untreated cells). The obtained results suggest that there isn't correlation between the anti-proliferative activity of *I. tinctoria* extracts against ATC cells and the antioxidant phenolic compounds. We previously characterized the glucosinolate profile of It-J and It-A extracts (2); at present LC/MS analysis of glucosinolates contained in It-B and It-F extracts is ongoing in order to establish whether these bioactives are associated with their anti-proliferative effects.

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4.3 = *CITRUS BERGAMIA* RISSO & POITEAU (RUTACEAE) ESSENTIAL OIL EXPLOITED IN STUDIES FOR HUMAN HEALTH APPLICATIONS

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Citrus bergamia Risso et Poiteau (*Citrus aurantium* L. ssp. *bergamia* Engl.), Bergamot, is a species cultivated mostly in a restricted area of Southern Italy (Calabria), though there are crops in South America and the Ivory Coast. *Citrus bergamia* Risso et Poiteau is a tree species that presents an average force with posture ranging from assurgent to expanded. The leaf is large and very similar to lemon in color and shape. The flower of bergamot, and other Esperidee, is generally called "zagara" from the arabic "zahar" which means "flower". The flower, with petals of a beautiful, pure white and of pleasant scent, differs from that of many other species of the genus *Citrus* because the style and the stigma often persist even after the formation of the fruit (1). Bergamot is grown for its essential oil, a product in great demand by perfumery but also by cosmetic, pharmaceutical and food industries, that is obtained by cold pressing the fruit peel. Bergamot essential oil (BEO) comprises a volatile fraction (93-96% of total) containing monoterpene and sesquiterpene hydrocarbons (such as limonene, □-terpinene, □- and □-pinene) and oxygenated derivatives (such as linalool, linalyl acetate, geranial), and a non volatile fraction (4-7% of total) characterized by coumarins and furocoumarins, such as bergapten. Due to the photosensitizing effects of BEO, the essence deprived of bergapten is mainly used in perfumery (1). BEO is increasingly used in aromatherapy for the relief of pain and of symptoms associated with anxiety and depression (2). This latter application may stem from the observed modulation of release of specific amino acid neurotransmitters in discrete brain regions of rat under both basal (3) and pathological conditions (4). BEO also produces a dose-related sequence of sedative and stimulatory behavioral effects in normal rats (5); moreover, it affords neuroprotection against brain ischemic insult (4), and is endowed with analgesic effects (2, 6). *In vitro*, BEO protects cells against an excytotoxic insult (7), or, at higher concentrations, triggers caspase-dependent and -independent death of cancer cells (8). To get further insight into BEO actions we investigated its effects on autophagy, an highly regulated intracellular degradative process whose dysfunction has been implicated in a number of diseases including cancer, neurodegeneration and pain. The results obtained demonstrate that in cultured human cells BEO rapidly modulates, also at non cytotoxic concentrations, biochemical and morphological markers of autophagy. The autophagic process triggered by BEO does not seem to involve the canonical autophagy pathway dependent on beclin-1. Moreover, the upstream signalling events in BEO-induced autophagy appear to be distinct from those induced by serum starvation and converging on mTOR kinase. These findings highlight the complexity of BEO biological activity that needs to be investigated in more detail to provide a rational basis to the widespread use of this natural product.

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4.3 = *ONOPORDUM ILLYRICUM* L.: NEW ANTI-HIV-1 AGENTS FROM AN EDIBLE MEDITERRANEAN PLANT

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Onopordum illyricum L. is a wild plant belonging to Compositae family spread in the Mediterranean region, in Portugal and Albania (1). It's very common in Sardinia where its young scapes and capitula are eaten raw in salad as side dishes (2), representing a food of good nutritional value. In the traditional medicine a decoction or tea of the whole plant is used as a digestive and to treat cough and biliary diseases (2); the flowering tops are used as a febrifuge for the treatment of malarial fever and for washing exanthematic skin (3).
Within a project aiming to find new agents inhibiting the replication of human immunodeficiency virus type 1 (HIV-1) from the Sardinian flora, the ethanolic extract and the ethyl acetate, n-hexane, butanol and water fractions obtained from aerial parts of *O. illyricum* have been assayed on the HIV-1 reverse transcriptase (RT) associated ribonuclease H (RNase-H) activity, a multifunctional viral enzyme that is still a good target because, at the moment, in the antiretroviral therapy are not present drugs that target this function. Since the human immunodeficiency virus (HIV) has been established to be the etiological agent of the acquired immunodeficiency syndrome (AIDS) (4,5), an originally unpredicted number of drugs have been approved for the treatment of the HIV infected patients (6). The management of this disease, however, is still complex and worrisome due to problems such as monitoring of therapy efficacy, chronic administration drug toxicity, poor tolerability, drug resistance development or therapy adjustment after treatment failures (7). For all these reasons the search for new inhibitors with novel mechanism of action and effective on HIV drug-resistant strains is still a worldwide health care issue (8,9).
In this work the ethanolic extract obtained from aerial parts of *O. illyricum* have been tested on the HIV-1 RT associated RNase-H function on *in vitro* biochemical assay and it showed an IC_{50} value of 8.84 ± 0.53 µg/mL. Given that relevant and selective activity relates to IC_{50} values below 100 µg/ml for extracts and below 25 µM for pure compounds (10), *O. illyricum* extract have showed a significant antiviral activity. The extract was fractionated and the butanolic fraction was the most active one, with a similar inhibition value to the total extract (IC_{50} = 8.97 ± 0.13 µg/mL). Previous studies reported for this plant the presence of fatty acids, sesquiterpene lactones, triterpenes, polyphenols and caffeoylquinic acid derivatives (11,12,13). According to them, luteolin, apigenin, hispidulin, arctin, 1,5-dicaffeoylquinic acid, onopordopicrin, deoxyonopordopicrin were isolated from the butanolic fraction and their inhibitory effect was evaluated on RNase-H activity. In particular, luteolin, apigenin, arctin, and, deoxyonopordopicrin inhibited the HIV-1 RNase-H function with IC_{50} values between 56 and 70 µg/mL. Differently, onopordopicrin showed an IC_{50} value of 16 µg/mL. The most active compounds, hispidulin and 1,5-dicaffeoylquinic acid, inhibited the HIV-1 RNase-H activity with IC_{50} values of 8,68 and 3,66 µg/mL, respectively. We have identified new natural derivatives that are able to inhibit the HIV-1 RNase H activity.

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4.3 = DISCOVERING HOP IN ITALY

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Humulus lupulus L. is a medicinal plant whose therapeutic and healthy values are confirmed by a well-established use in traditional medicine and by biological activities published in literature. These findings are summarized in the monographs prepared by WHO (2001) and ESCOP (2003) and allowed the inclusion of dried inflorescences of hops in the European Pharmacopoeia, though its main standing is closely linked to the production of beer. In recent years, Italy is progressively re-discovering hops, both for an excellent crafts production of beer and the growing attention of consumers towards high quality local products. Those facts and the lack of characterization of cultivars grown in Italy lead to botanical, genetic and phytochemical studies aimed at description of features of hop cultivated in Italy starting from foreign or own cultivars, and selection of autochthone cultivars to be use in brewery. In this context, we carried out a two-year phytochemical study of two foreign cultivars grown in Northern Italy to evaluate influences of in-field practice on bitter acid composition, volatile organic fractions and polyphenols, particularly for their potential application in traditional medicine.
From a comparison between samplings of the two years, it is apparent that Saaz hop revealed a good stability in its volatile emission. The main compound present in its headspace was myrcene (84.4 and 89.1% in the first and in the second year, respectively), followed by β-pinene (2.0 and 2.6%), α-humulene (5.9 and 2.1%) and β-caryophyllene (2.9 and 1.1%). On the contrary, the Cascade hop showed a very variable profile of emission. In the first year, the main volatiles detected were myrcene (29.1%), β-caryophyllene (28.1%) and α-humulene (27.0%). The same compounds passed in the second year to 93.0, 0.4 and 0.5%, respectively. This trend was also observed for the chemical classes of volatiles. For Saaz the most represented one was that of monoterpene hydrocarbons (88.3 and 94.0%). Conversely, for Cascade hop the main chemicals emitted during the first year were sesquiterpene hydrocarbons (77.7%), while in the second one they were replaced by monoterpene hydrocarbons (97.7%). The two harvests evidenced also different quantitative composition of both bitter acids and polyphenols.

4.3 = *CRYPTOTAENIA THOMASII* (TEN.) DC. (APIACEAE) *N*-HEXANE EXTRACT AS A SOURCE OF POTENTIAL ANTICHOLINESTERASE INHIBITORS

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Plants belonging to the family Apiaceae contain an exclusive group of secondary metabolites, known as sources of important biological activities for the treatment of several diseases including Alzheimer's disease (AD) (1, 2). AD is a progressive neurodegenerative disease, wherein a progressive loss of cholinergic synapses occurs in neo-cortex hippocampus. Decreased concentration of the neurotransmitter acetylcholine appears to be a critical element in the development of dementia, and a therapeutic approach to treat AD and other form of dementia is to restore acetylcholine levels by inhibiting both major form of cholinesterase: acetylcholinesterase (AChE) and butyrylcholinesterase (BChE). In recent years, researches have focused their attention towards finding cholinesterase inhibitors from natural products. Many plant-derived compounds, particularly terpenoids, have been studied as new cholinesterase inhibitors.
In the present work, the *n*-hexane extract of the aerial parts of *Cryptotaenia thomasii* (Ten.) DC. (Apiaceae, subfamily Apioideae, tribe Oenantheae) collected in Sila Grande, Calabria (Italy), was investigated for its AChE inhibitory activity using the *in vitro* Ellman's method (3).
C. thomasii is a very distinctive diploid species, belonging to a genus with transoceanic disjunctions (4). This species is endemic to Southern Italy in Basilicata and Calabria (5, 6), and it is the only representative of the genus *Cryptotaenia* in Europe.
The extract inhibited AChE activity in a concentration-dependent manner with an IC_{50} value of 144.31 □g/ml. To identify the putative active compounds present within the *C. thomasii* non-polar extract, gas chromatography (GC) and gas chromatography-mass spectrometry (GC-MS) analyses were performed (7). The extract was characterized by the presence of sesquiterpenes, diterpenes, sterols, and fatty acids and their methyl esters as main classes of compounds. Among them the most abundant were stigmast-5-en-3-ol, phytol, linolenic acid methyl ester, and neophytadiene. Moreover, taking into account the role of free radical-induced oxidative damage in the pathogenesis of AD, the *in vitro* radical scavenging activity was also investigated by using the 2,2-diphenyl-1-picrylhydrazyl radical (DPPH) assay (8).
The obtained results demonstrated the health promoting properties of *C. thomasii* and its potential role for the treatment of neurodegenerative diseases, like AD.
C. thomasii could be a good candidate for further studies to isolate cholinesterases inhibitors.

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4.3 = PHENOLIC PROFILE AND BIOLOGICAL ACTIVITY OF ACHILLEA MOSCHATA WULFEN FROM ITALIAN AND SWISS ALPS

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The genus *Achillea* belongs to Asteraceae family and consists of more than 100 species worldwide. *Achillea moschata* Wulfen is one of the twenty-four species recognized in Italy (1). It grows on siliceous rocks, screes and stony pastures, along the Alps up to 3,400 m a.s.l., where it is extensively used by local people for its medicinal properties, as reported in recent ethnobotanical studies (2, 3). The two analyzed samples were collected, at flowering stage, in Val Mustair (Switzerland - 2,300 m a.s.l.) and in Valchiavenna (Italy - 2,200 m a.s.l.), respectively. Aerial parts were defatted with petroleum ether (PET) and successively extracted with dichloromethane (DCM), and methanol (MeOH). HPLC and ESI-MS/MS analyses of the methanolic extracts evidenced a series of flavonoidic and phenolics compounds. The results showed that flavonoids were the predominant compounds than phenolics in both samples (89.5 µg/mg DW and 33 µg/mg DW, respectively, in *A. moschata* Valchiavenna; 82.5 µg/mg DW and 40 µg/mg DW, respectively, in *A. moschata* Val Mustair). Among flavonoid derivatives, luteolin and apigenin were the predominant aglycones, free and glycosilated. *A. moschata* Valchiavenna extract was characterized by apigenin as main compound (60.36 µg/mg DW) followed by a caffeic derivative -, disuccinylcaffeoyl quinic acid (25.23 µg/mg DW). Apigenin-7-O-glucoside represented the principal metabolite of the *A. moschata* Val Mustair (44.66 µg/mg DW) followed by 4.5-O-dicaffeoyl quinic acid. The antioxidant activity of all obtained extracts was tested by **in vitro** methods using 2,2-diphenyl-1-picrylhydrazil (DPPH) and [(2,2'-azinobis (3-ethylbenzothiazoline-6-sulfonic acid)] radical-scavenging assays. The data obtained from both samples proved their increasing scavenger capacity in relation to the extract polarity (PET extract < DCM extract < MeOH extract), and a good correlation between the two employed methods. The extracts were also investigated against three Gram-positive (*Bacillus cereus*, *Enterococcus faecalis*, *Staphylococcus aureus*) and three Gram-negative (*Escherichia coli*, *Proteus mirabilis*, *Pseudomonas aeruginosa*) bacterial species using the disc diffusion assay. Antimicrobial activity of the extracts was expressed according to inhibition zone diameter. DMC and PET were the most active extracts (inhibition diameter ≥ 12 mm).

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4.4 = VEGETABLE WASTE FOR MEDICAL AND COSMETICS DEVICE: THE COFFEE PLANT EXAMPLE

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The re-use of vegetable waste is a process that can improve economic efficiency and can optimize agriculture's ecological impact. For examples coconut fibers and pine bark are used as manure for plants, whereas rice hulls are used as building material. However, the latest food trend outline the use of vegetal waste to obtain bioactive compounds useful for cosmetics and medical devices. In this context, we focused our attention on the vegetal residues obtained from coffee production process to investigate the occurrence of bioactive secondary metabolites from waste residues. Previous analyses suggested that *Coffea arabica* is rich of antioxidant and anti-inflammatory compounds occurring in seeds as well as in fruit and leaves. These metabolites are interesting for organic cosmetic formulation for topical use (1). In our work we analyzed several samples of *C. arabica* seeds and fruits collected in farm of El Salvador. Vegetal samples were subject to both hydroalcoholic and lyophilization extraction. The resulting extracts were analyzed by UPLC/ESI-HMRS to qualitative and quantitative evaluation of secondary metabolites. The most abundant molecule is chlorogenic acid (CGA) which is the ester of caffeic acid and quinic acid. Reference data suggested that CGA showed antioxidant activity (2); therefore we tested the antioxidant action of different coffee extracts with several chemical tests: hydrogen-atom-transfer-based assays (oxygen radical absorbance capacity or ORAC) and electron-transfer-based assays (as DPPH assay Trolox equivalence antioxidant capacity assay or TEAC, total phenols assay by Folin-Ciocalteu reagent). Results suggested that different coffee cultivars and plant portions show relevant differences in the concentrations of CGA and antioxidant activity. We conclude that coffee extracts are an important source of antioxidants for cosmetic sector for examples in sunscreens formulations to replace the common sunscreen synthetic molecules.
Our work will aim at identifying the coffee waste residues with the most amount of CGA as well as other secondary metabolites with antioxidant activity and to test their efficacy as active compounds in sunscreen formulation.

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4.4 = PLANT ANTIOXIDANTS IN THE PROTECTION AGAINST RADIATION DAMAGE

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The scientific community agrees to recognize a link between oxidative stress, mainly due to hyperaccumulation of free radicals, and many serious diseases, such as cancer, atherosclerosis, hypertension, Parkinson's disease, Alzheimer's disease, diabetes mellitus, colitis and rheumatoid arthritis (1). It is known that the levels of oxidative stress may decrease considerably following the introduction of antioxidant molecules, contained in foods, dietary supplements or drugs (2). The molecules able to decrease the levels of oxidative stress belong particularly to the phenols and carotenoids. This research is aimed to develop food supplements containing mixtures of antioxidant plant compounds, with proven non-toxicity, to be used in the prevention of damage from oxidative stress to which are subjected the astronauts during space missions, as a result of exposure to cosmic radiation and to numerous not optimal environmental factors. The obtained formulations can also be used in other fields where there is an increase in oxidative stress in the human body, such as exposure to ionizing radiation at radio-diagnostic purpose or radio-therapy (3). This study is part of the project PAPARD (2014-034 R.0 financed by ASI). For the preparation of the formulations will be used a pool of bioactive molecules most widely studied by our research group (4). It will be evaluated the effectiveness of different mixtures consisting of: catechins and procyanidins, resveratrol and its oligomers (viniferins), hydroxy-tyrosol free and/or linked to secoiridoidic molecules, anthocyanins obtained from red grapes and red fruits and ascorbic acid (vitamin C). All the molecules selected for this study are already present as components of food and/or food supplements and known for their antioxidant properties. The different formulations will be evaluated for their antioxidant capacity and prevention from oxidative damage in two model systems (e.g. erythrocytes and human intestinal tissue); it will be also investigated the stability of the molecules in time, when subjected to irradiation with ionizing radiation. The doses used will be from 0.01 to 1 Sv, so as to cover both the simulation of exposure radiodiagnosis and radiotherapy (up to 0.07 Sv), it is the one that characterizes the space missions of long duration (up to 1 Sv).
The research group coordinated by Prof. Gabriella Pasqua performed chemical extraction on the seeds from grapes cvs. Michele Palieri (red), Italia (white), and Pinot. Preliminary observations have shown that the most effective solvent for the extraction of the proanthocyanidins is mixture of ethanol and water at 70:30. The extracts were added with maltodextrin and inulin, due to the its use as fiber and its antioxidant properties (5) at a concentration equal to 5% to obtain a fine powder. In collaboration with the group coordinated by Prof. Nadia Mulinacci have been carried out the HPLC/DAD/MS analysis. The chromatographic profiles showed that the cvs. M. Palieri and Italia were the richest in proanthocyanidins both oligomers and polymers. The group coordinated by Prof. De Gara conducted a preliminary analysis of the total antioxidant capacity of a first preparation of extracts of *Vitis vinifera* cv Michele Palieri (without excipients and added with 5% inulin and maltodextrin). The results of the preliminary experiment showed a very high antioxidant activity of the extracts, comparable to the range of ascorbic acid or vitamin C (ASC) and glutathione (GSH), two potent biological antioxidants.

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4.4 = RADICAL SCAVENGING AND HYPOGLYCAEMIC POTENTIAL EFFECTS OF AN EDIBLE FLOWERS EXTRACT FROM *BORAGO OFFICINALIS* L. (BORAGINACEAE)

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Edible flowers are commonly used worldwide to improve appearance and nutritive value of meals. Flowers are served as a salad, to prepare cakes and drinks or as a side dish (1). Flowers are a rich source of healthy phytochemicals including flavonoids, anthocyanins and other phenols. Diabetes has emerged as a major threat to health worldwide. In 2014, 9% of adults 18 years and older had diabetes. In 2012 diabetes was the direct cause of 1.5 million deaths. More than 80% of diabetes deaths occur in low- and middle-income countries. Type 2 diabetes or non-insulin-dependent or adult-onset results from the body's ineffective use of insulin. There is growing evidence that the excess generation of Reactive Oxygen Species (ROS), largely due to hyperglycaemia as a consequence of glycation reaction, causes oxidative stress in a variety of tissues. In type 2 diabetic patients, oxidative stress is closely associated with chronic inflammation that may play a role in the development of complications in diabetes (2, 3).
Borago officinalis L. (Boraginaceae) (known as borage, burrage, bourrache, and bugloss) grows in Mediterranean region, Europe and Northern Asia. Traditionally *B. officinalis* is used in traditional medicine to treat gastrointestinal, respiratory and cardiovascular disorders (4).
In this research we have investigated *B. officinalis* flowers for their potential health benefits as source of radical scavenger and hypoglycaemic agents. Fresh flowers were exhaustively extracted with ethanol by maceration. The extract exhibited a total phenols content of 28.8 mg chlorogenic acid/g dry extract evaluated by Folin-Ciocalteu method. The scavenging capacities of *B. officinalis* flowers extract were measured and compared. *B. officinalis* showed a promising radical scavenging ability in both test with IC_{50} values of 30.2 and 44.3 µg/mL for DPPH and ABTS, respectively. The lowering of post-prandial hyperglycaemia through the inhibition of key-enzymes linked to type 2 diabetes mellitus is a critical therapeutic strategy used to control type 2 diabetes (5). In this study, the ability of flowers extract to inhibit □-amylase and □-glucosidase enzymes is presented. The ethanol extract showed a selective □-amylase inhibitory activity with an IC_{50} value of 23.9 µg/mL. In conclusion, this study supports the use of *B. officinalis* edible flowers in diet as healthy foods.

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4.4 = GLUTEN-FREE FLOUR DOUGHS: TEXTURE IMPROVEMENT BY PROTEIN CROSS-LINKS OCCURRING VIA MICROBIAL TRANSGLUTAMINASE FROM *STREPTOVERTICILLIUM MOBARAENSE*

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Gluten is the major factor involved in the structural properties of bakery products. Thanks to its ability to create aggregates, it gives dough viscosity, elasticity and cohesion, all features responsible for the baking performance (1). Gluten is composed of extensible, viscous gliadins and rigid, elastic glutenins. Moreover, the gliadin protein fraction is the main factor responsible for the development of celiac disease (CD) and other non-celiac gluten sensitivities. The only treatment for these people is lifelong adherence to a strict gluten-free diet (2). Nowadays, the development of new technologies in the food industry aims to both improve products already marketed, and to develop new gluten-free products. Cross-linking enzymes, able to organize and create protein networks, are suitable for the food industry as protein modifiers. Among those enzymes, Transglutaminase (TGase), due to its ability to improve the firmness, viscosity, elasticity and water-binding capacity of food products, could be the key factor for manipulation in order to achieve higher food quality (3). The aim of this study was to evaluate the effects of addition of microbial transglutaminase from *Streptoverticillium mobaraense*, on protein aggregation in gluten free flours as well as in baked products. In our research we assessed five gluten free flours deriving from different plant sources such as cereals, pseudo-cereals and legumes; in particular corn, rice, amaranth, quinoa and lentil flours have been analyzed. By biochemical tests, flours' total extracted proteins capacity to act as TGase substrate was evaluated. Since all tested flours showed considerable transamidase activity in microplate colorimetric assay, the possible formation of crosslinked products were analyzed by SDS-PAGE. For this purpose, micro-doughs prepared by adding the water, in 1:2 ratio (w/v), to different flours were treated with different amount of TGase enzyme (0.5; 0.75; 1; 2 Units). To clarify which protein fraction of the flours was the best substrate for the TGase, differential extractions were performed. Therefore, three sequential fractions, corresponding to albumins/globulins, prolamins and glutelins, were analyzed in relation to different enzyme amounts. Results showed that the microbial TGase has capacity to modify gluten free flour proteins and determine protein network formation. In particular, the lowest enzyme concentration (0.5 U) was able to cause differences in protein bands between treated samples and their non-treated controls. These preliminary results give a perspective in the gluten-free research and suggest their possible use to create innovative products with improved texture.

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4.5 = *IN VITRO* STUDY ON NEW DRUGS AND FOOD SUPPLEMENTS FROM PLANTS USED IN SAHRAWI (WESTERN SAHARA) CULINARY AND MEDICINAL TRADITION

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Sahrawi population subsists on humanitarian aids. In recent years, thanks to cooperation projects, a laboratory has been set-up to produce drugs used for the treatment of infections, in particular caused by bacteria and viruses, that with different etiology affect gastro-intestinal tract and respiratory ways seasonally, and for the treatment of some pathologies as diabetes or - vitamin A and C, Fe and Zn salts deficiency due to food shortages (1).

The inconstant flow of these humanitarian aids, that usually allow the production in the laboratory, could cause the dearth of some active principles preventing the preparation of drugs, essential for the population. Therein lies the need to recover the basics of the traditional ethnic medicine, related to the plants used, in order to support pharmacology treatments depending on humanitarian aids and to solve problems connected to this shortage. This project also moves in the direction of the biodiversity preservation and enhancement, an increasingly emergent international topic in the debates about climate, environment and health. Furthermore the complementarity of this traditional medicine could aid to minimise risks related to drug-resistant of the infectious diseases.

Nowadays there is a small number of scientific and ethnobotanical papers about Western Sahara medicinal plants traditionally used by Saharawi population (2). In the first step of this research project we have selected plants used in culinary and medicinal tradition. Preliminary results for *Ammodaucus leucotrichus* Coss. & Dur. (Apiaceae) fruits, *Artiplex halimus* L. (Chenopodiaceae) leaves highlighted good nutritional values with particular reference to high level of macronutrients (K, Na, Ca, Mg) and micronutrients (Fe, Mn). The chemical composition showed a good content of total polyphenols for *A. leucotrichus*, also characterized by 4% of lipids, with 86% of oleic acid and a high amount of volatile components. The essential oil obtained from fruits was mainly composed by perillaldehyde (80%). *A. halimus*, particularly rich in Na (110 mg/g), highlighted an interesting profile of the small amount of lipids (1.5%), characterized by linoleic (18.91%) and linolenic acid (16.46%). The decoctions, the traditional preparations, didn't evidence antioxidant and antimicrobial activities for all drugs, with the exception of essential oil of *A. leucotrichus* that showed good properties against *C. albicans* (MIC=156 µg/mL) and high fungistatic capacities versus phytopathogens. Enzymatic tests related to diabetes are currently in progress.

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4.5 = MORPHO-HISTOLOGICAL CHARACTERIZATION AND NUTRITIONAL PROPERTIES OF THE PRICKLY PEAR [*OPUNTIA FICUS-INDICA* (L.) MILL]

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Opuntia ficus indica (L.) is a l- succulent shrub, native of the Mexico; it has been introduced in 16th century in several continents and now it is widely grown in the warmer parts of the world. In some countries as Mexico, Italy and South Africa, the Prickly pear is cultivated on considerable surfaces for industrial ends because of their edible fruits. Several searches show that the Prickly pear fruit is very rich in vitamins, minerals, amino acids and in sugars. It is used as foodstuffs, for medical applications and cosmetics.
Another important compositional factor of prickly pear is the presence of pigments, as betalains, which gives particular attractiveness to fruit and products. Betalains are vacuolar pigments present in all varieties of reported *O. ficus indica*; their main function in the plant is the attraction of animals for pollen transfer in order to ease plant propagation and to avoid the damages of UV radiation. Two betalain derivatives are present in cactus-pears: betacyanin, which gives the red-purple color, and betaxanthin, which gives a yellow-orange color. These pigments show important antioxidant activities without toxic effects in humans. Currently the *O. ficus indica* is naturalized throughout the Mediterranean basin and in the temperate zones of America, Africa, Asia and Oceania. In Italy and Israel it is planted for fruit consumption. It is also used for ornamental purpose, wind protection fencing, land reclamation and rehabilitation, and erosion control. In Italy the 90% of prickly pear cultivation is located in Sicily, where there are three production centers, two of which were awarded by the Community trade mark D.O.P.
Given the considerable interest aroused by this species, the aim of our research is to study the morphological and cyto-histological characteristics of the fruit from two different varieties of the prickly pear "*Sanguigna*" and "*Muscaredda*" o "*Sciannarina*". The quantification of some functional compounds of Prickly pear as the betalains, the carotenoids and chlorophyll (*a* and *b*) were also carried out.

4.5 = NATIVE TAXA OF SICILIAN FLORA UTILIZED AS VEGETABLES

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In the last forty years the eating habits of Sicilian people, like those of the populations living in the Western countries, have greatly changed. Young people prefer new food flavors created by industries, while those of wild vegetables are almost unknown.
The elders and those who still have strong links with the country follow a Mediterranean-style diet instead. They know the seasons of wild vegetables and they are able to recognize and cook them according to established traditional methodologies.
People used to go almost daily to the countryside, margins of cultivated fields and woods, looking for wild vegetables.
This alimentary habit derived substantially from the situation of poverty in which the majority of the rural and urban population lived.
In order to preserve the important cultural patrimony linked to the employment of taxa of indigenous flora as food, some years ago a study of wild plants utilized traditionally as vegetables started in Sicily.
For this purpose, besides the consideration of the available literature (1, 2, 3, 4), numerous surveys have been carried out in different areas of the Island through interviews to shepherds, farmers, loggers, etc.; they led to the identification of edible taxa directly in the collection localities and indicated for each of them dialectal name, collection procedures and culinary preparations, as well as any popular beliefs.
All the acquired data were processed and some reports were drawn up, in which there are for each plant: the scientific name, synonyms, Italian names, Sicilian names, the main botanical characters, the properties and medicinal uses, the edible parts, the collection procedures, the traditional food uses, any merchantability, notes and curiosities, any warnings for the consumption.
The research showed that the wild species used as vegetables are about two hundred. These are mainly hemicryptophytes, therophytes and geophytes, belonging to 37 families; the most widespread ones turned out to be: Asteraceae with 31 genera and 80 infraspecific taxa; Brassicaceae with 16 genera and 28 taxa; Umbelliferae with 9 genera and 11 taxa; Fabaceae with 4 genera and 11 taxa; Alliaceae with 1 genera and 6 taxa; Valerianaceae with 3 genera and 4 taxa; Liliaceae with 3 genera and 4 taxa; Malvaceae with 2 genera and 4 taxa; Asparagaceae, Plantaginaceae and Urticaceae with 1 genus and 4 taxa; Boraginaceae with 3 genera and 3 taxa; Caryophyllaceae, Chenopodiaceae and Solanaceae with 2 genera and 3 taxa, etc.
It was also found that the aerial parts of wild vegetables are mainly collected, while the subterranean parts are less considered.
It sometimes happens that multiple parts of a plant are utilized; in particular the leaves (41.5%), the young shoots (37%), the basal rosette (31.5%), the flowers and inflorescences (10%), the bulbs (5%), the turions (3%).
Wild vegetables, rich in functional nutrients, still represent an important resource for the territories in which they grow since they can enrich the table of strong flavors, bitter or delicate, giving unique taste sensations: rustic, primitive, rough but genuine, able to join man and nature.
When skillfully collected by expert hands, they can become "healthy and genuine" ingredients for delicious local traditional and ancient recipes, fundamental for the revitalization or the reinforcement of quality food, strictly connected to the bio-territory.

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5. = *PERENNIPORIA FRAXINEA* POPULATION: A CASE OF STUDY IN AN URBAN PARK AT PAVIA (ITALY)

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Perenniporia fraxinea (Bull.) Ryv. is either a parasite or a saprotrophic fungus growing on a wide variety of hardwood trees species. Sporomata are perennial, pileate, large, single or imbricate, woody. The fungus is reported in forests, gardens and parks, but also on street trees where it usually causes white rot on the lowest parts of the stem, rarely on emerging roots (1). Therefore, this species can affect the stability of trees, with possible consequences for people and properties.
The populations of pathogens like *P. fraxinea* are often studied by using molecular techniques associated with morphological studies such as somatic incompatibility tests (2). Despite its wide distribution, areas where *P. fraxinea* sporomata are abundant look uncommon, so that intra- and interpopulation comparison can be hard to perform (3). In Pavia municipality (Lombardy, Italy) this species is abundant and is likely to be one of the most widespread tree pathogens in urban areas.
The present investigation aimed at better understanding *P. fraxinea* populations. In particular, genetic variation among *P. fraxinea* isolates collected from sporomata found on closely located trees was investigated, coupling molecular analyses with somatic incompatibility tests.
Sporomata were mostly collected in Pavia municipality, namely "Parco della Vernavola" (the widest park at north of the town); a few samples were also collected from surrounding urban sites and from other Italian municipalities as outsiders (Zerbolò and Santa Maria della Versa in Pavia Province; Illasi in Verona Province).
Analyses through RAMS (random amplified microsatellites) pointed out a high genetic diversity among isolates even if they were collected only few meters far from each other. This finding may suggest the spread through root contacts is unlikely for this fungus. Somatic incompatibility tests confirmed the diversity, but failed to distinguish all genotypes identified through RAMs.
A significant correlation between genetic diversity and spatial distribution of isolates collected in "Parco della Vernavola" was observed.
This is one of the first reports on the populations of *P. fraxinea*.

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5. = ARE SHOOT FEATURES OF ACTUAL DIAGNOSTIC VALUE IN EUROPEAN BLADDERWORTS (*UTRICULARIA* L., LENTIBULARIACEAE)?

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In Europe, seven species of *Utricularia* L. (bladderworts) occur: *U. australis* R.Br., *U. bremii* Heer, *U. intermedia* Hayne, *U. minor* L., *U. ochroleuca* Hartman, *U. stygia* Thor, and *U. vulgaris* L. These species are mainly distinguished by the flower features (1, 2). As concerns the vegetative parts (shoots), we can subdivide all species in three aggregates: A) *U. intermedia* aggr., also including *U. ochroleuca* and *U. stygia*, B) *U. minor* aggr., also including *U. bremii* and C) *U. vulgaris* aggr., also including *U. australis*. Within the same aggregate, the species share very similar shoot morphologies, so that distinguishing them without flowers is hard. However, several authors (1, 2, 3, 4, 5, 6) described shoot features that might help to discriminate units, even without flowers. Indeed, many populations of these species do not flower or rarely do (2). Concerning the use of vegetative parts as a diagnostic tool, we tested their reliability from a statistical point of view. For species in aggregate A, we investigated the teeth along the ultimate segment of the leaf margin and the angle at the apex of ultimate leaf segments. For species in aggregate C, we studied the ratio of the setula length to teeth (from which setulae arise) length. We also investigated bladders' quadrifid glands for all the European species, performing a geometric morphometrics analysis. This analysis transforms a descriptive character, as the shape, in Cartesian coordinates, making possible the use of multivariate statistics. Besides shape, also the centroid size (a proxy of their absolute dimensions) of the glands was calculated.

Thor (1988) built an identification key using only features of quadrifid glands for discriminating the six Scandinavian species (*U. bremii* not occurring in this peninsula). We attempted to extend Thor's approach to all European species and using geometric morphometrics. Geometric morphometrics on quadrifid glands' shape (all species together) did not allow any species discrimination, except separating aggregate B from the rest of the species. Thus, our result severely questions Thor's identification key.

For *U. intermedia* aggr., results showed that number of teeth on the leaf margin appears to be as a good diagnostic feature, especially if combined with other features, such as the angle on the tip of the leaf segments. Our study confirms the trend found by (4, 5): the three species overlap concerning glands' shape, but averaging measurements by traps and individuals, a certain separation between species was found. Typical quadrifid glands of *U. intermedia* bear four parallel arms, but we found a large variability also within the same trap, already reported for all species belonging to this aggregate (4, 5) and largely confirmed here. Thus, some tools for a reliable identification based on shoot features do exist for this aggregate.

Centroid size of quadrifid glands in *U. minor* aggr. was unsuccessful for distinguishing species, not supporting the statements of (6), according to which the species in this aggregate would have different sizes of quadrifid glands. The shape of quadrifid glands in the two species resulted very similar, and intraspecific was higher than interspecific variability.

Concerning aggr. C, *U. vulgaris* bears a higher ratio of setula length to tooth length than *U. australis*, which appears more toothed. Actually, in *U. australis* it is rare to find setulae arising directly from the leaf margin, which always bears teeth, whereas in *U. vulgaris* it is quite common to find setulae directly arising from the margin. This feature may help to distinguish species, but it would be more effective if paired with other characters. One of the investigated populations bears intermediate values between *U. australis* and *U. vulgaris*, possibly representing a hybrid. Quadrifid glands' shape resulted very similar between species, and intraspecific was higher than interspecific variability.

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5. = SHAPE MATTERS IN SAMPLING PLANT DIVERSITY: EVIDENCE FROM THE FIELD

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The identification of shape and size of sampling units maximising species richness has major implications in conservation planning and monitoring actions. In this study we tested the effect of three sampling unit shapes (rectangles, squared, and randomly shaped sampling units) on the number of recorded plant species. We used a large dataset gathered in the network of protected areas of the Siena Province, Italy, as part of a joint project with the Italian National Forest Inventory. This dataset is composed of plant species occurrence data recorded from 604 plots (10m x10m), each divided in a grid of 16 contiguous subplot units (2.5 m x 2.5 m). Furthermore, we evaluated the effect of plot orientation along the main environmental gradient, related to slope, to understand how the selection of plot orientation (when elongated plots are used) influences the number of species collected. In total, 1041 plant species were recorded from the study plots. Species richness was significantly higher using randomly shaped sampling units compared to rectangular and squared units. Although rectangular units captured a significantly larger number of species than squared ones, plot orientation along the main environmental gradient (terrain slope) did not show a systematic effect on the number of recorded species. The decision of whether or not using rectangular *versus* squared plots should therefore depend upon the objectives of the survey and the characteristics of the area to be surveyed, with squared plots being more suitable for assessing species composition of more homogeneous vegetation units while rectangular plots being more suited for recording more species in the pooled sample of a large area. If the aim of a survey is to evaluate the biodiversity and conservation value of a ecosystem/community, sampling protocols that maximise the number of recorded species given the same sampling effort should be used. Furthermore, considerations on the orientation of rectangular plots should be carefully made to enhance reproducibility of collected data and correctly interpret the results of a survey. This information is crucial to develop cost-effective surveys and monitoring programs aimed at maximising the number of collected species.

5. = BURNED AREAS DETECTION AND FIRE WEATHER. A CASE STUDY

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Forests hold a fundamental role as biodiversity sources, as interchange medium between earth surface and atmosphere through photosynthetic and respiration processes and as territory protection in the hydrologic sphere. Therefore the burned areas monitoring and the study of fire danger turn out to be topical subjects.
The study is focused on the detection of burned areas and the evaluation of the fire danger. The first aim was to extract the areas affected by fire in a semiautomatic way from Landsat images. The second objective was to evaluate the use of the Fire Weather Index (FWI) (1, 2) to define the fire danger, as the weather factors influence a lot the fire phenomenon, because they can affect on their ignition and propagation.
The study was carried out in the Liguria region (NW-Italy). The Automatic Burned Area Detection Software (ABAMS) (3) was selected as method of detection and the best indices to be utilized in his two phases (seed and growing) were chosen. The burned areas, extracted in the first part of the research, and the FWI daily data were studied considering a period of four years (2000-2003). The statistical analysis of U Mann-Whitney was applied to verify the relation between burned areas and weather indices.
The results of the test seem to show that the FWI and the Drought Code (DC) can explain well the fires event and their extension in the Liguria region. According to this analysis, a first definition of the indices thresholds that could be used to discriminate among classes of danger was made.
The study highlights the connection of fires frequency and extension with the weather conditions, confirming that these factors, combined in a meteorological index, can be useful to estimate the fire danger.

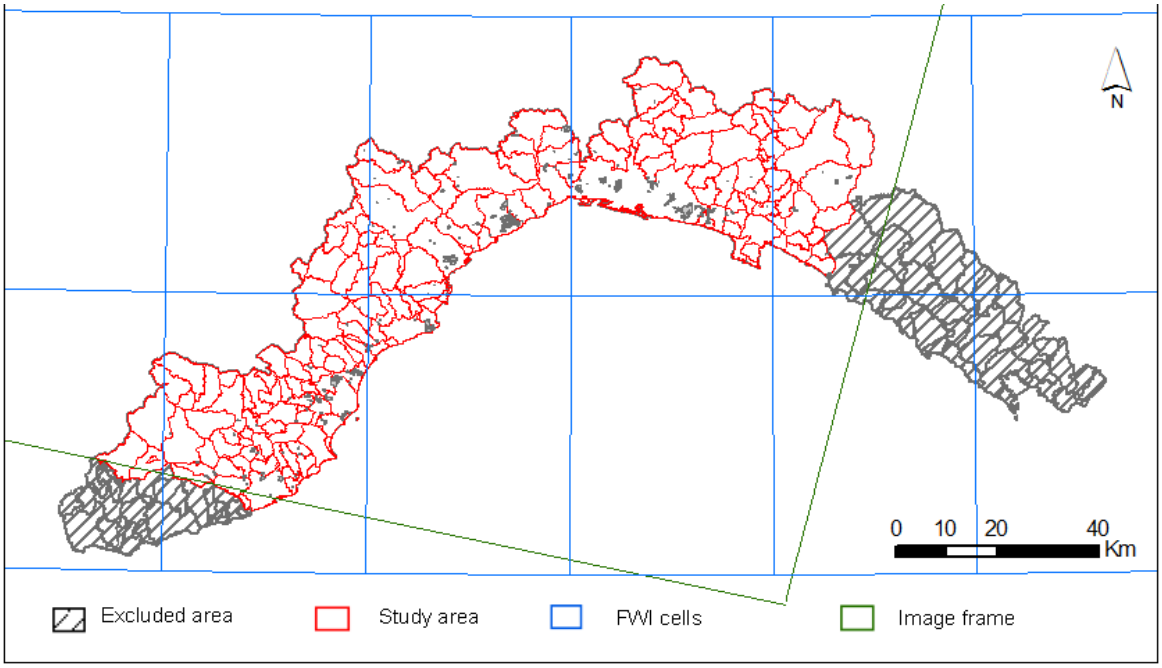

Fig. 1 Study area and obtained results (burned areas in black and gray)

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5. = VASCULAR PLANTS IN A MEDITERRANEAN PINE COASTAL FOREST: WHICH DRIVER PREVAILS?

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Coastal forests of *Pinus pinea* L. (Fig. 1) are included in the ‘‘Habitats Directive’’ (HD, 92/43 EEC) as priority habitat. The need to increase knowledge of this understudied habitat is a priority. These forests provide important functions by stabilizing sand dunes and defending back crops by sea-salt spray (1). Besides all, they acquired over the years an important recreative role although the spread of human settlements in coastal areas has also increased environmental concerns. The importance of this biota has always been underestimated by plant ecologists who have always considered it unattractive because of the presence of non-natural stone pine stands. Our goal was to examine how environmental conditions affect the composition and diversity of vascular plant in areas within the habitat 2270*. Three localities within protected areas along the Tyrrhenian coast were sampled in 2014 in both freely accessible and fenced zones. Sampling was done by means of 30 plots (100 m^2) using a stratified (open-closed) random sampling design. In each site all vascular plants were surveyed. In addition, soil chemical parameters (pH, electrical conductivity) were examined as well as distance from the sea later measured using GIS. Unconstrained (DCA) and constrained (CCA) unimodal analyses have been used to interpret the assemblage of vascular plants. A total of 158 taxa were recorded. The result of DCA revealed a main gradient of distinctiveness between open-closed areas concerning both composition and diversity. In addition, CCA testing the whole environmental variables did not give statistically significant results. When tested separately, it emerged that plant composition in Mediterranean pine coastal forests is significantly affected by the distance from the sea ($p = 0.006$), which is one of the primary driver of species composition in agreement with other coastal Mediterranean studies (2). Ordinations failed in explaining pH and electrical conductivity. However, the seeming unimportant role of soil chemical parameters it was attributable to the rather homogeneous sandy soil matrix of this habitat. Although first results give interesting indications on the diversity and compositional arrangement of vascular plants in Mediterranean coastal stone pine forests and their hiding mechanisms, the study requires other efforts especially in expanding the study area and environmental variables considered, in order to provide some practical guidelines for protected areas that maintain these forests.

Fig. 1. Coastal forests of *Pinus pinea* L.

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5. = NATIONAL CATALOG FORM BNB: EXAMPLES OF APPLICATION IN THE HERBARIUM UNIVERSITATIS SENENSIS (SIENA)

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Herbaria kept in universities, museums, schools and private collections represent a huge source of historical data for basic biological investigations on plant diversity. Often, these data are dispersed and with heterogeneous (if not absent) levels of computerization.
The Herbarium Universitatis Senensis (1) since 2002 uses the platform http://www.anAnarchive.it to store, retrieve and analyze herbarium, floristic and vegetation data. It is an open source web geodatabase through a participatory approach and sharing of data by various parties (2, 3). AnArchive is built around an application written in java to enter and handle data and taxonomic list and use a web browser to search and retrieve data (4).
During 2013 the Herbarium Universitatis Senensis (University of Siena) was involved in the project approved by MIUR (Ministry for Instruction, University and Research) with other 11 universities, entitled "The computer technologies and the new realities for knowledge, networking and valorization of scientific cultural heritage: the role of the network of the university museums". The first goal of this poject was the inventory and the computerized cataloguing of specimen and objects of the museums with the national standards for cataloguing (Fig.1).
In this work we report two examples of application of the national standards for cataloguing for the Herbaria collections: BNB catalog form, as required by the recent legislation for Cultural Heritage (Statute Book for Cultural Heritage and Landscape, Legislative Decree 01/22/2004 and subsequent amendments) about the Catalog Information System (www.iccd.beniculturali.it) on the web SIGECWEB of the Central Institute for Catalogue itself (http://www.sigecweb.beniculturali.it/it.iccd.sigec.axweb.Main/).

Fig. 1. Seeds collection.

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5. = MORPHOLOGICAL CHARACTERIZATION, MOLECULAR ANALYSIS AND CHROMOSOME COUNTING FOR THE IDENTIFICATION OF *SERAPIAS* HYBRIDS (ORCHIDACEAE)

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The regional natural Park of Portofino hosts a conspicuous number of about 30 orchid species, among which *Serapias lingua* L., *S. neglecta* De Not., and *S. parviflora* Parl. are widely recognized within the area, while *S. vomeracea* Burm.f. and *S. cordigera* L. are only cited in literature but have not been recorded recently. Spontaneous hybrids, such as *Serapias* ×*meridionalis* E.G.Camus (*S. lingua* × *S. neglecta*), have been reported, but unclearly defined units were also observed ("*S. olbia* type"). Therefore, a morphological and molecular investigation, as well as chromosome counting, were carried out on different accessions, species and hybrids, currently present in the Park, aimed to characterize the existing entities and clarify their classification. Such morphological markers as hairiness of hypochile and epichile, type of callosity, length of labellum, width of epichile, width of hypochile and position of epichile were considered. Genomic DNA was extracted from leaves and molecular analyses were performed using Tubulin-Based Polymorphism (TBP) and Start Codon Targeted (SCoT) marker systems (1-2). Data were collected and analyzed by NTSYSpc 2.0 software with different coefficients, and estimated similarity values were used for cluster analysis.

Dendrograms revealed, for both morphological and molecular analyses, a strict correlation among *S.* ×*meridionalis* and the "*S. olbia* type" unit, with values greater than those observed for parent species. This suggests that the accession defined as "*S. olbia* type" could be referred to *S.* ×*meridionalis*, while morphological differences (plant height or hairiness degree) could be related to the role of parents in the cross (e.g. *S. lingua* as ovule donor and *S. neglecta* as pollen donor, or vice-versa). A comparison with typical *S. olbia* is needed to verify this hypothesis.

Chromosome counting was performed on root apices from *S.* ×*meridionalis* and its parental species. A combined protocol, specific for the Italian endemic *Serapias* species, was developed (3, 4), which allowed us to confirm the chromosome number of *S. neglecta* as having 2*n* = 36 chromosomes, and to characterize *S.* ×*meridionalis* as as 2*n* = 54. These findings have provided evidences for a correct identification of orchid hybrids, upholding the existence in the Park of spontaneous crossings between diploid (*S. neglecta*) and tetraploid (*S. lingua*, 2*n* =72) species.

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5. = EXPLAINING PLANT BIOTECHNOLOGY TO PEOPLE

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In the last two decades, numerous misconceptions were constantly made public on genetic engineering and Genetically Modified (GM) plants (1). This situation is due to a strong dissemination of twisted views and spread of inaccurate, biased and false scientific data through Social Media and Internet that creates a disinformation chaos all around these issues. The reasons include unawareness of the sources, but many times represent unclear and vested economic and political interests.
For researchers that participate in public sessions, hearings with decision makers/politicians or tv/radio debates, it's hard to reply to "simple" phrases that provoke fear on people, such as: "GM Crops causes health problems in humans and animals" or "GM crops provokes damages on biodiversity and environment". The audience's first reaction is to believe in it. It's neither easy nor fast to explain that these claims are not correct and why. The time available in those contexts is not enough to accurately explain why this kind of information is not correct.
The long term consequences of this disinformation chaos include a strong impact on European Union (EU) policies and regulations that causes a strong limitation in the adoption of Agrobiotechnology and the profit of its technical, environmental, economic and social benefits (2,3). This EU political context has a great negative influence in many developing countries (4,5) where the use of Agrobiotechnology would introduce benefits to their agriculture practices and to the quality of life of their populations. An important example is the "eternal" delay of golden rice delivery that could avoid about 250.000–500.000 cases of child blindness each year because of Vitamin A Deficiency and 1.5-2.3 million deaths/year (6).
CiB - Centro de Informação de Biotecnologia (Center for Biotechnology Information, Portugal) - www.cibpt.org - is a private nonprofit association created in 2002. Our team devotes special attention to Agrobiotech aiming to explain its utility and potential benefits in Portugal and to the Portuguese speaking communities. We make great efforts to explain and clarify these issues to different publics: farmers; technicians; government representatives; decision makers; researchers; journalists; high school students and teachers. We produce contents to promote the public understanding of Agrobiotech, communicating on different Internet platforms.
Over the past years CiB has participated in public debates and in political hearings; organized workshops for journalists on plant sciences and GM crops; organized workshops for researchers and farmers on agrobiotech communication; an annual meeting to promote discussion and interaction between audiences and stakeholders from different countries and regions of the world. The outcomes of these activities include the demystification of the unsustainability of this technology, the increasing knowledge on the conditions of adoption, the potential benefits and constrains of deciding to use of Agrobiotech and the support of the adoption of GM crops in Portugal, in Europe and worldwide.

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5. = MACROMORPHOLOGICAL TRAITS AND MOLECULAR ANALYSIS IN CENTRAL-ITALIAN POPULATIONS OF *QUERCUS* GR. *PUBESCENS*

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There are still controversial positions about the taxonomic arrangement of some oak species belonging to the *Quercus pubescens* complex e.g., with special reference for Central Italy, to *Q. pubescens* Willd. subsp. *pubescens*, *Q. dalechampii* Ten., recently lectotypified (1), and *Q. virgiliana* (Ten.) Ten. According to the most recent taxonomic revisions, the latter should be considered as a synonym of *Q. pubescens* subsp. *pubescens* and interpreted as an expression of a wide range of phenotypic variability (2). In order to point out if any among the several proposed macromorphological traits of leaves and acorns actually play a truly informative role in the diagnosis of these taxa, and to test the existence of genetic grounds for any of these traits, we took into account seven sites in central Italy representing a wide range of altitude and substrata. For each site we randomly sampled 10 individuals belonging to the *Q. pubescens* complex, picking and measuring leaves and fruits. Macromorphological measurements have been carried out on 3 leaves and 3 fruits for each individual. The measured traits of the leaves were: length of leaf peduncle, distance between the left and right leaf bases, total length of the leaf, distance of the apex of the longest lobe from the central vein, distance of the longest lobe from the base, distance of the sinus of the longest lobe from the central vein, distance of the sinus of the longest lobe from the base, width of the first (proximal) lobe, angle formed between the vein of the longest lobe and the central vein. The measured traits of the acorns were: cupule diameter and height, cupule margin's thickness and shape, nut length, width and shape, scales length, width and shape, presence and length of protruding scales along the cupule's border. For both sets of data, further indirect traits have been derived from the ratio between pairs of the measured ones. Samples of leaves and buds from the same individuals were collected and used for molecular analysis. Genomic DNA was extracted using DNAeasy Plant Mini Kit (Qiagen). Six nuclear microsatellite loci (QpZAG1/5, QpZAG9, QpZAG36, QpZAG104, MSQ13, QrZAG96) were amplified. Allele scoring was carried out by means of capillary electrophoresis using an ABI PRISM 310 Genetic Analyzer (Applied Biosystems). GeneMapper v4.0 was used for genotyping. The presence of genotyping errors and null alleles was checked through Micro-checker. Linkage Disequilibrium between each pair of loci was tested through GenePop. GenAlEx 6.4.1 was used to evaluate allelic richness and private alleles and test for the Hardy-Weinberg equilibrium for each locus as well as allelic frequency for each locus, expected and observed heterozygosity and the pairwise F_{ST} values (AMOVA method). A Bayesian approach was performed through Structure 2.3.3 with the aim to estimate the *a posteriori* probability of a given number of pool. A first overview of the macromorphological data set shows a large variability with reference to all the considered traits. It is well known that, in oaks, several morphological traits often used for diagnostic aims can show overlapping ranges of variation (3). However, the crossed analysis with the molecular data can highlight those patterns which show a clear correlation. Results will allow to verify the molecular basis of the well-known large morphological heterogeneity of this taxonomic group and will offer useful tools to disentangle a number of issues still open in several fields of knowledge, including vegetation analysis and phytosociological syntaxonomy.

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5. = DIGITAL KEY FOR THE IDENTIFICATION OF ITALIAN ORCHIDS

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Orchids are a very complex group, which has also an economic relevance. Accordingly to the checklist of Italian vascular flora (1, 2), 29 genera and 190 specific and subspecific taxa are known to occur in the country. Among these, some infrageneric taxa are subject of legal protection, because they are rare and/or listed as threatened in the IUCN Red List. Hence, an accurate knowledge of their occurrence in the country is of high relevance for conservation and management issues.
However, collecting observational records of orchids is difficult, because of the lack of a complete and up-to-date identification key. The better resource, till now, was the Flora of Italy (3), which is however outdated, because of the discovery of many new taxa, and the continuous taxonomical changes (some genera are under revision, and new taxa are delimited each year). Hence, a new, up-to-date identification key, which could be updated in real time by a continuous flow of information, is proposed.
In this study we developed a database of morpho-anatomical, ecological and distributional information in combination with the software FRIDA (FRiendly IDentificAtion). The resulting information system can produce digital identification keys accessible online, and/or in the form of apps for smartphones or tablets (both for iOS and Android OS). The keys can be produced by organizing the characters in any sequence decided by the author(s), targeting different users. They can follow a "classic" systematic scheme for University students and experts, or they can make use of more easy-to-observe characters for laypersons. Hence, these keys can be used also by ordinary citizens, such as in campaigns of citizen science. A taxonomic database, connected to the digital keys, will be developed as well, as an effort to put some order in the changing nomenclature of this family.

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5. = FIRST DATA ON THE POLLINATION ECOLOGY OF *CAMPANULA MEDIUM* L. (CAMPANULACEAE), A SPECIES OF ORNAMENTAL AND CONSERVATION INTEREST

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Campanula medium L. (Campanulaceae) is a biennial herb, endemic to Italy (NW Tuscany and Liguria) and SE France (Provence) (1). However, presently it is widespread as sub-spontaneous across Europe, probably due to its high ornamental interest as cut flower, with many existing cultivars. The plant is also protected in Tuscany, listed in the annex A of the regional law n° 56/2000. Given the high economical and local conservational interest of this species, we decided to investigate its pollination ecology, whose knowledge represents a crucial step to optimise cultivation and conservation plans.
It is well known that Campanulaceae show secondary pollen presentation mechanism, in order to increase the accuracy of pollen capture and transfer by pollinators, and *Campanula* in particular shows also proterandry (2). Despite this, the pollination ecology of *C. medium* has never been investigated before.
A natural population in the Apuan Alps (Tuscany, Massa Carrara) was investigated from June to September 2008. To determine the breeding system, four experimental treatments were applied on 48 randomly chosen flowers from 24 different individuals: (i) spontaneous self-pollination, to test autonomous self-pollination within a single flower, (ii) forced self-pollination and (iii) forced geitonogamy; (iv) forced xenogamy, to test for outcrossing ability. A control group (14 flowers freely exposed to environment) was selected. Each flower was marked and checked daily, to document flower senescence, fruit production and fruit seed set. According to (3), Index of Self-Incompatibility (ISI) and Index of Automatic Self-pollination (IAS) were calculated. In addition, the percentage of pollen limitation within the population was assessed as indicated in (4). We investigated secondary pollen presentation and the temporal dynamics of the sexual phases. Flowers in different flowering stages were randomly sampled in order to estimate pollen viability (by means of the tetrazolium assay, using MTT) and stigma receptivity (by means of Perex test) (5). Concerning stigma biology, its opening and receptivity were studied in relation to the presence of the pollen on the style. In addition, the pollinators and their behavior during the visits were recorded. The most representative ones were collected for identification. To test correlations and significant trends ($p \leq 0.01$), the Spearman Index and Kruskal-Wallis test were applied.
Our study showed that *C. medium* is partially self-compatible, partially autogamous and mostly outcrossing. The fruit set from experimental xenogamy was higher than that resulting from controls, suggesting a pollen limitation in the population studied. Proterandry for the species is confirmed: male and female phases were clearly separated and the presence of pollen on the style affects negatively the receptivity and the opening speed of the stigma. In addition, our study highlighted that the time of anthesis was longer in plants with a self-pollination treatment (14.13 ± 4.32 days) than plants subject to xenogamy (4.15 ± 1.52). This delay in flower senescence, possibly due to a lower production of ethylene (6), could be an interesting issue concerning the knowledge of floral life span, which is one of the most important parameters in species of horticultural interest, and especially in cut flowers. The most frequent visiting insects, which can be regarded as putative pollinators, were bees belonging to Apoidea, such as *Apis mellifera* L., *Bombus terrestris* (L.), *Xylocopa violacea* (L.) and *Anthidium* sp.; Curculionidae were also observed as pollen foragers.

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5. = DETERMINING THE COMPOSITION OF BIOFILMS LIVING ON STONE MONUMENTS

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Biodeterioration is the process of stone weathering due to the effect of living microbial populations (1). Cultural heritage, such as stone monuments and archeological sites, are exposed to the attack and proliferation of a large variety of microrganisms; autotrophs as cyanobacteria and algae and Heterotrophs such as bacteria and fungi coexist in complex biofilms, whose composition is influenced by the supporting substratum and micro-climatic parameters (2). Understanding biofilm composition is a necessary step in the conservation of cultural heritage.
The aim of this project is to set a "*diagnosis kit*" based on multiplex real-time PCR. This powerful assay allows to identify the presence/absence of different species in the sample through the use of fluorescent probes, and makes possible to quantify the signal in terms of biomass. Finding correlations between biofilm composition and deterioration status with this non-invasive assay will permit to prevent deterioration and plan the best restoring strategy.
An exhaustive list of microrganisms found on stone monuments, frescoes and historic buildings all over the Europe has been compiled (3,4,5,6). Barcoding markers for each class of chosen microrganisms were selected as follows: *16S* and *NifH* for Cyanophyceae; *18S* and *rbcL* for Chlorophyceae, *ITS1-ITS2* for Fungi. Nucleotide sequences of each chosen marker, when present in databases were downloaded and aligned with CLUSTAL W (7). Multiple alignments were adjusted by eye and trimmed with UGene software (8). PCR primers were built on most conserved regions and then tested by in silico PCR with Amplify4 software (8).
Primers were also checked with nBLAST in order to prevent unspecific annealings in the mixture.
Best in *silico* results of PCRs, in terms of sensitivity and non cross-amplification, were obtained with *16S*, *rbcL* and *ITS1-ITS2*.
In order to confirm these results, next step of this ongoing study will need the performing of control PCRs on both axenic cultures of microrganisms and artificial mixtures of microrganisms in known quantities.
The advantage of a multiplex real time PCR is not only the possibility of profiling microbial biofilms; a second step of this sensitive assay will focus on identifying sub-classes or even single deteriogen microrganisms with the only use of a specific different fluorescent probe. Conversely it will also be possible to detect bacteria that perform a bioconsolidant activity (9).
Our final goal is to develop a low cost "*diagnosis kit*" based on this assay, that could be used in routine analysis on biodeteriorated monuments.

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5. = TAXONOMIC REMARKS ON *ISATIS TINCTORIA* (*BRASSICACEAE*) FROM POLLINO NATIONAL PARK (BASILICATA, ITALY)

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Isatis L. (*Brassicaceae*) is an Eurasian genus including 79 species (1). It is one of the most difficult cruciferous genera (2). Some species, in fact, are highly polymorphic in fruit morphology, the structures that provide the most diagnostic characters (3). In addition, due to the extreme variability in all morphological characters, the limits of many species are uncertain (4). Most if not all diagnostic characters used in earlier classifications are very variable and because of the unreliability of vegetative and floral characters it is difficult or impossible to identify many specimens when mature fruits are missing (5). The patterns of variation suggest that hybridisation may be widespread (4). Moreover, intermediate specimens are rather frequent, even between some taxa that are morphologically easily recognisable (4). In Italy, according to Conti & al. (6), *Isatis* is represented by 3 species: *I. apennina* Grande (= *I. allioni* P.W.Ball), endemic to Italy and France (south-west Alps and central Apennine), *I. praecox* Kit., an European taxon restricted to Lombardia, and *I. tinctoria* L., an Asiatic species widespread in central and south Italy. Floristic investigations in the Pollino National Park (on the Lucanian side) led to the discovery of a little population referable to *I. tinctoria*. However, a comparative study of the plants showed that they differ in many relevant characters. The fact that this Lucanian population has morphological dissimilarities involves its critical revision that could bring to a new taxonomic delimitation.

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5. = A POWERFUL TOOL FOR A SAFE COLLECTION OF MUSHROOMS

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Wilde edible fungi represent one of the most economically exploited resources of forests. Mushrooming is an important part of local traditions and rural culture and it plays a relevant role also from a recreational point of view. For these reasons, a lot of mushroom hunters explore woods often without knowledge about the territory and mushroom toxicity. Consequently, every year, a significant number of people intoxicated after eating fungi or they are lost in forest. In turn, this involves a significant waste of money and man effort.
In this context modern Information and Telecommunication Technologies (ICTs) may offers many opportunities to face the previously mentioned problems. In fact, in the last decade, smartphones and tablets have become popular and widely used because of their mobility, friendly user-interfaces, communication interfaces, and their increasing computing power. On these bases, an APP for smartphones (see Fig. 1) has been implemented with the aim to provide a powerful tool for mushroom hunters exploring forests.
The App development involved a multidisciplinary approach, ranging from the construction of thematic cartographies (which take into account the geological, pedological, and botanical characterization of woods) and the mycological surveys (needed to check-list the principal species growing in the considered forest) to the implementation of the software itself.
The application for smartphone provides a series of functionalities, such as: the localization of mushroom hunters in a forest even if the area is not covered by mobile telephony; the list and iconography of the main fungal (edible and toxic) species growing in the area; the current mushrooming legislation; the potential distribution maps of edible fungi; the boundaries of territorial consortia (where it is compulsory to pay per mushrooming).

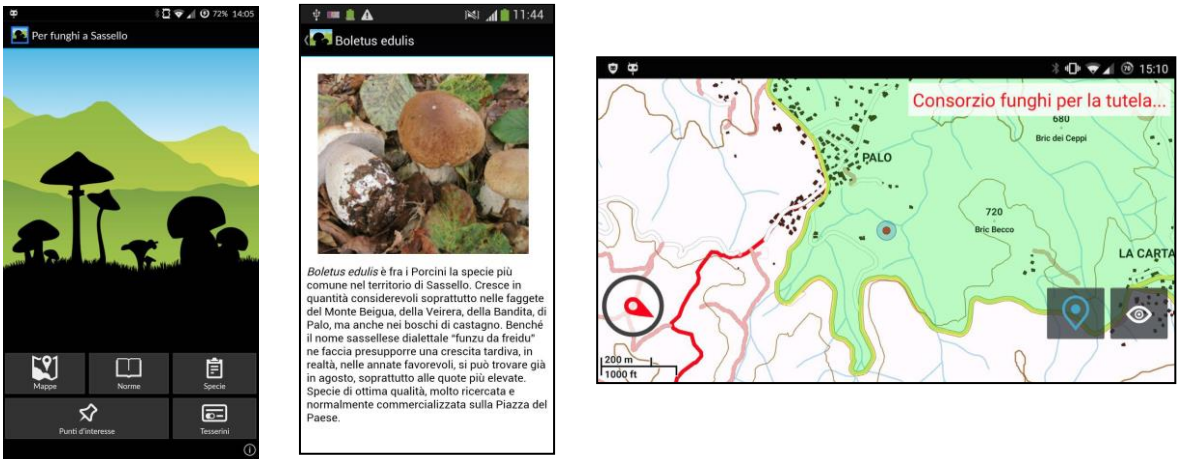

Fig. 1 – Three snapshots of the App "Per Funghi a Sassello"

5. = THE DISCOVERY OF PLANT BIODIVERSITY BY CHILDREN THROUGH THE ANIMATED MOVIES: ALICE IN WONDERLAND

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The discovery of biodiversity in childs occurs through various channels: either through direct contact with the outside world, and indirectly through paintings, games and the media.
Since 1937, Walt Disney Animation Studios realized animated movies inspired from novels. The first feature film was “Snow White and the Seven Dwarfs”. Since this, Disney’s studios realized more than 10 dozen movies.
The feature that distinguishes these works from a technical point of view is the attention to detail especially for the scene where the action takes place. Animals are often the protagonists of these stories, rarely plants.
A good example of the latter is in “Alice in wonderland”, a movie distributed in 1951 that tells the story of the discovery by Alice of an unmusical, fantasy-filled world beyond her imagination populated by odd human and plant and animal characters. The plot of this film is taken from “Alice's Adventures in Wonderland”, an 1865 novel written by Charles Lutwidge Dodgson under the pseudonym Lewis Carroll with several inclusions of the sequel “Through the Looking-Glass” another novel by the same Lewis Carroll, published in 1871. Both are novels full of symbolism, mathematical allusions and satire and contain the customs of England in the Victorian Age.
Alice, in the real and in in her imaginary world, moves within scenarios in which plants are the main background. But it happens in the scene in which Alice is in the flower garden that flowers come alive and become characters of the story. This scene is inspired by the second chapter of the “Through the Looking-Glass” titled “The garden of live flowers”. In the novel, Alice comes up a hill along a winding path, that houses a flower garden populated by daisies, a weeping willow, a lily, a rose, a larkspur and a violet. The real work of exaltation of plant biodiversity is made in the film by Walt Disney where in a scene of about 5 minutes more than 20 flowers are presented.
In the scene are clearly identifiable: *Bellis perennis*, *Cestrum elegans*, *Chrysanthemum indicum*, *Convallaria majalis*, *Cyclamen persicum*, *Delphinium* sp., *Hyancinthoides non-scripta*, *Ipomoea violacea*, *Iris germanica*, *Leucanthemum* ×*superbum*, *Leucanthemum vulgare*, *Lilium candidum*, *L. davidii*, *Narcissus incomparabilis*, *N. pseudonarcissus*, *Nelumbo nucifera*, *Ranunculus asiaticus*, *Rosa indica*, *Salix babylonica*, *Syringa vulgaris*, *Taraxacum* sect. *Taraxacum*, *Viola odorata*, *V.* ×*wittrockiana* and *Zinnia* sp. It is a real triumph of sounds and colours that leave the spectator delighted.
The theme of the plants between knowledge and representation has been the subject of international meetings and scientific contributions. Studies about the representation of plants in pre-Christian (1), during the Middle Ages and in European art of the XIV - XVII have been done (2). In our case the plants drawn in Alice in Wonderland seems to belong more to the plants cultivated in the Americans gardens of during 1950’s, when the film was made, rather than to the English gardens of the Victorian era where the novel is set. Nevertheless these drawings approach children, and the adults accompanying them in the vision, to the biodiversity of flowering plants cultivated. Offering to the spectator a wide representation of plants commonly grown with their shape and colours.
Certainly, "Alice in wonderland" is a good example of how even cartoons, such as fairy tales, can play an educational role, as they allow the child to learn while having fun. In particular, this cartoon can be a good teaching tool for knowledge of plant biodiversity. In fact, it can provide many ideas for developing a number of educational activities to be proposed in both the kindergarten and in the primary school, intended to make understand even the youngest children the meaning and the value of plant biodiversity, as well as to know some plants and their growth environments. Therefore, it would be desirable in the future to take into account such aspects, uncommon in animated films for children. In consideration of the now urgent need to fill several gaps of knowledge, widespread in the younger generation (3) in respect of the plants and the environment in general.

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5. = IMPLEMENTATION OF AN ONLINE INFORMATION SYSTEM FOR THE "FLORA CRITICA D'ITALIA"

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Since the early 2000s, within the Italian Botanical Society was discussed the opportunity to realize collaboratively a modern Flora articulated in monographs on the model of others under construction or recently completed in several European countries (1).
This initiative had a milestone in 2007 with the institution of the "Fondazione per la Flora Italiana", with the aim of supporting the activities of study, conservation and cultural and scientific enhancement of the flora of Italy and of sensitizing the national community about the many functions that it implements, in the context of natural and semi-natural ecosystems.
The "Foundation for the Italian flora" is responsible for the publication of the "Flora critica d'Italia". During the course of the project, it became apparent the diminishing impact of the hard copy in favour of an online information system more flexible and upgradeable and thus more suited to the current needs of the research but also in all fields of everyday life.
Last 25th February the first 3 contributions, concerning the families of *Isoetaceae*, *Heliotropiaceae* and *Hydrophyllaceae* (2, 3, 4) have been published online (http://www.floraditalia.it/index.php?page=flora_critica). These early contributions were prepared in the traditional way with a text editor.
In order to create the online information system, under the indications of the Editorial Committee, the Board of Directors decided to rely on the experience gained by the staff of Palermo in the field of biological databases (5, 6) and to support by itself the development of a specific software.
It is currently under construction and implementation a relational database that serves as a base for the entire project. It include information related to nomenclature, taxonomy, description, keys, distribution, images, karyology, uses, vernacular names, etc.
Data and tables are standardized according to TDWG (Taxonomic Database Working Group) standards (http://www.tdwg.org) and comply with the The Global Biodiversity Information Facility (GBIF) data architecture.
The system is composed by two main parts: a database and a web application. The database currently contains 16 relations (although more will be built within the end of the project) and relies onto MySQL Database Management System.
The web application is built using PHP scripting language and the CakePHP framework for the server-side part, running on a Linux server. The client-side part leverages onto Javascript and Jquery library. It follows the MVC[1] (Model View Controller) design paradigm. It deals with the database manipulation in terms of data management and data presentation along with user management. The whole system addresses several main security issues such as passwords hashing, accesses logging and code injection protections.
Data inputting is online by a web browser. The authors will input data in user friendly forms or to import tables.
Outputs produced include online ones coming from queries and pdf production to be done periodically to fix what done or to prepare chapters of the volumes to be eventually printed and included in a hard copy volume.

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5. = THE FOUNTAIN OF ST JOHN THE EVANGELIST'S ABBEY IN PARMA: DIAGNOSTIC ANALYSIS FOR RESTORATION PURPOSES

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The founding of the monastery located in the historic center of Parma, as it appears today, dates back to 1490 and includes three cloisters of which the one known as "the Portal's" or "St John's" was the last built between 1537-38, about thirty years later than the adjacent one, called "The Chapter's", with which it forms a compact and organic building suitable for use as a processional route. The Portal's cloister is the one you see as you enter the square, with columns of white marble from Verona with Ionic capitals, which for the first time are widely used in Parma. It is reported that between 1538 and 1542, the abbot Don Marco Cambi completed the works of the cloisters; in particular, the Portal's cloister was originally decorated by Leonardo da Monchio in the circles and triangles next to the arches. The only certain information regarding the fountain in the middle of the Portal's cloister is found in the deed dated October 17, 1589: Father Basilio devoted himself to several restorations of the monastery and with the deed of the notary Garbazza, Pietro Martino acquired from the community of Parma the water for the fountain of the New Cloister (or St. John's Cloister), and built the necessary pipelines (1). The fountain is composed of a square base with four steps on which is located the pedestal with a square base surmounted by an octagonal nut. Above the nut is the pillar that presents a Scotia molding in the lower part, while the real stem is shaped and is decorated with acanthus leaves. The tub above is shallow and shell-shaped. In the middle of the tank is a nozzle for water flow which has been modified in recent times; it is made of metal and plastic and looks inappropriate in the artistic context in which it is inserted. The fountain was built with the use of three different materials: some parts of the base with steps are in red granite of Baveno, other parts are made of limestone, while the tub and pillar are in marble. The different components of the base steps are linked by iron clamps with abundant material oxidation. Degradation, both of biological and non-biologic origin, was detected; for the macroscopic superficial alterations, diagnostic analysis and the biodeteriogen sampling methods, we made reference to the NORMAL 1/88, 3/80, 24/86 and UNI 10923: 2001 Recommendations (2). Visual examination showed that recently the artifact was improperly plastered on the pedestal and the exterior of the tub. Instead, a blackish crust of modest thickness, together with superficial deposits, covers the inside of the tub. In the base there are fissures and cracking, biologic and non-biologic layers, detachment of plaster, efflorescence and wax stains on the granite. The biodeteriogens macroscopically detected belong to algae, lichens, mosses and higher plants. Various species have been identified with the use of a stereo microscope and of an optical microscope. The patina of algae is widespread in the tub and in the base: it is made up of unicellular green algae including *Chlorella vulgaris* and *Ulothrix sp.*. The lichen colonization, instead, is limited and concentrated in the tub (*Caloplaca flavorubescens*). The plaster of the steps is populated by mosses (*Tortula muralis*). Higher plants appear sporadically in the cracks of the base (*Oxalis corniculata, Parietaria, Saxifraga tridactylites, Taraxacum officinale, Erigeron annuus*) (4). A future restoration should involve material consolidation and removal of biodeteriogens by a biocide treatment as provided by NORMAL 37/92 and 38/93 Recommendations (3).

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5. = *IN VITRO* GERMINATION OF TROPICAL EPIPHYTE ORCHIDS *CATTLEYA BOWRINGIANA* VEITCH AND *EPIDENDRUM NOCTURNUM* JACQ.

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Recently "orchid fever" has broken out again, although perhaps the passion for collecting these plants has never really been suspended from the XIX century until today. Many hybrids are commercially available, new every year, but the sale of botanical species is still strong among collectors. In this work we tested the *in vitro* germination ability of two famous species in orchid collections: *Cattleya bowringiana* Veitch and *Epidendrum nocturnum* Jacq. Flowers of the studied species were self-pollinated under glasshouse conditions in the Azienda Agricola Nardotto e Capello, Camporosso (Imperia). Immature pods were collected, sterilized with NaClO for 30 minutes and then flambéed for 10 seconds. Seeds were sowed on Petri dishes filled with two different media: 1) Malmgren growing medium modified by Pierce et al. (1) [Activated charcoal 2g/L; $Ca_3(PO_4)_2$ 75mg/L; KH_2PO_4 75mg/L; $MgSO_4$ $7H_2O$ 75mg/L; NH_4NO_3 100mg/L; Coconut water 50ml/L; Sucrose 20g/L; Agar 6g/L; pH 5.8] and 2) MS half strength (2) + Coconut water 50ml/L, Sucrose 20g/L and activated charcoal 2g/L; Agar 6g/L; pH 5.8. Petri dishes were then put in the growth chamber and cultivated at 21°C ± 2°C with 16h of photoperiod, with 40W Osram cool-white fluorescent tubes providing a quantum flux density of 30µE m^{-2} s^{-1}.
Seed germination started after 20 days from sowing for *Cattleya bowringiana* and after 45 days for *Epidendrum nocturnum* on both Malmgren modified growing medium (MALM-CO) and on 1/2 MS; the number of germinated seeds on the total number of sowed seeds was evaluated after 60 days (using immature pods some seeds were embryo-free and for germination percentages only seeds with embryo were considered). In *Cattleya bowringiana* germination percentages were 90.45% and 92.65% on MALM-CO and 1/2 MS, respectively. In *Epidendrum nocturnum* germination percentages were 83.85% and 88% on MALM-CO and 1/2 MS, respectively. For both species there were no statistically significant differences between the two media. However, differences were observed in the vigor of protocorms and seedlings. 60 days after sowing, *Cattleya bowringiana* on MALM-CO was at step 3 of seed development (appearing of protomeristem) according to Dutra et al. (3) and at step 6 (emergence of second leaf), in some cases also over, on 1/2 MS (Fig.1). The same results were observed in *Epidendrum nocturnum*, that reached step 2 (and in some cases step 3) on MALM-CO and step 4 on 1/2 MS (Fig.2). Also pigments were more colorful in protocorms and seedlings developed on 1/2 MS.
Germination percentages obtained for tropical epiphyte orchids, especially *Cattleya bowringian*a, are similar to those reported in literature (4) but, differently to previous works, the medium 1/2 MS showed an increase in the development of seedlings, suggesting the use of this new medium to propagate these orchid species.

Fig. 1. *Cattleya bowringiana* seed development after 60 days from sowing. On the left step 3 on MALM-CO; on the right step 6 on 1/2 MS.

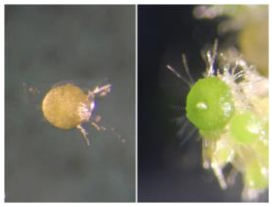

Fig. 2. *Epidendrum nocturnum* seed development after 60 days from sowing. On the left step 2 on MALM-CO; on the right step 4 on 1/2 MS.

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5. = GROWTH OF THE WOOD DECAY FUNGUS *PERENNIPORIA MERIDIONALIS* ON *MEDICAGO SATIVA*

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Perenniporia meridionalis Decock & Stalpers is a wood decay macrofungus which grows on hardwoods (seldom on conifers) in Central and Southern Europe, as far as Caucasus towards East. Since the species was quite recently taxonomically revised (1), only few isolates of this species are available and literature concerning biotechnological applications is almost absent. Mycological researchers of DSTA (University of Pavia) obtained its own isolate in 2010 from a sporoma collected in Italy, near Iseo Lake (BS), whose identity was checked by molecular analysis (2).
Enzymes of wood decay fungi can be exploited as a tool to achieve biodegradation of lignocellulosic agricultural residues aimed at sustainable production of biofuels and chemicals (3). *P. meridionalis* was therefore tested for the ability of growing on stems of *Medicago sativa* L., one of the most important plants cultured in Northern Italy. Fungal mycelium grown in liquid cultures was inoculated onto previously triturated and sterilized dried stems in Petri dishes; the chambers was moistened with sterile water; growth at 37°C was subsequently observed week by week for a month. This temperature was chosen after a preliminary growth profile test on 2% Malt Extract Agar (MEA). Incubation at 37°C resulted in a wide colonized area: even though quantitative determination was not possible, mycelium in fact appeared homogeneously woolly and substratum completely embedded.
Microscopy was performed both by: a) optical microscope 100x to 1000x (Zeiss Axioplan); b) SEM (Tescan FE-SEM, MIRA XMU series) which is equipped with EDAX spectrometer, at an accelerating voltage of 15-20 kVolt in high vacuum. Observations were only partially consistent with each other, since SEM allows a much finer discrimination between fungal structures and vegetal ones.
Stem appeared deeply destructured by chemical (enzymatic) degradation particularly affecting the not-lignified component. Typical structures of wood decay fungi such as abundant arboriform skeleto-binding hyphae spread as a capillary net were observed. The growth of hyphae with a wide diameter clearly engaged in stem penetration was observed; such hyphae showed sharp distal ends, while proximal ones looked bell-shaped. The surface of both hyphal types was often sprinkled with scarce to abundant encrustations likely to be oxalate crystals., fungi are well known to precipitate cations (and particularly Ca^{2+}) even in substrata poor in minerals, this is reported for several alleged functions among which pathogenesis, pH regulation, removal or reservoir of metals (4).
These preliminary results show that *P. meridionalis* displays remarkable versatility being able to efficiently colonize and degrade a substratum unusual for a lignicolous fungus.

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5. = ENVIRONMENTAL EDUCATION ACTIVITIES AND GUIDED TOURS AT THE BOTANICAL GARDEN OF CAGLIARI

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Environmental education is incorporeted in all the major international strategies for biodiversity conservation and sustainable development and botanic gardens have a key role to play in implementing these strategies. The International Agenda for Botanic Gardens in Conservation, published by BGCI (2000), stressed the need for all botanic gardens to develop themselves as centres for environmental education and sustainability. The Botanical Garden belongs to the University of Cagliari and was established in 1866 by Prof. Patrizio Gennari, it has a surface area of 5 hectares and is home to many species arranged in five core collections, mediterranean, tropical, cacti and succulent plants, palm grove, medicinal plants or "hortus medicus". The Botanical Garden of Cagliari is dedicated to the research and conservation of biodiversity, but is also actively involved in environmental education and in education for sustainable development. With a mission to inspire public understanding and appreciation of plants and the natural environment, the Botanical Gardens organize educational tours for children and adult groups, environmental education activities, exhibitions and social and cultural events. The aim is to increase environmental knowledge and awareness and to educate people about the urgent need to conserve plants. Starting with the 2014-15 school year the Botanical Garden of Cagliari has widened the didactic offer with environmental educational programmes, it has developed 5 environmental education activities for primary schools and 9 environmental education activities for secondary schools. Some topics are related to botany (plants morphology, plants as key regulators of ecosystem functions, properties of medicinal plants) while others are related to biodiversity and sustainability. Activities for secondary schools are: "Il gioco delle terre nascoste", an orienteering activity with the aim of exploring the biodiversity of the botanical garden, "I semi viaggiatori" the focus here is on the different types of seed dispersal and how dispersal is connected to form and function, "Alla scoperta del fiore" in this activity, students observe the similarities and differences between flowers of different plants and they also learn the anatomy of a flower. "La bottega dell'erborista", an activity where students learn about medicinal and edible plants. Environmental education activities for primary school schools are: "Quanti anelli hai?" an activity where students by reading a tree's rings discover the age and other amazing things about a tree, "Arcobaleno vegetale" an activity where students learn about plant pigments. "Dalla carta alla carta" the aim of this activity is to increase awareness of forest products and their uses and to discover how important it is to recycle and how easy it is to make recycled paper. These activities encourage learners to see, hear, feel, taste or smell the objects under discussion (learning through experience).In addition to these environmental education activities the Botanical Garden of Cagliari organizes guided tours for groups (school – infant primary and secondary, tourists, horticulturists, landscape architects, amateur and home gardeners) given by an expert guide. Data shows that every year about 25000 people, coming from different parts of the World, visit the Botanical Garden of Cagliari, especially during spring and summer. In 2013 the Botanical Garden of Cagliari received 3476 visits (guided tours), 53% of infant schools and 22% of primary schools. In 2014 the number of visitors decreased in 3352, 36% of lower secondary school and 47% of primary schools. From January to May 2015, 2789 visitors in guided tour enjoyed the Botanical Garden of Cagliari, 24% of lower secondary school and 61% of primary schools and 6% of school-age visitors chose to participate in environmental education activities rather than go on guided tours. In 2016 the focus will be on increasing the number of visitors and also to devising new environmental education activities for infant schools and for upper secondary schools and to provide new guided tours for the public in order to communicate the message that all life depends on plants.

5. = ANALYSIS OF WOODY FLORA DIVERSITY IN TWO SECTORS OF CENTRAL ITALY CHARACTERIZED BY DIFFERENT ENVIRONMENTAL FEATURES

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Climate, altitude, lithology and morphology are known to be the main drivers of floristic diversity at different scales; woody flora represents a good surrogate of total vascular flora diversity in heterogeneous environments at medium scales (1). We analyzed woody flora diversity in two sectors of central Italy (Colli Albani and Sabina, Lazio), to explore the relations between woody flora spatial pattern and environmental factors.

The study areas (Fig.1), even if are characterized by similar elevation range (from 250 to 950 m a.s.l.), belong to different phytoclimatic (mainly Transitional Temperate vs. mainly Temperate Region) and lithological (volcanic vs. sedimentary formations) units. Our dataset consists of 56 floristic relevées carried out in 2011-2014; 104 specific and subspecific taxa, and 66 genera were collected with 842 occurrence data. Nomenclature follows (2).

Using binary data, similarity between relevées was assessed by hierarchical classification (Ward's method) and by unconstrained ordination (NMDS in two dimension) on dissimilarity matrix (Jaccard index); differences between groups of relevées were tested by ANOSIM. Indicator Species Analysis identifies the associations between species patterns and groups. Moreover we analyzed species richness, chorological spectra and physical features of relevées (location, phytoclimatic, lithological and morphological features) to identify floristic and environmental patterns. Statistical analysis were performed by software R.

The dendrogram (Fig. 2) identifies two groups (1, 2) and four subgroups (1.1, 1.2, 2.1, 2.2), confirmed by ordination diagram (Fig. 3) and ANOSIM. Group 1 consists of 30 relevées mainly located in Sabina and falling into a Temperate bioclimate; sedimentary formations prevail; the taxa associated with this group are 11, such as *Asparagus acutifolius*, *Daphne laureola*, *Acer monspessulanum*, *Carpinus orientalis*, *Juniperus communis*, *Juniperus oxycedrus*, *Pistacia terebinthus*. The subgroups 1.1 and 1.2 differ in relative abundance of Eurasian (39% vs. 50%) and Mediterranean (35% vs. 23%) taxa, in species richness (greater vs. less figures) and in associated taxa. Group 2 consists of 25 relevées mainly carried out in Colli Albani, falling into a Transitional Temperate bioclimate and characterized by volcanic formations; the taxa associated with this group are 12, such as *Castanea sativa*, *Corylus avellana*, *Sambucus nigra*, *Cytisus scoparius*, *Euonymus europaeus*, *Acer pseudoplatanus*, *Rosa arvensis*, *Lonicera caprifolium*, *Tilia cordata*. The subgroups 2.1 and 2.2 differ in species richness (13-30 vs. 2-16 *taxa*), in relative abundance of Eurasian (50% vs. 63%) and Mediterranean (27% vs. 31%) taxa and in associated taxa.

A clear difference ìin woody flora composition between Colli Albani and Sabina has been detected, mainly associated with lithological and phytoclimatic features. These results represent the first step to confirm the efficiency of woody flora in analyzing total vascular flora diversity and spatial pattern at a fine scale.

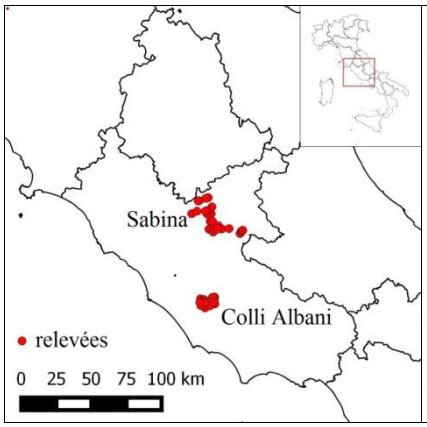

Fig. 1. Study areas.

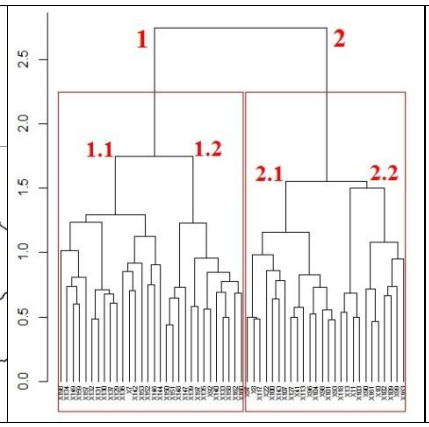

Fig. 2. Dendrogram of relevées.

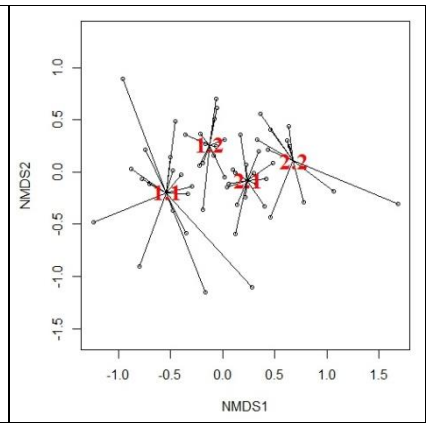

Fig. 3. NMDS plot (Stress = 0.2).

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5. = FLORISTIC AND VEGETATIONAL CHARACTERIZATION OF DEGRADED AREAS IN THE PROVINCE OF PISA: THE CASE OF A DISMISSED QUARRY AND THE NEIGHBORING TERRITORY (MUNICIPALITY OF VECCHIANO)

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The natural restoration of abandoned mining areas is one of the most complex environmental challenges due to the multiplicity of issues involved. In general, the spontaneous dynamics of renaturation in the quarry areas are very slow and altered because of limiting environmental conditions. So, recovery actions should promote ecological processes interrupted by mining and then the acceleration of the natural dynamics. Presently, in the province of Pisa several abandoned quarries have not undergone the necessary environmental recovery and, for this reason, they are classified as "areas of geophysical degradation". These include the former quarry C.E.I. dismissed in 1992 and located on the Monti d'Oltre Serchio in the municipality of Vecchiano (PI). The present work was focused on the flora and vegetation of this area. The study area, including the quarry (2.9 ha), the remaining sides of the Poggio dei Cavoli and the adjacent ridge of "Sassi Grossi", is characterized by limestone formations of the non metamorphic Tuscan Falda. It is characterized by a rather hard carsism, with almost no soil and high pH in the quarry area, while it is rich in oxides and lacking humus in the outer portions. Inside the cave, however, the scarcity of vegetation cover and the strong albedo, due to the light-colored rock, cause a different microclimate, characterized by dryness and higher temperature. The flora consists of more than 200 vascular species, of which about 50 are restricted to the quarry and 85 to adjacent areas. Particularly important is the number of floristic emergences, including rare orchids and ferns, which induced some scientists to propose the inclusion of the study area within the Regional Park of Migliarino-San Rossore-Massaciuccoli. This makes the site interesting from an environmental perspective and therefore, worthy of further valorisation and protection.
The analysis of aerial photographs and surveys conducted in the field has allowed the identification of the main types of vegetation (Fig. 1). In the territory outside the extraction site, the Mediterranean Macchia (high, medium and low) with evergreen sclerophyllous (*Quercion ilicis*) in catenal contact with the Garrigue (*Rosmarinetalia officinalis* and *Thero-Brachypodietalia*), and the mixed forest with dominant oak and transitional pruneto (referable to the alliance *Quercion pubescentis- petraeae* Br. Bl. 1931), can be found.
The quarry area is, instead, dominated by a pioneer vegetation that is hygrophile on the plain of the quarry, and lithophile on the front. This latter, at least in part, has species in common with the surrounding garrigues. This is interpreted as the initial stage of the series of vegetation that should lead to the reconstitution of the original forest cover, represented by the actual vegetation of the adjacent areas. The widespread presence of species such as *Rubus ulmifolius*, *Clematis vitalba* and *Dittrichia viscosa* (L.) Greuter, and of invasive exotic species, such as *Cortaderia selloana*, *Robinia pseudoacacia*, *Buddleja davidii* and *Ailanthus altissima*, reveal that, without proper action, it is difficult to imagine a fast recovery of the vegetation consistent with the area around the quarry.

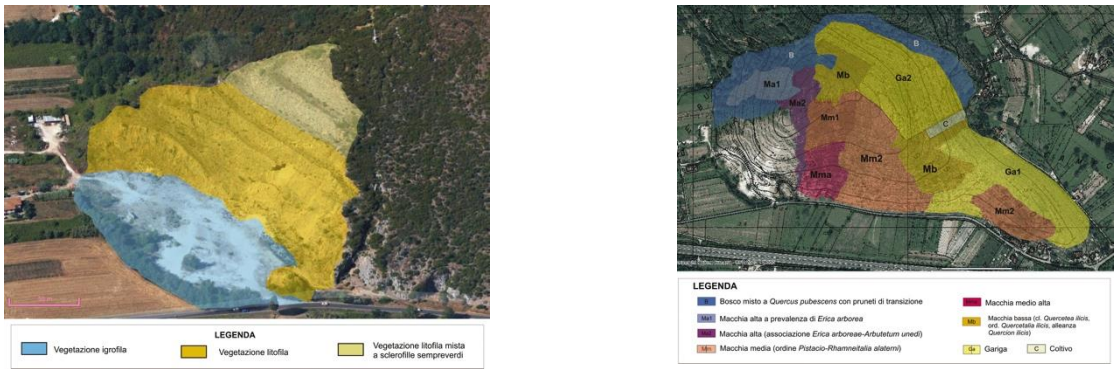

Fig. 1. Map of the real vegetation in the quarry (A) and surrounding areas (B)

5. = SEED DORMANCY AND GERMINATION IN *CAMPANULA MARTINII* F.FEN., A.PISTARINO, PERUZZI & CELLIN. AND *C. BERTOLAE* COLLA (CAMPANULACEAE)

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The Campanulaceae are highly understudied in terms of germination and embryo morphology. Members of this family possess typical small seeds (0.2-2mm) with dormant and not dormant embryos (1). Assignment of seeds to a dormancy class is a prerequisite to understand the ecological and evolutionary relationships of seed dormancy among different taxa (2). Since dormant embryos may or may not be underdeveloped, the purpose of our investigation was to determine if embryo growth occurs in seeds of two species within *Campanula* sect. *Heterophylla*: *C. martinii* and *C. bertolae*. The latter is endemic to the western Alps of Piedmont on altered serpentine rocks, while *C. martinii* has been only recently recognized as a new species, endemic to the Prealps of Lombardia and Trentino-Alto Adige (3). Germination tests were run on seeds of ten different populations. Germinating seeds were counted weekly and data expressed as final percentage germination and employed to calculate the germination mean time (GMT). About 500 seeds were employed to calculate the embryo to seed-ratio (E:S ratio) in dormant seeds, and after 24-h of imbibition, and 7-d and 140-d incubation at 4°C. The average length of dormant embryos was 291.55±14.68 m and 281.75±24.67 m, in *C. martinii* and *C. bertolae* respectively (ANOVA, p< 0001) and the E:S ratio was 0.294 and 0.335. After 7-d at 4°C, however, embryos considerably enlarged (360.15±22.64 m and 431.20±31.41 m in *C. martinii* and *C. bertolae* respectively, with significant differences between species, p<0.001). Final percentage germinations varied between the two species and among their populations, going from 49.54±2.25% (Val Degagna, BS) to 2.92±1.85% (Tremalo, TN) in *C. martinii* and from 97.96±1.18% (Caselette, TO) to 9.93±2.45% (Lanzo Torinese, TO) in *C. bertolae.* The two species differed significantly for their mean germinations starting from 75 days of incubation (p = 0.028). Populations differed statistically only in *C. bertolae*. GMT values were 77.35±5.54 days and 88.18±5.70 days in *C. bertolae* and *C. martinii*, respectively.
To germinate, embryo size increased from 99.84% to 128.17% in *C. bertolae* and from 144.71% to 215.20% in *C. martini* (E:S ratio = 1). In *C. martinii* the average increase of the embryo largely exceeded the range of 57–182% reported in literature for some Campanulaceae having underdeveloped embryos (2).
On the contrary, the E:S ratio of not germinated seeds was 0.555 and 0.505 in *C. bertolae* and *C. martinii*, respectively. These results demonstrated that: (1) embryos of *C. martinii* and *C. bertolae* are underdeveloped at seed dispersal, and need to reach a critical length before germinating; (2) seven days at 4°C are sufficient to start embryo growth; (3) lengths of dormant embryos differed significantly in the two species and showed a different rate of growth. Dormancy is an evolutionary strategy which is influenced by many environmental constraints but it is also under genetic control according to the species and their population diversity.

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5. = PRELIMINARY COMPARISON OF THE DEVELOPMENT OF SPONTANEOUS VEGETATION BETWEEN DEGRADED SOILS AND RECONSTITUTED ONES (PIACENZA, ITALY)

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Land degradation is a complex process of progressive loss of soil fertility due to soil exploitation, that causes alterations of its physical and chemical properties. Nowadays it is one of the most critical worldwide issues, that many policies and researches are trying to contrast through new technologies and new approaches. Reconstitution is an innovative technology (patented by the company mcm Ecosystemi) of mechanical and chemical treatments applied to degraded soils. This process affects the structure and the amount of organic matter, to create soils with better agronomical properties (1, 2). The efficiency of this technology is tested with the project called *New Life*, LIFE10 ENV/IT/000400, which is co-funded by the European Union. To compare the natural succession of spontaneous vegetation between degraded soils and their respective reconstituted ones, 28 plots have been created in 2013. These plots are divided in 3 groups according to the degraded soil origin. They are described below.

1) Group n. 1: total plots 10; 5 plots of degraded soils; 5 plots of reconstituted soils. The degraded soil origins from the landfill.
2) Group n. 2: total plots 12; 4 plots of degraded soils with four different textures (sand, silt and 2 kind of clay); 8 plots of reconstituted soils.
3) Group n. 3: total plots 6; 1 plot of degraded soil, 5 plots of reconstituted soils. The degraded soil is a cultivated one.

All plots are located in a rural area near Piacenza, in Gossolengo. In order to analyse the main chemical and physical parameters of soils, for each plot 3 samplings were carried out between September 2013 and June 2014. Analysis were carried out according to standard methods (3,4). To monitor vegetation, phytosociological relevés were carried out monthly from June to October 2014, for a total of 140 relevés. They are carried out according to the method of the Zurigo-Montpellier school.

Concerning flora, 71 species were found during the sampling, 67 of them belonging to vascular flora, 1 belonging to *Musci*, 2 are *myxomycetes* (*Lycogala terrestre*, *Stemonitis axifera*) and 1 belonging to *fungi* (*Volvariella hypophitis*). The most important family is that of *Poaceae* and the most common biological form is that of therophyte (e.g. *Chenopodium album*). Geophytes present high values of coverage and sometimes they are the dominant life form (e.g *Cynodon dactylon* and *Sorghum halepense*), even if their low number of species. According to Landolt's index, a lot of species are typical of bright habitats (e.g *Convolvulus arvensis*) and typical of nitrofile substrates (e.g. *Abutilon theophrasti* and *Amaranthus retroflexus*).

All these conditions are usual of a pioneer stadium (5) of a secondary succession that appears similar in all 3 groups. Soil seed bank and the invasion from neighboured vegetation source are two very important conditions that drive pioneer stadium of colonisations (5,6). Despite the short sampling period, the colonisation of plots appeared faster on degraded soils, perhaps due to a different condition of the seed bank that reconstitution treatments may have reduced on reconstituted soils.

Further analyses are needed to investigate seasonal fluctuations and succession development.

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5. = VISUALIZATION OF E2F-DEPENDENT TRANSCRIPTIONAL ACTIVATION IN *ARABIDOPSIS THALIANA* PLANTS

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The regulatory pathways that control cell cycle progression are conserved in animal and higher plant cells and include the the cyclin D/retinoblastoma/E2F pathway, a major regulator of cell proliferation that plays a key role in the G1-to S-phase transition during the cell cycle. Beside this role, the E2Fs have been also involved in the control of other cellular processes. The typical E2Fs can bind DNA as heterodimers together with DP (Dimerisation Partner) proteins and activate the expression of genes necessary for the synthesis of nucleotides and DNA. However, based on their interaction with retinoblastoma (RB) proteins, the E2F-DP complexes have also been shown to play repressive roles. Moreover, in both plants and animals, atypical E2Fs with repressive capacity have been discovered and appear to increase the complexity of this family of transcription factors.
Arabidopsis thaliana contains three typical E2Fs, named E2FA, E2FB, E2FC, that interact with the DP proteins DPA and DPB, and three atypical E2Fs called E2FD, E2FE, E2FF (1). It has been shown that both E2FA and E2FB factors are able to transactivate a synthetic E2F-responsive promoter, confirming their ability to act as positive transcriptional regulators, whereas E2FC has been shown to function as a transcriptional repressor, in association with the RBR1 protein. The atypical E2Fs have no transactivating ability and are believed to compete with the activating E2Fs for binding to the same *cis* elements (1). According to current models proposed for plant cells, the regulation of the E2F activity balances cell proliferation and differentiation. When released from RBR1 upon phosphorylation of the retinoblastoma protein by cyclin-dependent kinases, the E2FB factor appears to stimulate cell proliferation by activating genes involved in cell cycle progression. Otherwise, E2FA dissociated from RBR1 can determine the switch from cell proliferation to endoreduplication in cells committed to differentiation (2).
The purpose of our study is to visualize the E2F-dependent transcriptional activity in *Arabidopsis thaliana*. Plants have been transformed with a GUS reporter construct in which the expression of the GUS gene is regulated by a synthetic promoter, composed of 10 repetitions of the E2F binding sites (TTTCGCGC) placed upstream to a minimal –60 CaMV 35S promoter. Synthetic promoters have been previously devised and have several applications (*e.g.* the DR5 synthetic promoter is commonly used to study the auxin response in plants) (3). As for our work, this experimental design has been setup to visualize the patterns of expression conferred *in planta* by activating E2F factors that bind their specific *cis* elements. Surprisingly, histochemical analyses of GUS activity in the transformed *Arabidopsis* lines showed two distinct patterns of expression of the reporter gene and in 50% of the transgenic lines the GUS activity was localized in the root apex, which includes the highly proliferating cells of the root apical meristem. According to the models mentioned before, this result suggests a likely activation of the synthetic promoter by E2FB that determines this tissue-specific pattern of expression of the GUS reporter. Further analyses are currently underway to verify whether this meristematic activation is phase-specific during the cell cycle. Moreover, because 50% of the lines did not show GUS expression in root apical meristems, considering that the E2F sites could be a *hot spot* of methylation because they possess C and G nucleotides, studies are underway to verify whether the activation of the synthetic promoter is correlated to epigenetic mechanisms. Finally, another aspect under investigation concerns the use of this synthetic promoter to monitor the over-expression of an exogenous E2F factor, such as the DcE2F (4), in transgenic plants.

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5. = INTERSPECIFIC VARIATION IN TOTAL PHENOLIC CONTENT IN TEMPERATE BROWN ALGAE

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Phlorotannins are polyphenolic secondary metabolites found in almost all brown algae that function as defense against grazers, pathogens and epiphytes but are also involved in photoprotection mechanisms (1, 2). These compounds, produced in the Golgi apparatus, are accumulated in the cytoplasm, within vesicles called physodes, or bound to the cell wall (1, 3, 4). The concentration of phlorotannins differs within and between species, showing geographical as well as ontogenetic variations but may be also affected by environmental (e.g. temperature, light intensity) or biotic (e.g. grazing, epiphytes) factors (2, 5, 6, 7). The aim of this study was to focus on interspecific variation of total phenolic contents (TPC) in temperate brown algae also considering their relationship with environmental conditions. In particular, we compared the TPC of four species characterized by different thallus morphology, growth cycle and bathymetric level: *Cystoseira amentacea* (C. Agardh) Bory, *Cystoseira compressa* (Esper) Gerloff & Nizamuddin, *Dictyopteris polypodioides* (A.P. De Candolle) J.V. Lamouroux and *Padina pavonica* (Linnaeus) Thivy, collected in summer 2011 from the north-western coast of Sicily.
The TPC was determined colorimetrically with the Folin-Ciocalteu reagent. Results showed significant differences in TPC among the four species with *D. polypodioides* showing the highest value (0.74±0.001% of DW) and *C. compressa* the lowest (0.25±0.016% of DW). Significant differences in TPC were observed between the two leathery algae ($p<0.05$), with *C. amentacea* (0.48±0.056% of DW) showing higher TPC than *C. compressa*, and also between the two sheet-like algae ($p<0.001$), with *D. polypodioides* showing a significantly higher TPC than *P. pavonica* (0.30±0.024% of DW).
We hypothesize that the variations of TPC observed among the four species might result from a combination of internal and external factors such as thallus morphology (two species are leathery and two sheet-like), growth cycle (three species are semi-perennant and one annual), bathymetric level (two species inhabit the infralittoral fringe and two the upper infralittoral zone) and herbivore pressure.

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5. = THE BOTTARI-CHIEREGHIN (XVIII-XIX CENTURY) HERBARIUM OF THE NATURAL HISTORY MUSEUM OF VENICE (ITALY)

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In the second half of the eighteenth century the physician Bartolomeo Bottari (1732-1780) made a herbal consisting of plants collected in the territory around Chioggia and the Venetian coast, in support of an unpublished work entitled *Prospectus Florae Clodiensis et litorum Venetiarum*. At a later stage, the herbal passed to zoologist and botanist Stefano Chiereghin (1745-1820) who carried out the updating of the nomenclature of the species and added other specimens. In 1819 Stefano Chiereghin finally gave it to the Liceo Convitto of S. Caterina, now Convitto Nazionale "Marco Foscarini" of Venice. According to the documentation gathered, over time the herbal was consulted and rearranged by several scholars (1, 2). The herbal is in excellent condition for the most part. It includes native species such as *Pancratium maritimum* L., *Montia fontana* L., *Soldanella alpina* L., *Lysimachia vulgaris* L., but also many exotic *taxa* such as *Stapelia hirsuta* L. (native of South Africa), *Monarda dydima* L. (originating from North America) or *Pentapetes phoenicea* L. (from Southeast Asia and the northern Australian coast). Equally varied is the system used to prepare the individual sheets. Some plant specimens appear glued on the sheets; other secured by wide adhesive strips with the scientific binomial nomenclature marked on them; other specimens were sewn with white thread or inserted between two cuts made in the sheet; some specimens were placed loose between the sheets, while other were secured with staples, the latter procedure probably adopted in relatively recent times. Unfortunately the information on the tags is very scarce. The data that recur most often, in addition to the scientific binomial nomenclature, are references to the *Systema Naturae* of Carl Linnaeus or to the works of other naturalists such as Bauhin and Dodonaeus; in some cases information about the habitat was recorded, but it is very seldom as specific as in the writing <<in nemore dei Nardi>> found next to a *Potentilla*. Among the very few dates given are 1813, reported on the tag accompanying a specimen of *Plantago adriatica*, the date 1769 written on two small pieces of paper that accompany some lichens and, again next to the lichen *Collema pulposum*, the date of 1767: <<…Martio mense in ageribus sabulosis _ atrasim vulgo a la Morosini 1767>>. Three distinct handwritings are easily distinguished that may be associated with the different phases of the collection. The first handwriting, found on almost all of the specimens, recorded the binomial nomenclature (sometimes more than one) as well as the bibliographical references. The second is not generally associated with any information, and appears to have carried out a revision of the nomenclature (Fig. 1, from the archive of Natural History Museum of Venice). Finally, a third handwriting is associated with some specimens that may have been added to the collection at a later time (Fig. 2, from the archive of N. H. M. of Venice). Currently the material studied is preserved in the Natural History Museum of Venice. At present research activity is undertaking the review and the digitisation of individual *exsiccata* with a view to compiling a general catalogue of samples for better data analysis.

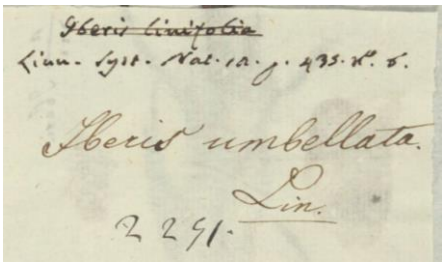

Fig. 1. Label of *Iberis umbellata* L.


Fig. 2. Label of *Nardus stricta* L.

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5. = FLOWERY VAULTS: STUDIES ON THE FLORA OF GENOESE FRESCOES OF THE 17th CENTURY

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The identification of plants in the Italian works of art, both in terms of botanical systematics and about their symbolic meaning, it is a topic of great interest for its multiple applications (1, 2). The recognition of different plant species within the works of art in Genoa has been addressed so far only for paintings on canvas and wood in Liguria in a time span ranging from the sixteenth to the eighteenth century (3, 4). Examining particularly the seventeenth century - the period that identifies perhaps the moment of greatest power and wealth of the Republic of Genoa and of its ruling aristocracy - it is easy to see how one of the most rich and varied artistic productions is that of fresco painting, which was used to adorn nearly every major noble residences and the main religious buildings. Actually, the great artists of fresco, at the head of the most prosperous and rich Genoese workshops dominate the Ligurian scene and catalyze the attention of the clients, creating great decorative cycles which have in the reproduction and the use of plants for decorative purpose one of their most interesting characteristics. The possibility of reconstructing, through the study of the repertoires available to artists, the scientific, cultural and 'workshop' panorama which may have contributed to a botanical production so scientifically correct, is set up as a line of research very interesting and innovative if designed in conjunction with the study of outcomes in Liguria. It is therefore interesting to be able to reconstruct an operational framework of the art workshop in this restricted geographic area, by searching the book sources, if available, also through a meticulous identification of plant species found in the fresco decorations of the period investigated; possibly, in such a way, the precise descriptive choices of a species rather than another could be understood, even as a specific iconographic and meaning intent, within the complex framework of the whole work. As part of a project on the 'culture of flowers in the Genoese Baroque' which has been recently undertaken between historians and botanists of Genoa University, we present some preliminary research concerning the vault decorated by Bartolomeo and Domenico Guidobono in the Palace Centurione-Cambiaso in Genoa, in the late seventeenth century and that one of the Aula Magna of the University of Genoa (former Hall of literary exercises of the Collegio of the Society of Jesus), painted by Gio. Andrea Carlone and Gio. Antonio Haffner in 1683. This study, currently in progress on other important decorative Genoese cycles of the same period, in addition to the identification of the species depicted, aims at a wider survey in order to define a scientific framework of botanical knowledge on the part of the artists themselves - whenever possible also through the repertoires hosted in libraries which they could consult - until to get to the possible identification of precise iconographic and semantic choices through which we could give a more complete and thorough interpretation of the whole work and of the role - perhaps too often considered secondary - of the botanical decoration.

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5. = SEED GERMINATION REQUIREMENTS AND EFFECTS OF SODIUM CHLORIDE AND POTASSIUM NITRATE IN THREE TYRRHENIAN COASTAL SPECIES OF THE *SILENE MOLLISSIMA* AGGREGATE (CARYOPHYLLACEAE)

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Silene mollissima (L.) Pers. aggregate is included in the *Siphonomorpha* Otth. section of the genus *Silene* L. and currently comprises 11 endemic species of the Western Mediterranean Basin. Five of these species have a distribution which is centered in the Tyrrhenian area. The three species investigated were *S. velutina* Pourr. ex Loisel., *S. ichnusae* Brullo, De Marco & De Marco and *S. badaroi* Bestr., all occurring in coastal habitats. Inter- and intra-specific variability in the responses to light, constant (5-25°C) and alternating temperatures (25/10°C), NaCl (0-600mM), KNO_3 (20 mM) under salinity and recovery of seed germination was evaluated for all the species. Seeds of the three taxa were non-dormant and light significantly improved their germination, which showed high percentages (> 80%) at the low temperatures (5-15°C) and under the alternating temperature regime (25/10°C) decreasing significantly at the highest temperature (25°C). *S. velutina* and *S. ichnusae* seeds germinated up to 300 mM NaCl, while *S. badaroi* until 100 mM. For all the species, salt did not affect seed viability and recovery did not decrease whit increasing salinity and temperature, except for *S. badaroi*. Inter-population variability both in salt tolerance and recovery was detected for *S. velutina*. The adding of KNO_3 did not affect germination and its recovery under salt conditions. Our results highlighted that these species have their optimum of germination during autumn-winter when, under the Mediterranean climate, water availability is highest and soil salinity levels are minimal, but they are also able to germinate until spring.

Fig. 1a-1b-1c. Habitus of *Silene ichnusae*; germination tests and *S. velutina* germinated seed.

5. = A METABOLOMIC APPROACH TO EVALUATE THE EFFECTS OF ENVIRONMENT ON THE GARGANEGA GRAPE METABOLOME DURING RIPENING

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Grape metabolome includes a wide set of secondary metabolites, in particular phenolic compounds, essential for defining some wine quality traits such as colour, taste and aroma. Since their biosynthesis and accumulation are affected both by genetic and environmental factors, there is a great interest in understanding how these compounds are accumulated within the berry during ripening and which are the molecular mechanisms at the base of the *terroir* effect. This work focuses on the white grape variety Garganega, a traditional cultivar of the Veneto region which gives rise to very fine wines such as Recioto di Soave, but whose metabolomic information is still scarce. The aim of this work is to unravel the berry metabolome of Garganega and its changing during ripening, evaluating the effect of the environment on the composition and the amount of the phenolics accumulated. Garganega berries were sampled at four different time points during ripening from four vineyards located in the Soave (VR) area and their methanolic extracts were analyzed by HPLC-ESI-MS and HPLC-DAD,allowing the identification and quantification of the grape phenolics. Chromatographic data subjected to multivariate statistical analysis showed that the qualitative composition in phenolic compounds was not affected by the different location of the vineyards, with the most abundant molecules belonging to the classes of hydroxycinnamic acids such as caftaric acid, flavan-3-ols such as catechin and its oligomers and flavonols, especially quercetin and kaempferol glycosides. Besides, the effect of environment was predominant in defining different levels of accumulation of these molecules, highlighting clear differences between the samples taken from plain and hill vineyards.

5. = A COMPARISON BETWEEN PEDUNCULATE OAK AND BLACK LOCUST VESSELS IN TREE RINGS: IT IS ALL A MATTER OF SIZE

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By means of tree-ring anatomy investigation, it is possible to assess wood cell anatomical features (i.e. vessel number, size, and distribution) along a series of dated tree-rings and to characterize the relationships between tree growth and environmental factors (1). Recent methodological improvements allow today to identify new parameters that contain valuable information on wood response to environment, this providing new knowledge on tree plasticity to climate. As regards European broadleaves, in the last years several studies have investigated anatomical features of different oak species. On the contrary, only few data are available on tree-ring anatomy of black locust, one of the most widespread non-native tree species in Italy.
Herein we present preliminary results of an investigation conducted in the "Siro Negri" Forest Reserve (Pavia, N-Italy), a natural broadleaves mixed forest, in which anatomical features of pedunculate oak (*Quercus robur* L.), the dominant tree species, are compared with those of black locust (*Robinia pseudoacacia* L.), established in the Reserve more than 70 years ago. Within the Reserve, we sampled 10 oak and 15 black locust trees by a 5-mm increment borer. After dendrochronological preparation, tree-ring measuring and cross-dating, a core from each tree was scanned at 2400 dpi resolution and 24-bit colour depth. Digital images were processed with ROXAS (2), an automated image analysis system, specifically designed for tree-ring anatomy, which allows the extraction of xylem vessels according to morphometric criteria. A large set of vessel variables was directly obtained by ROXAS or subsequently calculated. We finally retained 23 variables, related to total ring (i.e. ring width, total ring area, net wood area), to vessel number, size and distribution within the ring (i.e. mean and total vessel area, vessel size corresponding to 10°, 50°, 90° percentile of intra-ring distribution), and concerning particular groups of vessels (vessels of the first row or vessels larger than the mean). Some additional derived variables were calculated, i.e. vessel density, accumulated potential hydraulic conductivity, percentage of conductive area. For each variable, raw time series were transformed into residual series, removing autocorrelation. Descriptive statistics were then obtained for both raw and residual series in the common period 1954-2005. Correlation analyses were performed to compare variables within and between the species.
A total of 47289 vessels were measured for the two species (27853 in black locust, 19436 in oak). Number and size of measured vessels varied greatly. In general, the mean vessel area was much lower in black locust then in oak (27100 and 42600 μm^2 respectively). This difference was only partially balanced out by the higher number of vessels per ring in black locust (31 against 18), so that the accumulated potential hydraulic conductivity (proportional to squared vessel ray) was higher for oak. Both the species produced larger vessel at the beginning of the vegetative period (fig. 1), so that mean vessel area from the first row was higher than the mean area for all the vessels, but oak concentrated about 40% of all the vessels near the ring boundary, while black locust 34%. Correlation between inter-annual variations of anatomical parameters in the species was variable depending on the considered parameter: significant values were found comparing tree-ring width, tree-ring and net wood area, vessel density and percentage of transport surface within the ring, but the highest correlation was obtained when considering mean area of the vessels above the fiftieth percentile. All the other parameters had specific inter-annual variability, which probably reflected specific ecological requirements. Despite both the investigated species are considered ring-porous, differences in wood anatomy and intra-ring vessel distribution indicated diverse wood structure. Different intra-annual variations in the investigated parameters suggested that the species have distinct modes to adjust the xylem system, and thus the capacity to transport water, to environmental variations.

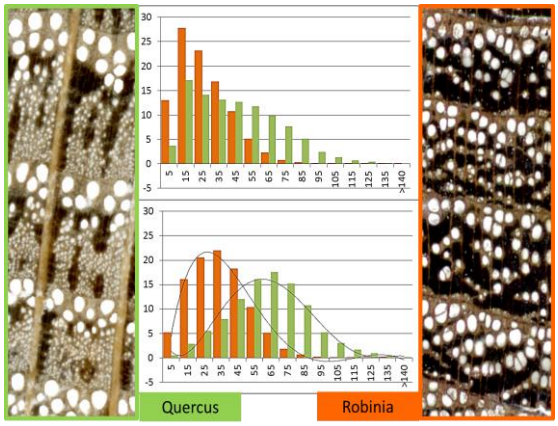

Fig. 1. Vessels in *Quercus* (right) and *Robinia* (left) and their size distribution within the ring (up) and in the first row (down).

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5. = WIKIPLANTBASE: A COLLABORATIVE PLATFORM FOR FLORISTIC DATA. FIRST STEPS TOWARDS ITALIAN REGIONAL FLORAS ONLINE

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Online databasing of plant diversity data became one of the major issues in biodiversity informatics in recent years. An increasing number of databases is now available concerning nomenclature and taxonomy, herbarium specimens, invasive alien plant monitoring, plant traits, ecology, vegetation, chromosome numbers, plant rDNA sites, genome size, DNA sequences. On the contrary, there are still few publicly available databases storing floristic data, especially at local level (1). Floristic records provide baseline data for researches in plant biology, linking a certain systematic unit to the localities where it is known to occur. Traditionally, printed floras are the "one-stop-shop" to find floristic data for a territory; unfortunately, printed floras are quickly outdated. Therefore, researchers must look for updated data in the scientific literature and herbarium specimens: the longer the time elapsed since the issue of the flora, the heavier the effort.
In consideration of this, in 2013 we have started the project "Wikiplantbase #Toscana" (2) to provide a framework where the full set of georeferenced floristic records of Tuscany can be entered, stored, updated and freely accessed through the Internet. In few months, thousands of data accumulated and, and in summer 2014 we started to modify the platform to store floristic records also for Sardinia. As a result, since 17 November 2014, Wikiplantbase #Sardinia is available online (3), and recently both platforms were upgraded to version 2.0, allowing a wider set or search possibilities.
As of 28 May 2015, Wikiplantbase #Toscana is storing 84149 floristic records, and Wikiplantbase #Sardegna 25000. Concerning the taxonomic coverage, for more than 90% of specific and subspecific taxa known for Tuscany there is at least one record available; while for Sardinia to a lesser extent, is still about 50%, but rapidly growing. The most recorded species are, so far: *Pteridium aquilinum* (L.) Kuhn subsp. *aquilinum* (Dennstaedtiaceae) for Tuscany (253 records from 161 different localities), and *Pistacia lentiscus* L. (Anacardiaceae) for Sardinia (292 records from 189 different localities).
The platform's design allows to store further regional records, so as to support a unified floristic database of the Italian Regions. With minor software tweaking, the online platform Wikiplantbase might be adopted in other contexts, resulting in a well connected network of regional floristic databases suited to exploit the involvement – still largely untapped – of non-academic collaborators, as advocated by citizen science.

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5. = INFLUENCE OF ABIOTIC AND BIOTIC FACTORS ON THE PRODUCTION OF TOXINS IN THE DINOFLAGELLATE *OSTREOPSIS* CF. *OVATA*

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Several dinoflagellate species are known to form dense blooms posing severe health issues and ecosystem threats through the production of toxins or other noxious substances. In the last decades, blooms of the benthic-epiphytic microalga *Ostreopsis* cf. *ovata* have been among the most intense along the entire Mediterranean coasts [1], leading to ecological and human health problems, that are associated with the production of palytoxin-like compounds (i.e. isobaric palytoxin and a wide range of ovatoxins), known as ones of the most potent marine toxins. Blooms have shown different features in terms of blooming time-periods, abiotic parameters, algal density and toxicity. Thus, batch culture studies were performed, using different strains isolated from Italian coastal areas, for assessing the role of abiotic (e.g. temperature, salinity, light) and biotic (e.g. presence of other competing algal species or bacteria) factors in the bloom development and toxicity.
Firstly, we highlighted that the production of the different ovatoxins by *O*. cf. *ovata* and their extracellular release increased during the growth [2]. The toxic profile revealed either a high qualitative (i.e. absence of some ovatoxins) and quantitative (i.e. toxin concentration per cell or culture volume) variability among the various strains. In general, temperature, salinity and nutrient amount resulted important factors in regulating cell toxin production [3,4,5]. More specifically, nutrients (N and P) were confirmed to be essential for the biosynthesis of ovatoxins, which are high molecular weight polyketides containing nitrogen, and results attested as mostly in N-deprivation but also in P-deprivation toxin amount was lower than in replete conditions.
Among the factors affecting *O*. cf. *ovata* growth and toxin production, the competition with other algal species (e.g. diatoms or benthic dinoflagellates) was relevant, while allelopathic effects of the ovatoxins towards the other microalgae may be excluded. The bacterial community associated with *O*. cf. *ovata* in culture influenced toxin production due to the re-mineralization of the organic matter, releasing amounts of N and P available, and suggesting a mutualistic algae-bacteria relationship [6].
Overall, these results attested that the physiological status of the cells is strictly linked to the physical-chemical conditions of the ecosystem, which, on the other hands, affect toxin production and release.

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5. = GROWTH INHIBITION OF *OSTREOPSIS* CF. *OVATA* CULTURE BY TOXIC ALGAL-DERIVED POLYUNSATURATED ALDEHYDES

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Ostreopsis cf. *ovata* is a toxic bloom-forming benthic dinoflagellate distributed both in tropical and temperate regions. During the last decade, massive *O.* cf. *ovata* blooms have occurred in different coastal areas of Mediterranean Sea including the northern Adriatic Sea where blooms have been observed since 2006 (1). These phenomena have been related to human intoxications and mass mortalities of benthic marine organisms due to the production of palytoxin-like compounds.
In the last decade, several microalgae have been shown to release a wide range of secondary metabolites. A variety of diatoms genera produce and release different types of long-chained polyunsaturated aldehydes (PUAs) (2). These compounds have been related to multiple functions such as chemical defense against grazers, intrapopulation signal molecules and have effects on a variety of marine organims as well, including co-occurring microalgae. Moreover, several recent studies have highlighted changes on growth, cell membrane permeability, flow cytometric properties and cell morphology due the exposition of microalgae to PUAs (3).
This study investigated the effects of three PUAs, namely the 2E,4E-decadienal, 2E,4E-octadienal and the 2E,4E heptadienal (which are among those produced by diatoms), on the growth and cell morphology of the toxic benthic dinoflagellate *Ostreopsis* cf. *ovata*. *O.* cf. *ovata* was cultured under controlled laboratory conditions at different PUAs concentrations (from 0.1 to 36 µmol l^{-1}) and using two different time expositions (24 and 72 h).
Results showed a marked decrease of *O*. cf. *ovata* growth when cultures were exposed to the various PUAs compared with the control. After 72 h of exposition, the growth inhibition respect to the control was of 74%, 48% and 44% for 2E,4E-decadienal, 2E,4E-octadienal and 2E,4E heptadienal respectively. Although a concentration-dependent decrease of the growth rate was observed for all three aldehyde compounds, the effect was stronger with longer-chained molecules than with shorter-chained ones.
Moreover, in corrispondence of the highest concentration of 2E,4E-decadienal (6 µmol l^{-1}), aberrant forms of *O.* cf. *ovata* cells were observed, showing a marked decrease of cell dimensions, a contraction of cytoplasm and the formation of abnormal vesicle-like structures. After 72 h exposition, the aberrant forms represented the 81% of the total.
Our results pointed out the negative effect of PUAs on the growth and morphology of *O.* cf *ovata* cells and confirm the possible allelpathic role of these molecules.

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5. = NEW FLORISTIC DATA OF MOUNT ETNA

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This work represents a contribution to the knowledge of the vascular flora of Mt. Etna. After the early floristic works of the Etnean territory (1, 2) many works provided new data about the Etnean flora. Nevertheless this flora is not enough well-known.
In this work the authors report data acquired through recent field surveys on Mt. Etna; during these surveys species of particular interest were found. They are newly recorded species and some rare or very rare species; for these species new sites for the Etnean territory are reported.
The newly recorded species are over 10: *Boerhaavia coccinea* Mill., *Salix alba* L. subsp. *vitellina* (L.) Arcang., *Lathraea squamaria* L., *Malva veneta* (Mill.) Soldano, Banfi & Galasso, *Viola aetnensis* (DC.) Strobl subsp. *messanensis* (W. Becker) Merxm. & Lippert var. *gracilis* (Sibth. & Sm.) Poli Marchese & Turrisi nom. prov., *Tragopogon minor* Mill. and others. *Tragopogon minor* Mill. is also a new finding for Sicily.
New sites of over 30 rare or very rare species of the Etnean flora are reported: *Aristolochia altissima* Desf., *Ampelodesmos mauritanicus* (Poir.) T.Durand & Schinz., *Oenanthe aquatica* (L.) Poir., *Cardamine glauca* Spreng., *Viburnum tinus* L., *Chamaenerion dodonaei* (Vill.) Schur ex Fuss var. *palustre* (Burn.) Poli Marchese & Turrisi nom. prov., *Trachelium coeruleum* L. etc.
The presence on the Etnean territory of *Salix pedicellata* Desf. and *Stipa austroitalica* Martinovky subsp. *appendiculata* (Celak) Moraldo is confirmed.
In addition some taxa (*Vicia cracca* L. var. *aetnensis* Fiori and others), which have been not included in the recent checklists of vascular flora of Sicily and of Italy (3, 4, 5), are reported.
Among the reported entities there are "guide species" of the Habitat 92/43 CEE Directive and species which are listed in Regional or National Red Lists.
The results obtained allow to update the floristic knowledge of Mt. Etna. This is of a particular significance due to the peculiarity of Mt. Etna, which is a natural park, recently included in the list of the World Heritage Sites of UNESCO.

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5. = SEASONAL VARIABILITY OF BRYOPHYTIC VS. VASCULAR SPECIES IN THE VEGETATION OF SUBMEDITERRANEAN TEMPORARY POOLS IN CENTRAL ITALY

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In the Mediterranean and Submediterranean regions the temporary ponds are very important habitats for the conservation of plants, including Bryophytes. Data on bryophytic flora of the temporary pools and waterlogged soils in Piana di Ferretto (Umbria, inland Central Italy), a biotope of great conservation interest and a Natura 2000 site (IT5210020), are here discussed and interpreted in the light of the vascular plant communities which house them. Bryological data from the area date back to the 80s (1, 2). The vascular component, formerly studied in the 80s, has been recently reorganized in a phytosociological framework including six amphibian vegetation types belonging to the alliances *Isoëtion duriei* Br.-Bl. 1936, *Cicendio filiformis-Solenopsion laurentiae* Brullo & Minissale 1998, *Eleocharition acicularis* Pietsch 1967, *Ranunculo ophioglossifolii-Oenanthion fistulosae* De Foucault 2012 (3). A remarkable richness of plant species and communities was detected in these systems, also emphasized by the presence of two Habitats from 92/43/EEC Directive's Annex I (3110, 3170*). However, these analyses didn't take into account the bryophytic component, suggesting the possible occurrence of a chronological shift between the bryophytic and the vascular flora. In order to test this hypothesis, we selected 50 permanent sampling areas (40x40 cm^2 plots) representative of the phytocoenotic variability of the site, taking care of the tiny ecological variations that characterize the amphibian systems, with special care for the water persistence and slope. Floristic and phytosociological relevés were carried out fortnightly during the vegetative season. The investigations allowed the detection of thirty-three *taxa* of mosses and liverworts (24 *Bryophyta*, 9 *Marchantiophyta*), identified and named following Cortini Pedrotti (4) and Aleffi *et al*. (5). Thirteen species are new records for Umbria: *Campylopus introflexus*, *C. pilifer*, *Dicranella cerviculata*, *D. staphylina*, *Ephemerum recurvifolium*, *Fossombronia wondraczekii*, *Plagiomnium ellipticum*, *Pohlia nutans*, *Riccia canalicul*ata, *Riccia subbifurca*, *Riccia crozalsii*, *Phaeoceros leavis*, *Riccardia camedrifolia*. According to Cortini Pedrotti *et al.*, Aleffi & Schumacker (6; 7) and the European Committee for Conservation of Bryophytes (ECCB), *R. canaliculata*, *R. crozalsii* are considered endangered (E) and *F. wondraczekii* and *R. beyrichiana* are considered rare (R); *D. cerviculata* is considered (E). The bryophytic species show a late-winter development and largely disappear or reduce their cover values before the vascular communities reach their maximum level of development (half/end of May, depending on the type). In many cases this prevented from the possibility to include bryophytes in the vegetation relevés, or to detect their role in the ecosystem in a quantitative way. With the present study, based on repeated diachronic samplings, it was possible to relate the bryophytic species to each vascular plant community. The transitional bioclimatic condition of the study area, between Mediterranean and Temperate, generates a remarkable shift between bryophyte and vascular species phenology, probably emphasizing a phenomenon that often affects the vegetation of the temporary ponds even in purely Mediterranean contexts.

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5. = GENETIC DIVERSITY AND CONNECTIVITY IN RELICT POPULATIONS OF *PLATANUS ORIENTALIS* AT THE EDGE OF ITS DISTRIBUTION

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Mediterranean ecosystem, due to its complex paleoclimatic history, hosts important residual of past vegetation that survives with a scattered natural range by occupying restricted ecological niches. Massive changes in vegetation have occurred recurrently during the succession of glacial and interglacial periods in Europe. Several members of the Tertiary tree flora disappeared from the European continent during Quaternary glaciations, with the exception of few relict species that survived in refuge areas in southern and south-eastern Mediterranean. For these relict species information about the distribution of genetic diversity and populations genetic structure is fundamental for determining in situ and ex situ conservation strategies. In particular, these relict species, often distributed in small and scattered populations are expected to display very low levels of diversity due to the effect of genetic drift and to the restricted inter-populations gene flow. For these species the genetic characterization of individuals, if coupled with information from their spatial distribution, age and pattern of relationship among individuals, can allow a fine-scale genetic spatial structure of the relict populations. This is of special interest because the spatial arrangement of related individuals within populations provides also insights into pollen and seed dispersal patterns. Finally, because pollination is often between near neighbours, an understanding of fine-scale genetic structure may allow predictions concerning the likelihood of selfing or biparental inbreeding. Among relict tree species, *Platanus orientalis* L. is an Eurasiatic species of warm riparian forests and southern Italy represents the westernmost limit of its distribution. Here we compared levels of genetic diversity and connectivity within and among populations of *P. orientalis* selected from the peripheral and central part of its distribution. For this aim we used microsatellites markers in order to: (1) measure genetic diversity at different levels (individual, population, species); (2) analyse gene flow within and among populations; (3) perform paternity and parentage analysis to evaluate the contribution of seed and pollen dispersion to population connectivity. The preliminary analysis was conducted considering five microsatellites loci for a total of 250 individuals from 13 populations distributed in South Italy, Greece, Turkey and Bulgaria selected as representative of peripheral and central distribution. The total number of detected alleles was 16. Populations from central distribution always show polymorphisms in all examined loci while most of loci were often found fixed in the peripheral Italian populations. These latter were also the ones with the lowest values of observed and expected heterozygosity (the lowest value, Ho = 0.037 and He = 0.065, found in the population from Campania). The genetic differentiation between populations, estimated as proportion of total diversity, was calculated through the coefficient of Fst. Fst values were higher among the Italian populations (average Fst = 0.137) than among populations from the central distribution (average Fst = 0.051) indicating a greater isolation and then a higher differentiation among the Italian populations. These results were also supported by AMOVA analysis that showed most of the observed genetic variation occurring within population (63%) and not among populations (37%). According to this preliminary analysis, the small population size and the low genetic connectivity detected among Italian populations support a scenario where genetic drift plays a major role in shaping the diversity of *P. orientalis* populations at the peripheral of its range. Ongoing analyses on seeds paternity will experimentally test the effective seed and pollen dispersion within and among isolated populations. The genetic bottleneck and the low connectivity among the peripheral populations could potentially determine extinction of these residual populations. Indeed, the maintenance of genetic diversity represents a crucial requirement for long-term survival of relict species because the absence of variation may strongly reduce the adaptability of the populations to changing environments.

5. = UPDATING THE WOODY FLORA OF TUSCANY: THE FIRST COMPREHENSIVE SURVEY, TWO CENTURIES AFTER SAVI'S WORK

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Woody species represent a significant component of the flora and vegetation of a territory. Their presence or absence directly affects the formation of whole communities and plays an important role in landscape sculpturing and in forest management. For this reason, the knowledge of woody species geographic distribution is of great theoretical and practical relevance.
Tuscany, with 3810 taxa of vascular flora, is among the Italian regions with the highest diversity (1), has a high number of woody native taxa (2), and is ranked 4th in forest cover (3). However, specific researches on its woody flora are lacking. Apart from the historical contribution of G. Savi (4), the only study on the whole regional territory is a list of shrubs (5).
In the context of the Global Tree Seed Bank project, promoted by Garfield Weston Foundation in collaboration with Royal Botanic Gardens, Kew (UK), we present the first comprehensive survey of Tuscan woody flora with sub-regional detail (presence/absence at province level) based on bibliographic, herbarium and field investigations, including the unpublished observations stored in the on-line database Wikiplantbase #Toscana (1). For the purpose of our study we define as woody flora those taxa whose life forms are reported as phanerophytes (P) and/or nanophanerophytes (NP) (6), including also plants occasionally growing as chamaephytes. Based on our checklist, a chorological spectrum was built at region and province level. In addition, a cluster analysis (UPGMA, Baroni Urbani & Buser's index) of provinces, based on floristic data of native taxa, was carried out.
We checked 404 specific and subspecific taxa: 268 native (66.3%) and 136 non-native (33.7%). Native taxa belong to 107 genera and 53 families. Rosaceae is the most represented family (56 taxa), and *Rosa* is the genus with the highest number of species (23). Stenochorous taxa are 8 (3%), two of them (*Rhamnus glaucophylla* Sommier; *Salix crataegifolia* Bertol.) endemic to Tuscany. The province with the highest number of native taxa is Grosseto (196), followed by Lucca (193), while the lowest number was documented in Pistoia province (128). Only 70 taxa are shared by all the provinces, whereas 26 occur just in a single province.
The regional chorological spectrum highlights a dominance of Stenomediterranean (22%), followed by Eurimediterranean (16%) and European (13%) taxa. However, considering Eurosibiric and Mediterranean species *sensu lato*, the former are slightly more abundant (46% vs. 40%). The same pattern can be found at province level, with the exception of Livorno and Grosseto, where Mediterranean taxa slightly outnumber Eurosibiric ones. In addition, the chorological spectra for each province highlight that Livorno and Massa-Carrara have the highest percentage of stenochorous taxa (3.1% and 2.9%, respectively).
As concerns the non-native species, the highest number (78) is found in Livorno province, whereas the lowest number occurs in Siena (16). At regional level, 12 exotic taxa (9.3%) are invasive, 58 are naturalized (45%), and 58 are casual. Their native range mostly concerns Asia (27%) and N America (24%).
Cluster analysis of provinces resulted in two main clusters: a first one uniting Livorno and Grosseto (encompassing the whole Tuscan Archipelago), a second joining all the remaining provinces.
As regards conservation status, six taxa are included in national Red Lists (7, 8) and 77 in the Re.Na.To inventory (9) and in the annexes of regional law 56/2000.

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5. = SUB- OR NEO-FUNCTIONALIZATION FOLLOWING GENE DUPLICATION: INFERENCE FROM GENE EVOLUTION ANALYSIS.

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The recent advances in DNA sequence technologies and the consequent growing availability of large genomic data isenabling the revaluation and improvement of plant phylogeny and taxonomy as inferred from large datasets. Moreover, the release of whole genome sequences for anincreasing number of taxa is providingnovel and comprehensive resources to studies on whole genome duplication (WGD) events occurred during the evolution of major Eukaryotic groups. In particular in plants, the relevance of these events in shaping genome architecture and new functionalization has been underlined.
Different mechanisms, not mutually exclusive, can be involved in the formation of copies of genomic contents, leading to duplication of single genes, chromosome segments (segmental duplication) or whole genomes. Subsequent genome evolution can follow different trajectories. In many cases, gene copies are lost and, in the case of WGD, in the long term a reduction of genomic redundancy verges to convert the polyploid organism to a diploid stage. Alternatively, processes like sub- and/or neo- functionalization can lead to the retention of gene copies.
In this work we investigated the possibility to use a phylogenetic gene tree to infer sub- or neo-functionalization events. The gene coding for B-glucuronidase (*GUS*) was chosen as it is widespread in plants and present in a limited number of copies in most of the investigated taxa.
Our results provide the evolutionary framework to trace the history of the duplication events that led to the formation of multiple copies of *GUS*. The retention in all the examined taxa of paralogous copies generated by a WGD event which preceded Angiosperm divergence suggested the occurrence of sub-/ neo-functionalization events; evidence of different expression of the two paralogous copies, probably associated to a different function, was provided by subsequent hystochemical analysis in *Arabidopsis*.

5. = AIS LIFE – AEROBIOLOGICAL INFORMATION SYSTEM AND ALLERGIC RESPIRATORY DISEASE MANAGEMENT - LIFE13ENV/IT/001107

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The most important biological component of ambient air is pollen, and its allergens are the main cause of airborne allergic respiratory diseases (1). In Europe, emissions of some air pollutants have decreased over past decades (2). Nevertheless, this does not always produce a corresponding drop in atmospheric concentrations, especially for particulate matter and ozone, which have significant impact on human health (2). Chemical air pollutants and anthropogenic aerosols can alter the impact of allergenic pollen, while pollen production rises in higher atmospheric CO_2 concentrations (3, 4). Changes in plant flowering season due to climate change will probably result in an increase in the duration and severity of the pollen season, alongside a higher frequency of episodes of urban air pollution (1). Therefore, exacerbations of allergic respiratory diseases will have a more pronounced effect in coming decades (5). In this context, AIS LIFE project (http://www.ais-life.eu) aims to develop an information base, to enable policy-makers dealing with environment and health issues to better manage pollen-related allergic respiratory diseases, improve the quality of life of patients suffering from them, reduce health system costs, and increase awareness among sufferers of pollen-related allergic respiratory diseases. **Expected results:** Establishment and consolidation of a multidisciplinary, transnational network of experts, with particular attention to pollen-related allergic respiratory health; implementation and dissemination of an Integrated Information System (IIS) and an enhanced Personalised Information Systems (PPI) in Italy, France and Austria; the widening of the Tuscan monitoring network for aerobiological components, by activating a sampling station in Pisa (Italy); educational campaigns (Italy, France and Austria) on the use of the Aerobiological Information System (AIS), promotion of improved lifestyles, and prevention of respiratory allergic diseases; raised awareness concerning the effects of interactions between pollens and chemicals on allergic symptoms across Europe, to guide environmental and health policy decisions. The aerobiological monitoring is in function since 1st June 2014 in Italy, France and Austria, with the continuous collecting of the data on the most important allergenic botanical families and fungal spores: Asteraceae, Betulaceae, Corylaceae, Cupressaceae - Taxaceae, Fagaceae, Oleaceae, Platanaceae, Poaceae, Urticaceae and *Alternaria*.

Acknowledgements: ARPAT and Consorzio LaMMA (Italy); ZAMG and MA22 (Austria); RNSA, Air Rhône-Alpes, Air Parif, Météo France and Infoclimat (France) are gratefully acknowledged for providing chemical, aerobiological, and climate modelling data.

Fig. 1. Pollen Trap – VPPS 2000

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5. = DIVERSITY OF *SMYRNIUM PERFOLIATUM* (*APIACEAE*) IN SICILY

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Smyrnium L. is an Eurasian genus of the family *Apiaceae*, including about 20 species. In the Italian flora, it is represented by 3 taxa also occurring in Sicily. These are: *Smyrnium olusatrum* L., *S. perfoliatum* L. and *S. rotundifolium* Mill. (1, 2). The last one has also been treated at the rank of subspecies under *S. perfoliatum* [*S. perfoliatum* subsp. *rotundifolium* (Mill.) Hartvig] (3, 4) or as a mere variety (5). *Smyrnium olusatrum*, often associated to nitrophylous habitats, unlike the other two, occurs throughout most of the Island. *Smyrnium perfoliatum* occurs in the underwood of deciduous *Quercus* sp. pl. or of *Fagus sylvatica* L. woods, in the submontane and montane belts of the Nebrodi mountains. *Smyrnium rotundifolium* is instead common in some less mesophylous environments like thermophylous open woodlands of central and western Sicily. In this sector of the island, the populations of the Madonie and of the Montains around Palermo referable to *S. perfoliatum* diverge for both morphological and ecological characteristics. Indeed, plants corresponding to *S. perfoliatum* subsp. *perfoliatum* occur only in the underwood of *Quercus* and *Fagus* woods in the Nebrodi mountains, under mesophylous conditions; dissimilar forms that are rather intermediate with respect to *S. rotundifolium* instead occur in C-W Sicily (Madonie and Mounts of Palermo). The study of the phenotypic characters, in particular of the leaf, makes possible to clearly distinguish these populations that, therefore, represent a taxonomically and perhaps even chorologically critical case, since similar plants of Greece have been described as *S. rotundifolium* var. *ovatifolium* Halàksy (6). Then, the latter taxon, today overridden, in the light of the Sicilian case should be re-evaluated.

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5. = A MAP OF ETHNOBOTANICAL KNOWLEDGE IN TUSCANY

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Ethnobotanical studies in Tuscany have documented a rich set of traditional uses of wild and domesticated plants: while most of them are used as folk medicine or food, others play a central role in housekeeping, handicraft, cosmetics, homemade dyes and liquors, religious rites, magic, and superstition (1). Several papers and books have been published on these topics, mostly about restricted territories like municipal or provincial districts (see e.g. 2), physiographic units (see e.g. 3), or specific uses of plants (see e.g. 4). The Tuscan Ethnobotanical Centre C.E.T. (Centro Etnobotanico Toscano), established in 1999 and part of the Department of Agricolture, Food and Environment, University of Pisa, is committed to becoming a regional and national point of reference for the dissemination of researches about folk uses of plants. In this context, its staff, in collaboration with the BioLabs of Scuola Superiore Sant'Anna, after gathering numerous ethnobotanical papers and books about the Tuscan territory, have established a bibliographical archive. A similar work has been accomplished by the Botany Laboratory of the Biology Department of Pisa University, with specific attention to veterinary uses (5).
However, no unified inventory for the whole region has been published, listing all species of ethnobotanical interest along with their diverse uses in distinct territories and relevant bibliographic references, and allowing an assessment of the distribution of ethnobotanical knowledge in the regional territory. As first steps towards this list, we have **a**) integrated the above mentioned bibliographical archives; **b**) screened the references; **c**) assembled a summary table of published works, reporting the number of ethnobotanical records and their breakdown in use categories grouped by province. Finally, we produced a colour-coded map of Tuscany showing the number of published works and the number of matching ethnobotanical records for each province.
Bibliographical references document 6432 ethnobotanical records of folk uses in Tuscany, involving over 700 species. Medicinal use account for almost 46% of the records, food use for almost 30%. Veterinary uses rank third (5% of records) and concern mainly prevention and care of diseases, mostly for bovines. The province with the highest number of published works and the highest number of ethnobotanical records is Lucca (30 published works and 1236 records); at the opposite end stands Prato (1 published work, 58 records). The coastal provinces of Pisa, Livorno, and Grosseto are better documented than the inner provinces of Florence, Siena, Pistoia, and Arezzo.
We conclude that the distribution of ethnobotanical knowledge in Tuscany is heterogeneous and shows remarkable gaps especially in the inner provinces, linked to the Apennine territories which are likely to be resilient areas in an ethnobotanical perspective.
This work is preliminary to the production of a map of ethnobotanical records, linking plant names and ethnobotanical uses to the smalles geographic unit supported by relevant bibliographic references. The map will be publicly accessible online and will provide a query interface, modelled after Wikiplantbase #Toscana (www.biologia.unipi.it/ortobotanico/FloraToscana/flotos_start.html) (6) and Wikiplantbase #Sardegna (www.biologia.unipi.it/ortobotanico/FloraSardegna/flosar_start.html) (7).

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5. = CITIZEN SCIENCE AT ROME: AN EXAMPLE IN PHENOLOGIAL ACTIVITY

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Aerobiological monitoring is very important in helping people allergic towards pollinosis.
By following weekly bulletin published on several websites it is possible to keep informed about concentration levels and forecasts.
Our experience started in 1996; in 2000 we began to publish a weekly bulletin at web address www.polline.uniroma2.it. In these years we decided to support aerobiological monitoring with phenological observations, contributing to a weekly release of phenological maps by IPHEN.
Rome University of Tor Vergata's Monitoring centre participates in Italian PHenological Network (IPHEN) collecting weekly observations on *Cupressus sempervirens* L. and *Callitropsis arizonica* (Greene) D.P. Little.
Accordingly we carry out observation every 7 days according to the method proposed by IPHEN, taking notes about the different vegetative and reproductive phenophases.
Phenological observation help us to evaluate accurately pollen presence in the air; in fact winter precipitations can delay pollen collection by the sampler determining the air runoff.
To improve the observation site last autumn we thought of involving ordinary citizen among our website visitors.
Citizen scientists is a recent way to collect data used in worldwide, especially in north Europe after project Open Air Laboratories (OPAL) (1). Also European Environment Agency underlines the importance and the utility to involve citizen in projects on biodiversity monitoring (2).
Also New York Botanical Garden, for example, has different projects to study the impacts of climate change on the Native Forest, by engaging volunteers in collecting important scientific data on different species of trees, shrubs, and herbs.
We published on our website only an invitation to participate. We also sent to respondents material to learn to identify the *Cupressus* species, and how to collect and input data on the seasonal biological processes of those species eg. when leaves, flowers, and fruits.
Some citizen began to send their observations every week, from the end of October to the end of February.
Data were collected in more than 60 different sites in and out of great ring road. By using Q-GIS, we have obtained a first map of different phenophases for *Cupressus* in Rome, in which we can see how the blooming of *Cupressus* starts before in south west of Rome and then towards the eastern and northern sector.

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5. = PATTERNS OF SEED GERMINATION WITHIN ALPINE GRASSLANDS OF DIFFERENT PROVENANCE AND HABITATS

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Seed germination is an important stage in plant life, which strongly influences seedling survival, lifetime fitness and the history of the species. In the alpine habitats characterized by a short growing season and extreme temperatures (1) germination timing have to be extremely controlled. Indeed, different germination phenologies and requirements have been identified on alpine flora (e.g. 2). However, a typical alpine germination behaviour has not been found yet, likely due to the large diversity of alpine plants and habitats. Hence, a community wide-scale study on seed germination is essential to better understand how germination and dormancy responses vary across alpine habitats.
The aim of this study is to identify patterns of germination and dormancy through alpine grasslands of different habitat (classification based on Natura 2000 Network) and provenance. In particular, we focused on habitats 6230 (siliceous grasslands with acidic soil pH), target plant community species-rich *Nardus stricta* grassland from Pyrenees (Spain) and Alps (Italy) and habitat 6170 (calcareous grasslands with basic soil pH), target plant communities *Caricetum firmae* and *Seslerio-sempreviretum* from Picos de Europa (Spain) and Alps (Italy). So far, seed germination of 30 species, collected from habitat 6230 (Val Viola, Sondrio), have been tested in the laboratory under alternating temperatures (15/5ºC and 25/15ºC, 12-h daily photoperiod), after the exposure to 0, 3 and 5 months of cold-wet stratification and 3 months of dry storage (after ripening). Further germination test investigated the effects of gibberellic acid (GA_3, 250 mg/l). The response of final germination (FG) to the different temperatures and treatments was analysed using a non parametric test (Kruskal-Wallis test).
Seed germination was significantly different across temperature, species and pretreatments. Cold stratification significantly increased the germination in 16 out of 29 species and decreased it in one species (*Poa alpina*). After ripening increased the germination in two species (*Semperivuvm montanum* and *Campanula barbata*) and GA_3 in six species (*Campanula barbata, Gentiana acaulis, Nardus stricta, Oreochloa disticha, Pedicularis tuberosa* and *Pulsatilla alpina*). *Pedicularis tuberosa* and *Gentiana acaulis* germinated only using GA_3. In three species *(Carex sempervirens, Carex curvula* and *Luzula spicata*) seed germination did not occur at any condition tested. Finally, in some species (12 out of 29) seed germination occurred during the period of cold stratification. In conclusion, our results show that only one species (*Arnica montana*) had non dormant seeds. The predominant dormancy type is physiological dormancy, being found in 13 species. Seeds of one species (*Lotus corniculatus*) have impermeable seed coat and was assigned to as physically dormant. Finally, seven species (*Campanula barbata, Gentiana acaulis, Gentiana nivalis, Pedicularis tuberosa, Pulsatilla alpina, Pulsatilla vernalis* and *Sempervivum montanum*) have seeds with underdeveloped embryos and were sensible to cold stratification and/or GA_3, so they were assigned to the morphophysiological dormancy. We show that low temperatures regime (e.g. 15/5ºC) are more effective to detect dormancy on alpine species, since they allow highlighting their conditional dormancy state (e.g. in *Achillea moschata, Antennaria dioica, Festuca halleri, Gentiana nivalis, Potentilla aurea, Pulsatilla vernalis, Scorzoneroides helvetica* and *Solidago virgaurea*). Further experiments are on the way to investigate the extent to which similar germination behaviours may occur also in other alpine grasslands from different habitat and/or provenience.

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5. = MORPHOLOGICAL AND ANATOMICAL DIFFERENCES IN TWO FERN SPECIES GROWING UNDER COMMON LIGHT ENVIRONMENTS

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The structure of leaves has important implications for the performance of plants in specific habitats (1, 2, 3). Adjustments of morphological, anatomical, biochemical and physiological leaf traits are integral features in the process of acclimating to contrasting light environments (4).
Leaf morphological and anatomical traits of two ferns, *Polystichum setiferum* (Forssk.) Moore ex Woyn and *Phyllitis scolopendrium* (L.) Newman which can grow in different environmental conditions, were analyzed. In particular, *P. setiferum*, occurs in the South, Western and Central Europe growing along rivers of the temperate region in moderately sunny habitats (5). *P. scolopendrium* occurs in South, Western and Central Europe, extending to Sud Est of Sweden and North of Ukraine, preferring shade environments (6). Leaf morphology and anatomy of the two species growing outdoor in the Botanical Garden of the Sapienza University of Rome were analyzed. Fully expanded leaves were collected in February 2015 in the same garden plot under the canopy of deciduous trees. Leaf anatomical traits were measured on cross-sections by light microscopy using an image analysis system (Axiovision AC software). Total leaf thickness (LT), abaxial and adaxial cuticle thickness, abaxial and adaxial epidermis thickness, palisade and spongy parenchyma thickness, the fraction of the mesophyll volume occupied by intercellular air spaces (F_{ias}), and stomatal size and density (by nail varnish impressions) were measured. Stomatal area index (SAI) was calculated by the product of the mean stomatal length and stomatal density. Leaf area (LA, by an Image Analysis System) and dry mass (DM, after drying at 80 °C to constant mass) were measured. The leaf mass per unit of leaf area (LMA) was calculated by the ratio between LA and DM, and leaf tissue density (LTD) by the ratio between LMA and leaf thickness.
The results, on the whole, highlight a significantly higher LMA and LTD in *P. setiferum* (by 20% and 147%, respectively) than in *P. scolopendrium*, driving to a 28% reduced volume per leaf area of air spaces which determines a higher drought resistance. This last is further attested by a 22% higher proportion of adaxial epidermides and cuticle thickness on the total leaf thickness (336.1±32.3 µm and 162.7±16.1 µm in *P. scolopendrium* and *P. setiferum*, respectively) and by the 107% higher SAI which determines a more efficient water use justifying the capability of *P. setiferum* to grow in moderately sunny environmental conditions. The 54% lower ratio of palisade to spongy parenchyma thickness in *P. scolopendrium* could improves light absorption in shade environments by scattering irradiance into the mesophyll (7). On the whole, the results highlight significant differences at morphological and anatomical level in the two considered ferns reflecting their adaptive capability to contrasting light environments.

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5. = HUMAN MYCODIVERSITY IN VARIOUS STAGES FROM DEATH

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Forensic mycology is a relatively new term which describes the role of fungi in criminal investigations (1). In fact these can be used as an investigation tool, for instance in providing trace elements, estimating time of death (*postmortem* interval), ascertaining time of deposition, investigating cause of death (either hallucinations or poisonings) and locating buried corpses(2). After death corpses are a plentiful source of organic matter, and they are not only colonized by the fungi which represent the normal body mycodiversity (i.e. *Candida* spp., *Malassezia* spp. etc.) but it is also possible to detect fungi which are not normally able to colonize living tissues(3).However there is a lack of information about the role that fungi play in human decomposition. Nevertheless, some studies showed that both the environmental humidity and temperature of the corpses position affect the fungal growth (1, 3).
Our work involves a multidisciplinary approach, which is performed both by mycologists and forensic pathologist. Our study focused on a fungal community developed, over a period of two months, on a cadaver. This was found into a flat, and the cause of death that was seen during autopsy was likely due to sudden cardiac death. Upon discovering, the corpse appeared with already moldy face and ears. Mycological investigations were carried out previously by direct examination, later by isolation in axenic culture. The fungal strains were identified by macro and micro- morphological, biochemical and molecular analysis as well.
Fungal colonies on the corpse were sampled every week for two months in order to monitor the fungal growth. During this period, the corpse was kept at 4°C in a refrigerator into the Genoa mortuary. This study allowed us to highlight the presence of 48 vital fungal strains belonging mostly to the genera *Penicillium* and *Mucor*. The fungi grew and recurred on several parts of the body so we are able to draw the "total body temporal colonization map" Actually fungal growth first appears to interest the face, ears, neck and feet, whereas only later involves the arms, abdomen, groin, legs, and autopsy suture.
This work represents the first step to draft an experimental protocol, which might support the estimation of *postmortem* interval. Furthermore fungal strains identification may contribute to assess a range of moulds involved in and causing the decomposition process.

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