



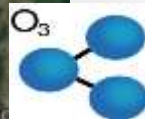
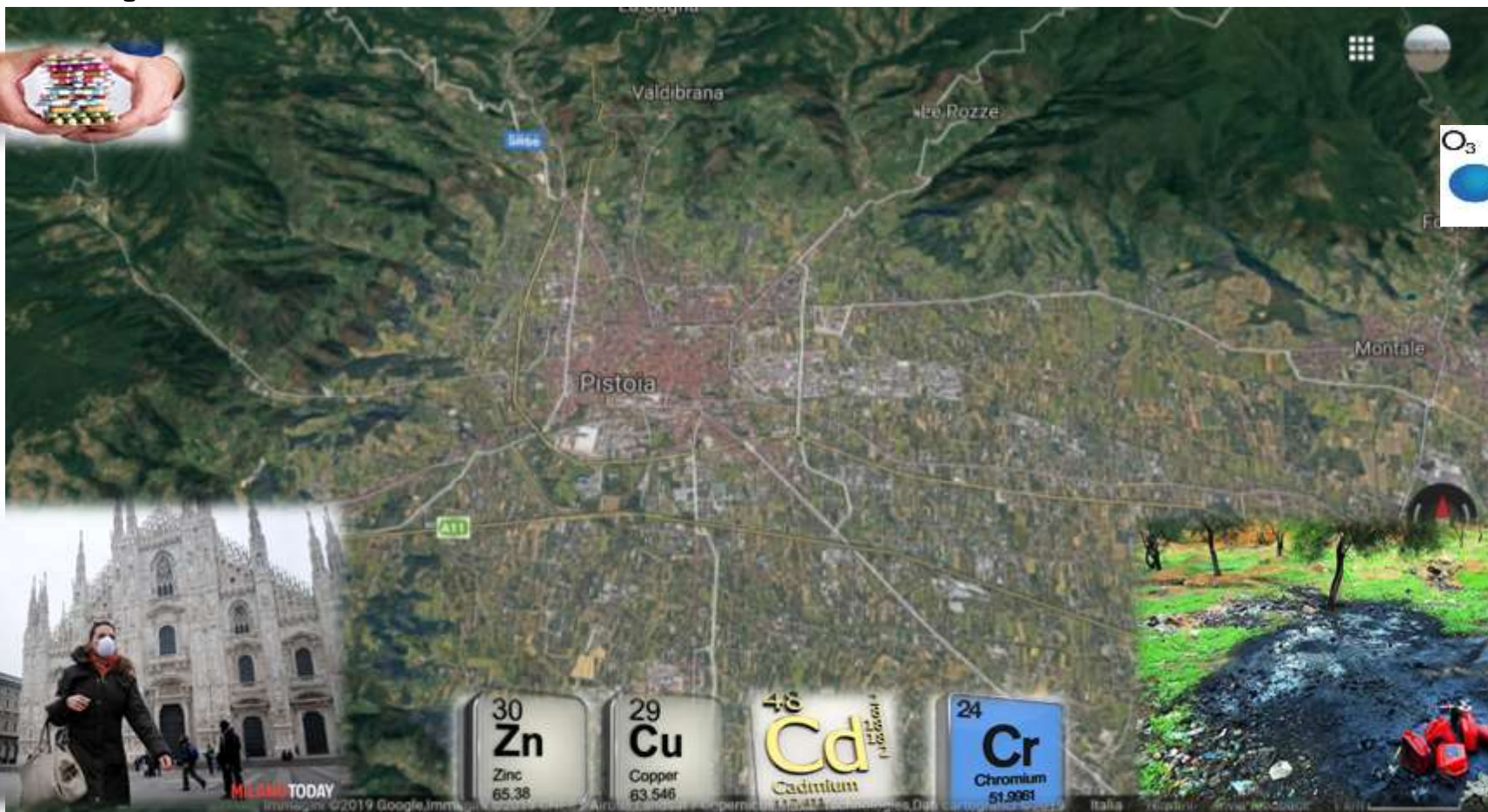
Alberi e inquinamento: cosa fa la ricerca

Luca Sebastiani – luca.sebastiani@santannapisa.it

25 Ottobre 2019



Inquinamento Quali Sostanze e Dove?



MILAM TODAY

Antropocène

(Paul Crutzen - Nobel per la chimica atmosferica)

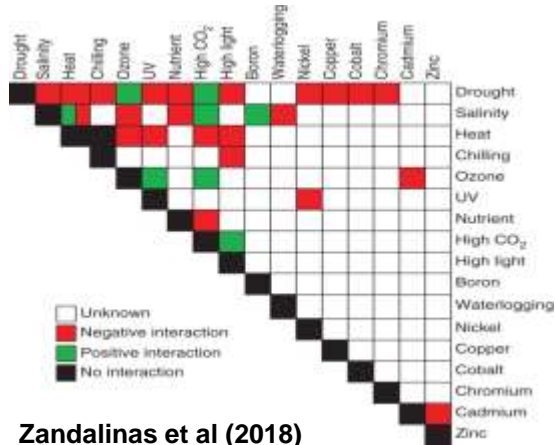
Cambiamento Climatico



Degradazione Suolo



Inquinamento

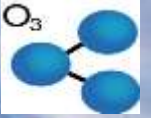


Zandalinas et al (2018)

Physiologia Plantarum 162: 2-12



Quali sono gli inquinanti più importanti ?



Inquinamento Atmosferico

L'inquinamento atmosferico nuoce all'ambiente e alla salute umana. In Europa, **le emissioni di molti inquinanti atmosferici sono diminuite in modo sostanziale negli ultimi decenni**, determinando una migliore qualità dell'aria nella regione. **Le concentrazioni di inquinanti sono tuttavia ancora troppo elevate** e i problemi legati alla qualità dell'aria persistono. Una parte significativa della popolazione europea vive in zone, in particolar modo nelle città, in cui si superano i limiti fissati dalle norme in materia di qualità dell'aria: l'inquinamento da **ozono, biossido di azoto e particolato** pone gravi rischi per la salute. Diversi paesi hanno superato uno o più dei loro limiti relativi alle emissioni per il 2010 per quattro importanti inquinanti atmosferici. Ridurre l'inquinamento atmosferico, quindi, continua a essere importante

Agenzia europea dell'ambiente

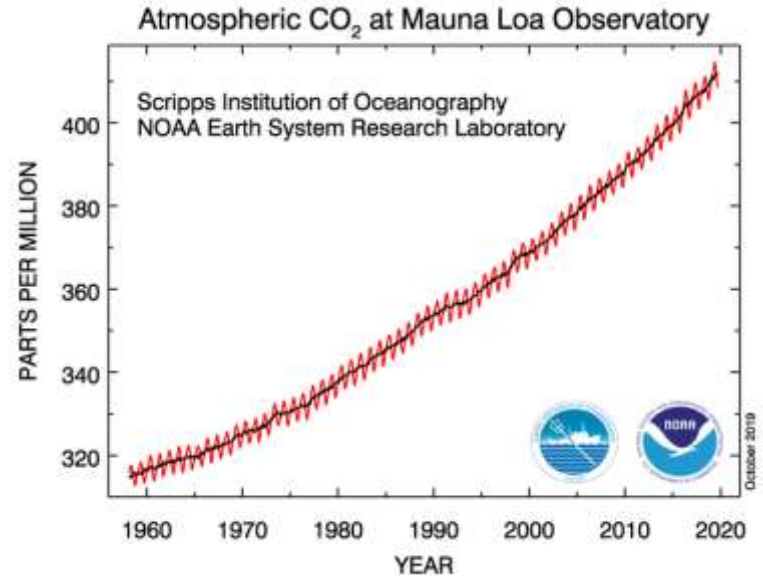


Inquinamento Atmosferico



2019-10-22 20:00

Ozono
Anidride Solforosa
Particolato
Anidride Carbonica

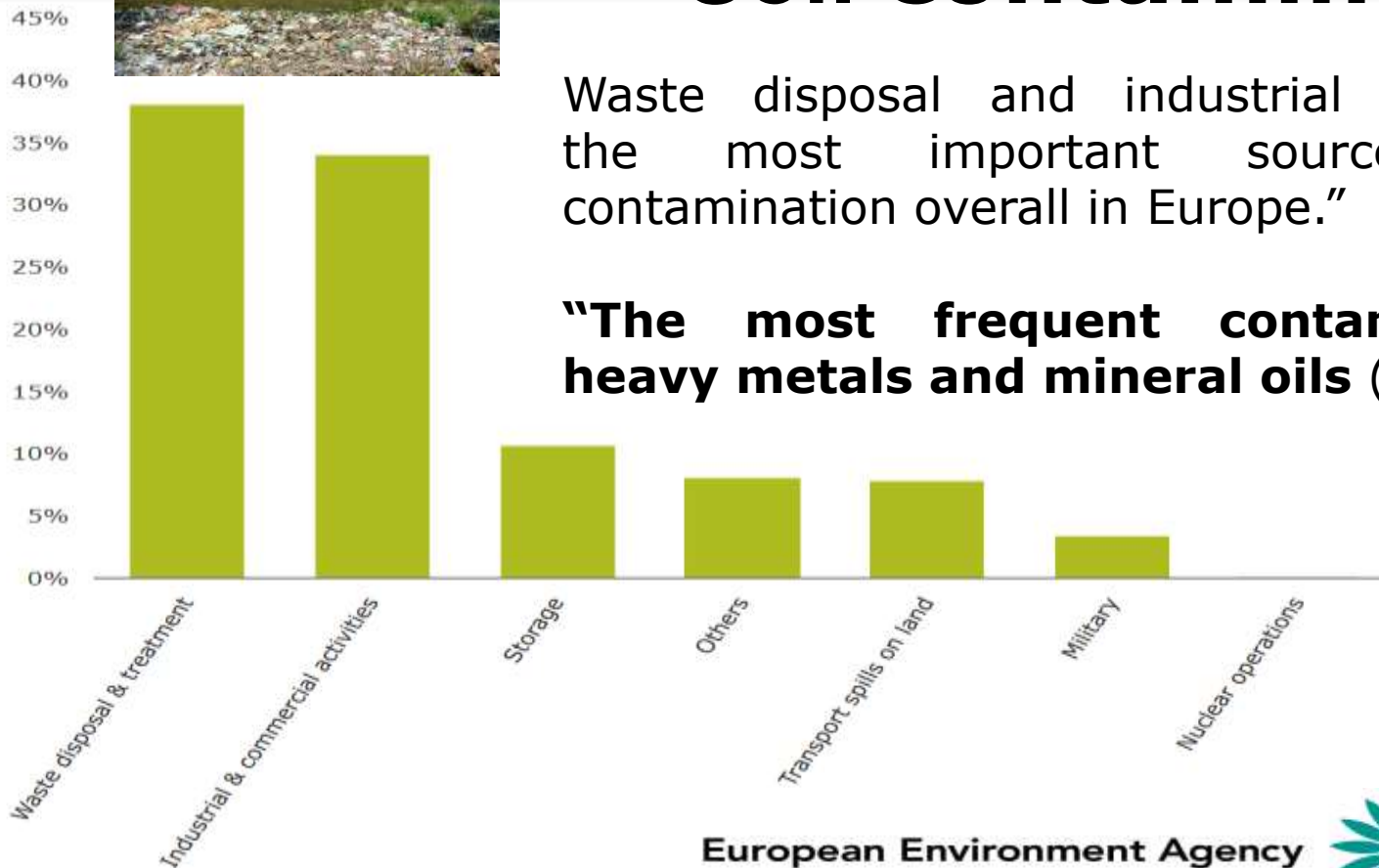




Soil contamination

Waste disposal and industrial activities are the most important sources of soil contamination overall in Europe.”

“The most frequent contaminants are heavy metals and mineral oils (EEA, 2014)”



European Environment Agency



Introduction to Organic Contaminants in Soil: Concepts and Risks

L. Valentin, A. Nousiainen, and A. Mikkonen



Table 1 Type and source of the most relevant group of contaminants in European soils

Contaminants	Example of compounds	Source of contamination ^a	Estimated percentage ^b	References
Heavy metals	Cu, Zn, Cd, Pb, Hg, Cr	Application of animal manure (D) Military facilities (P) Gasoline stations (P) Sawmills and wood preservation sites (P) Mining and metallurgical industry (P,D)	37.3	[21, 42]
Oil hydrocarbons	Alkanes, alkenes, cycloalkanes	Oil industry (P,D)	33.7	[23]
Chlorinated compounds	PCP, PCBs, PCDD/Fs	Manufacture of pesticide and herbicide (D) Wood preservation sites (P) Pulp and paper production (P) Municipal waste incineration (P,D) Plastics, fire-retardants manufacture (P,D)	Chlorinated phenols – 3,6 Chlorinated hydrocarbons – 2,4	[43, 44]
Monomeric aromatic hydrocarbons	Benzene, toluene, ethylbenzene, xylene (BTEX)	Oil industry (P,D) Gasoline stations (P) Manufactured gas plants (P,D)	6	[43]
PAHs	Benzo[a]pyrene, chrysene, fluoranthene	Oil industry (P,D) Gasoline stations (P) Manufactured gas plants (P,D) Wood preservation sites (P) Municipal waste incineration (P,D) Automobile exhaust (D)	13.3	[43, 45]
Nitroaromatics	TNT, nitrobenzene, nitrophenols, atrazine	Manufacture of aniline, dyes, drugs (P,D) Explosive industry, military facilities (P, D) Manufacture of pesticides and herbicides (D)	^c	[46]

^aP = point contamination; D = diffuse contamination

^bAccording to the European Environmental Agency, the estimated percentage is based on the frequency with which a specific contaminant is reported to be the most important in the investigated site [23]

^cInformation not available

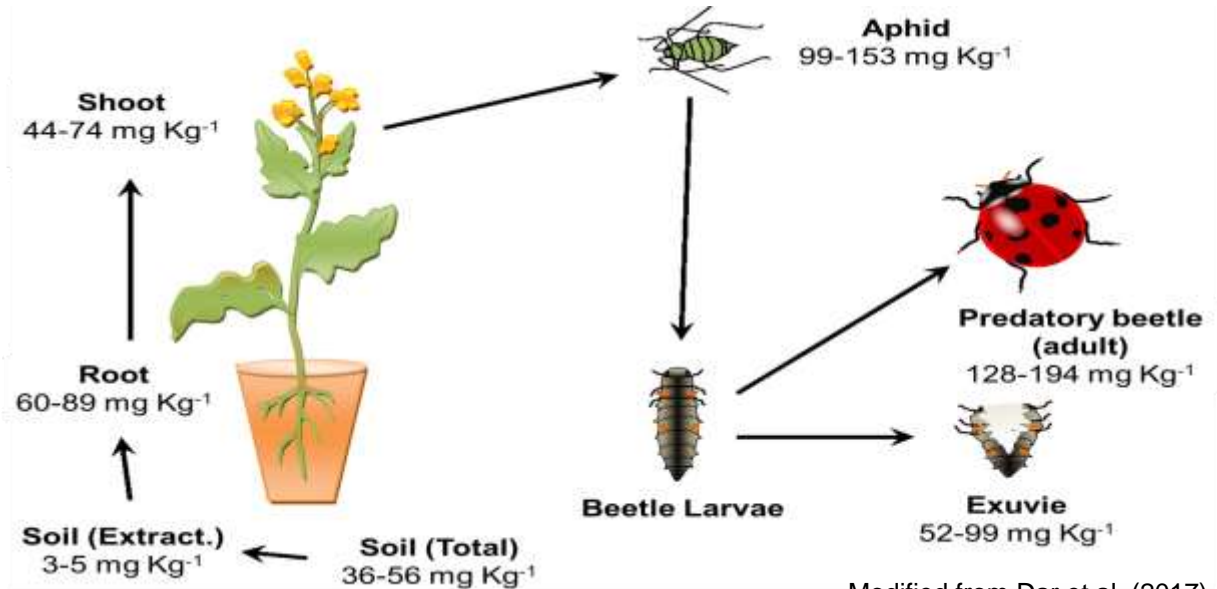
Zinc is an essential element for both plants and humans, but it is toxic in excess amounts



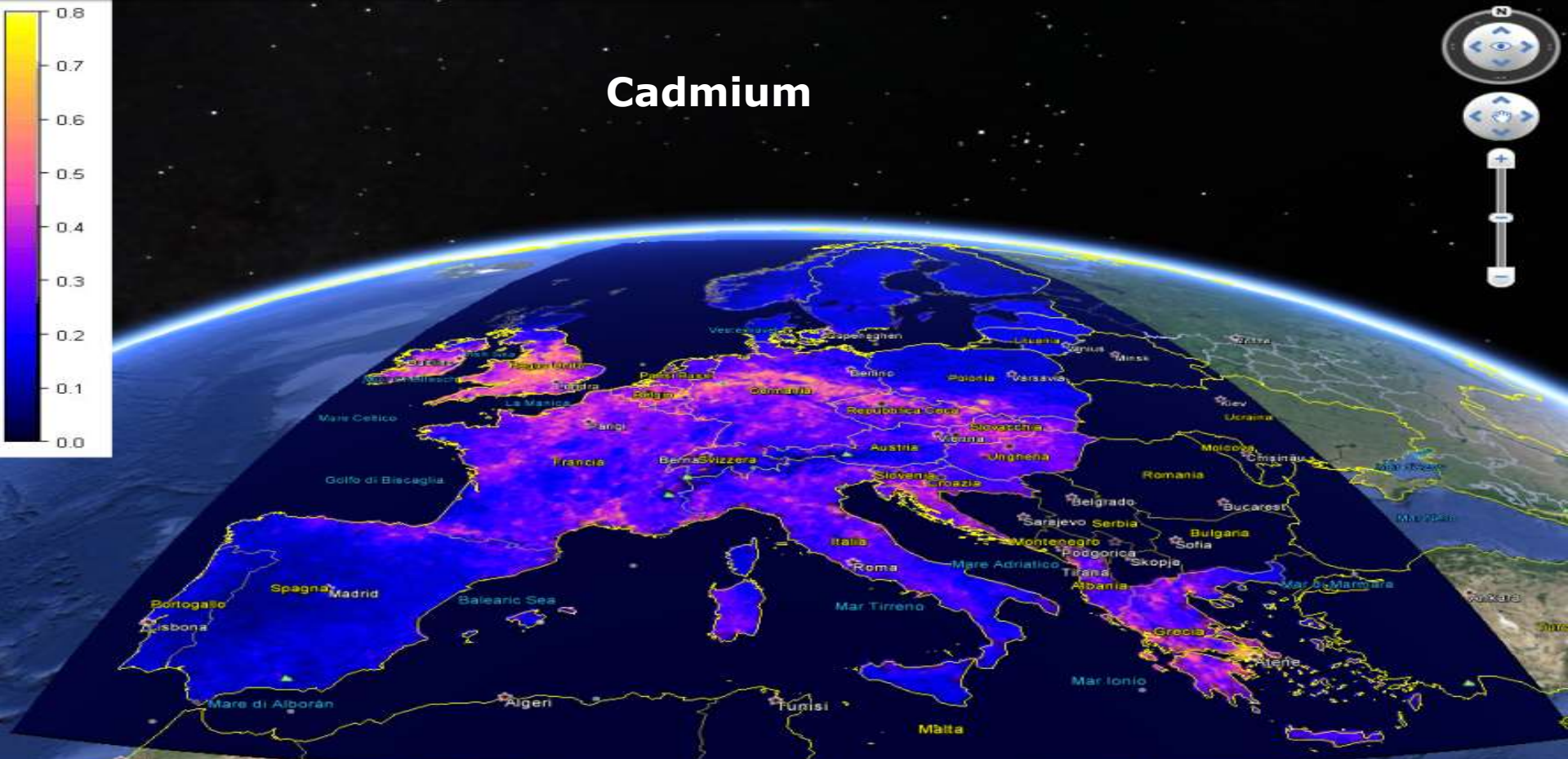
Zinc is propagated throughout the food chain by bioaccumulation



Zinc excess in soil might be either of geological or anthropogenic origin.



Cadmium



Spatial distribution in topsoils 1588 geo-referenced samples FOREGS Geochemical database (Source: European Soil Data Center, <http://eusoils.jrc.ec.europa.eu/>)



Nick Palmer (TWND)

A GLOBAL WATER-QUALITY CRISIS AND THE ROLE OF AGRICULTURE

Xenobiotici Emergenti



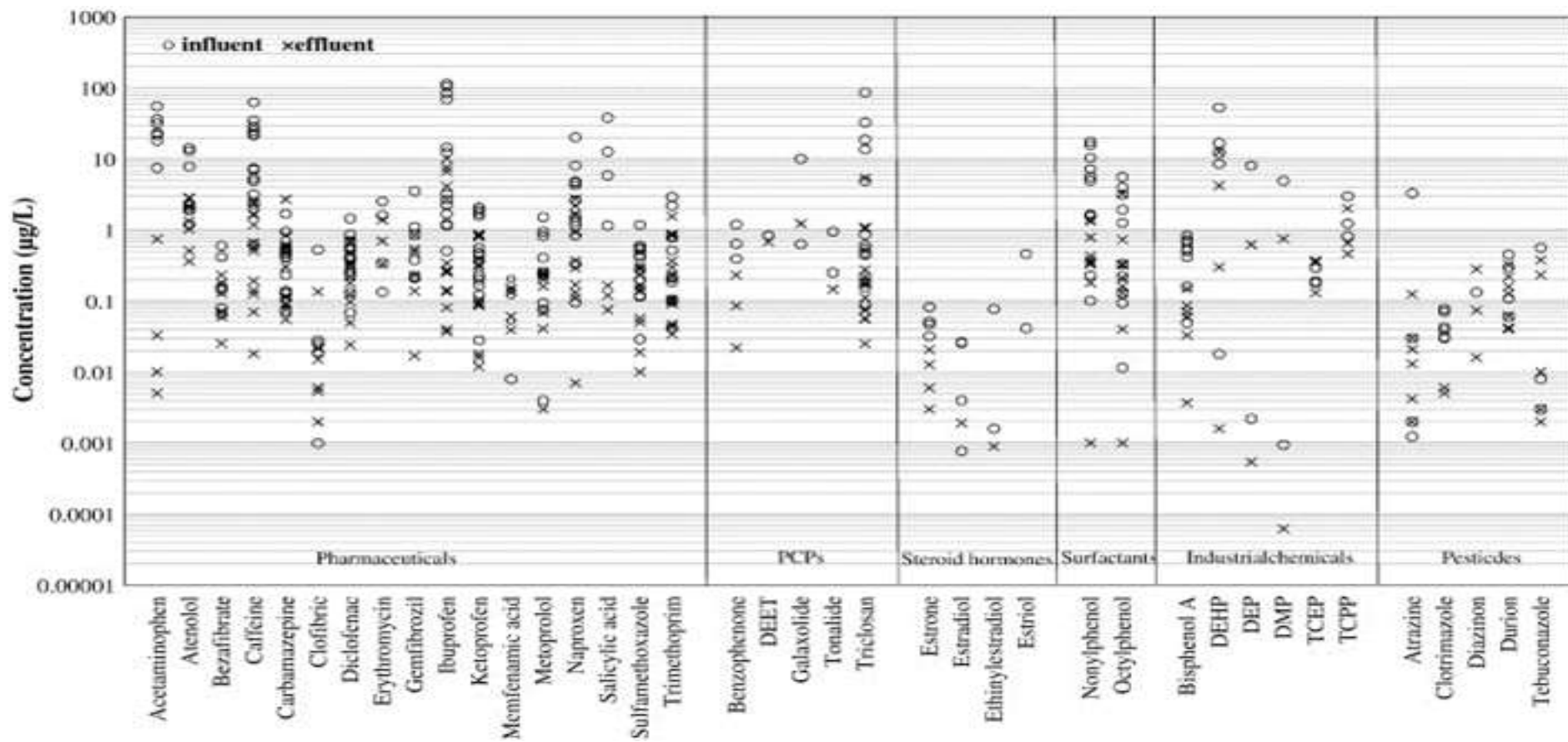
Emerging Contaminants

Pharmaceutical and Personal Care Products



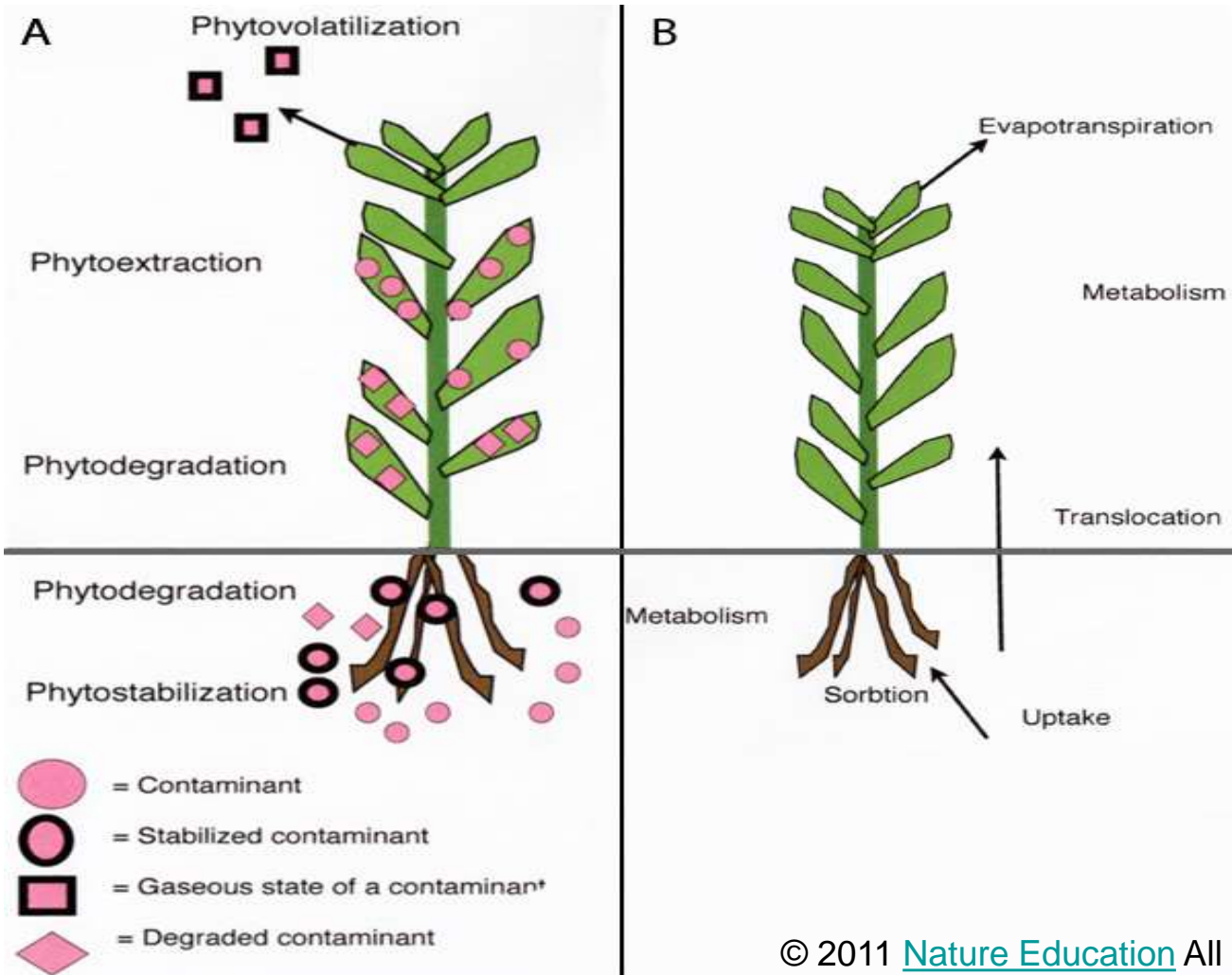
Continuously released into the aquatic environment

Not efficiently removed by traditional wastewater treatment plants

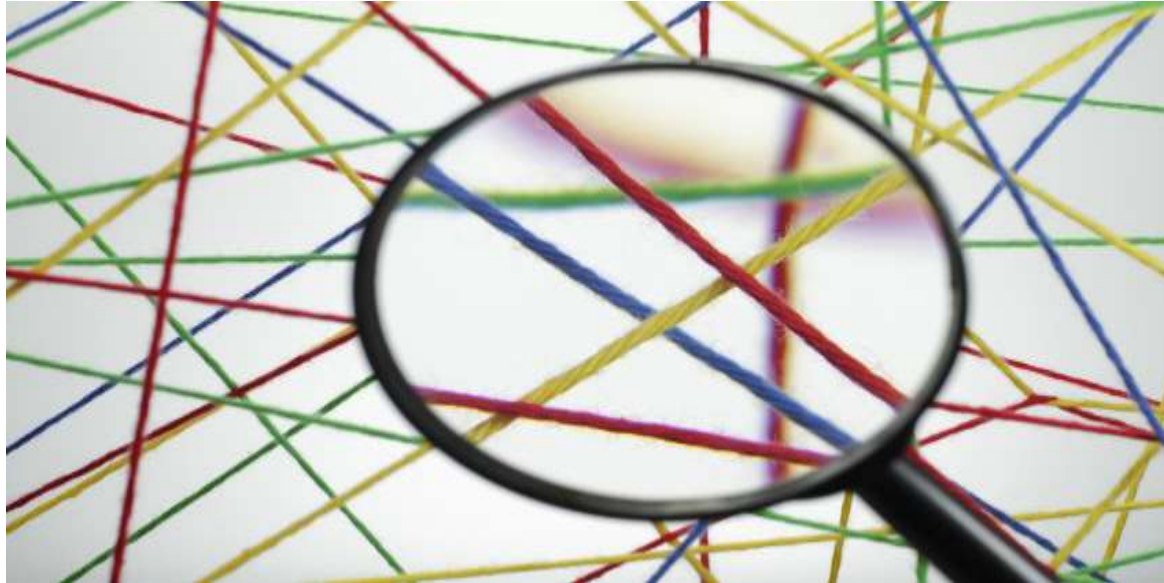


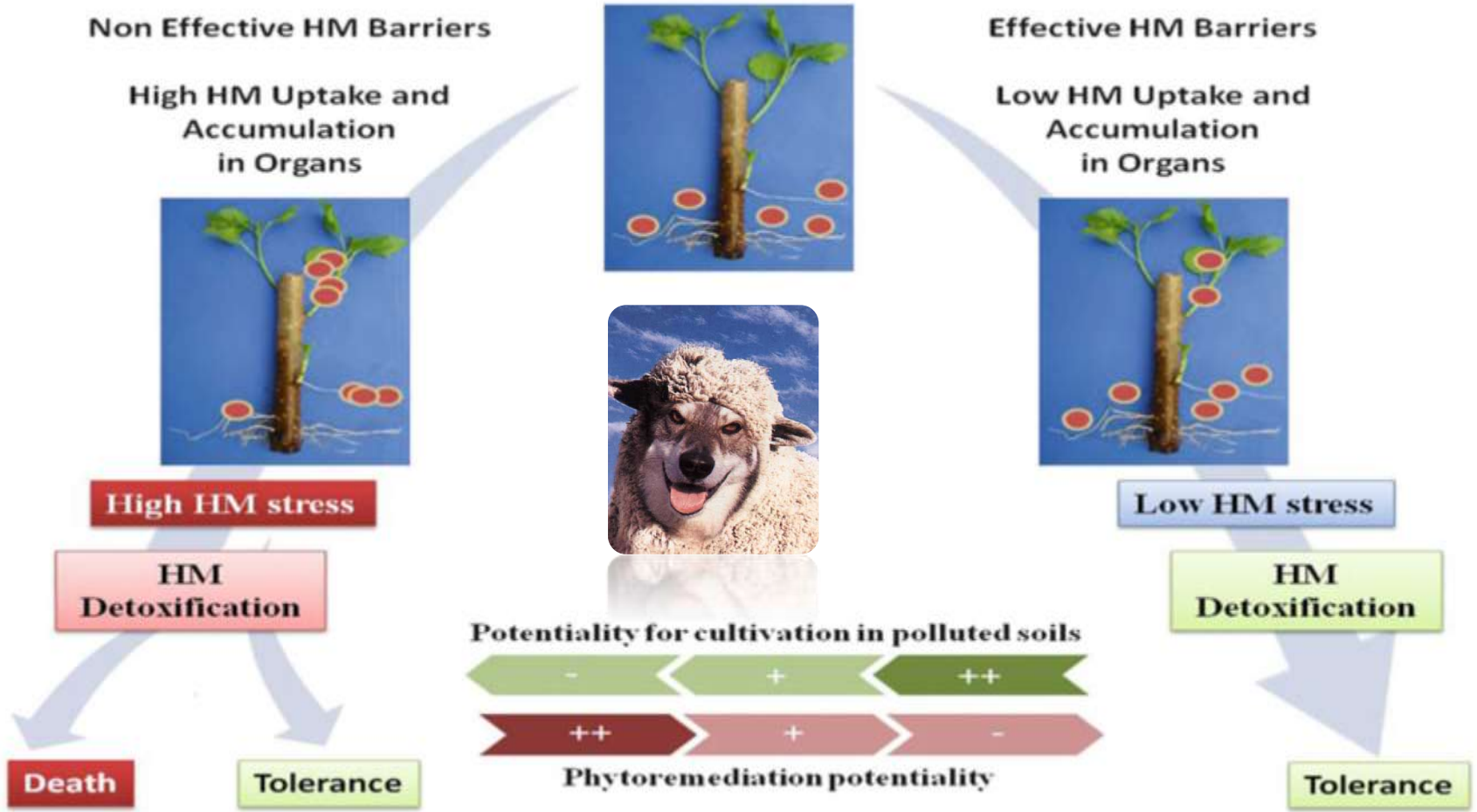
Piante ?





Il nostro approccio al problema





Sebastiani L. et al. (2014). Heavy metals stress on poplar: molecular and anatomical modifications. In: Approaches to Plant Stress and their Management, pp 267-279 – Springer.

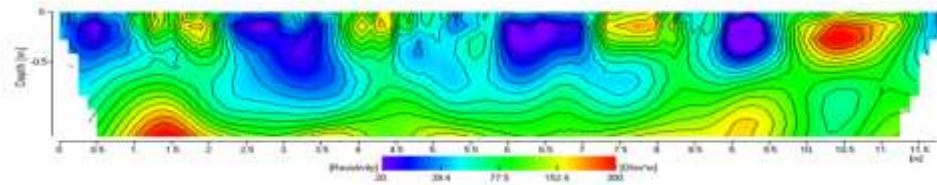
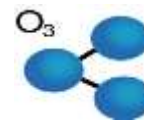
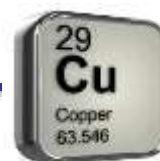


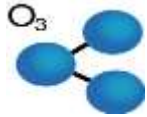
Campo

Serra

Cella di Crescita

Vitro





Ozone: **100 ppb**

Control: **< 3 ppb**
5 h day⁻¹

Tree Physiology 19, 391–397
© 1999 Heron Publishing—Victoria, Canada

Physiological and morphological responses of olive plants to ozone exposure during a growing season

ANTONIO MINNOCCI,¹ ALBERTO PANICUCCI,² LUCA SEBASTIANI,¹ GIACOMO LORENZINI² and CLAUDIO VITAGLIANO¹



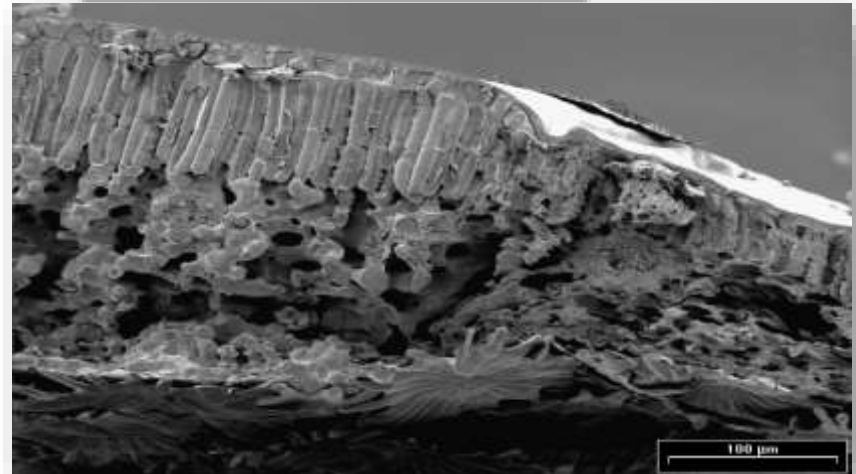
0 Cv. Frantoio
Cv. Moraiolo

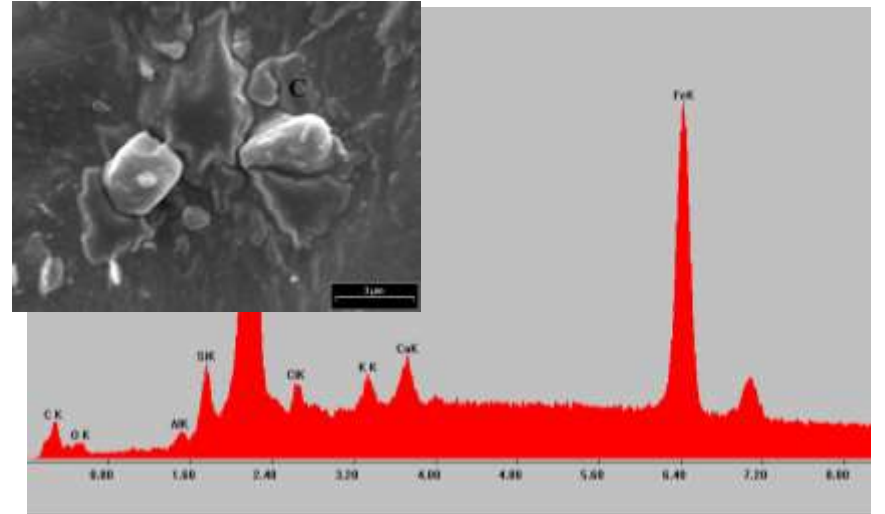
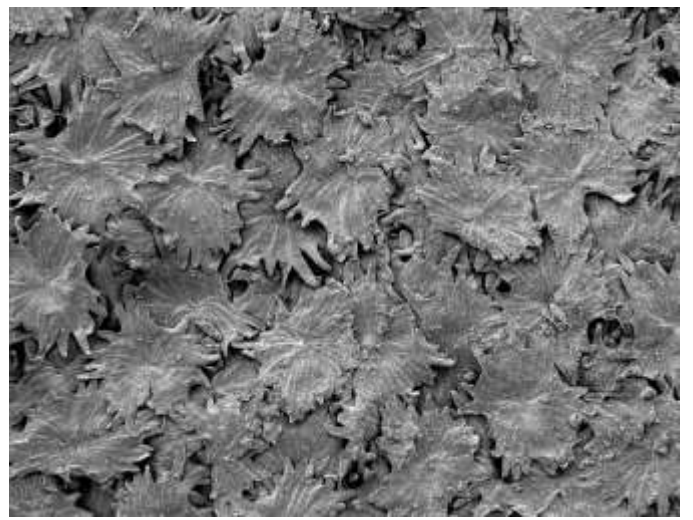
100

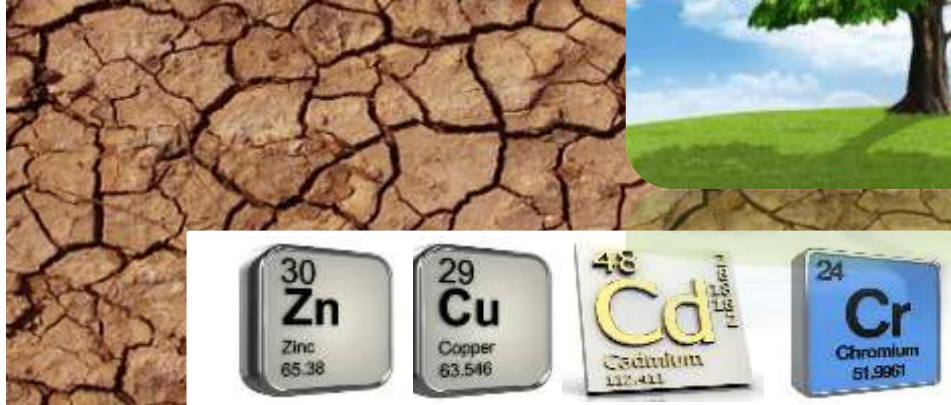
120

Leaf drop and development
of necrotic spots

(days)





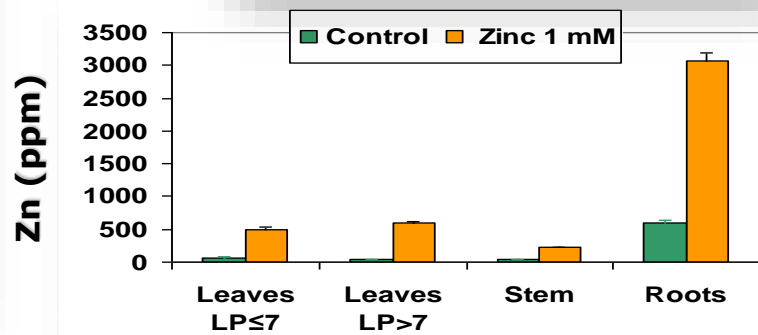
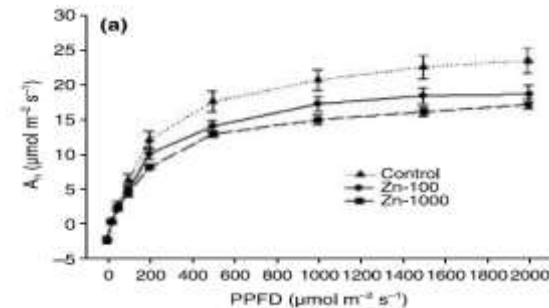
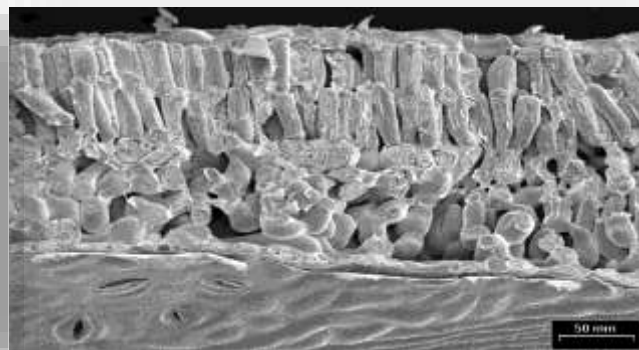


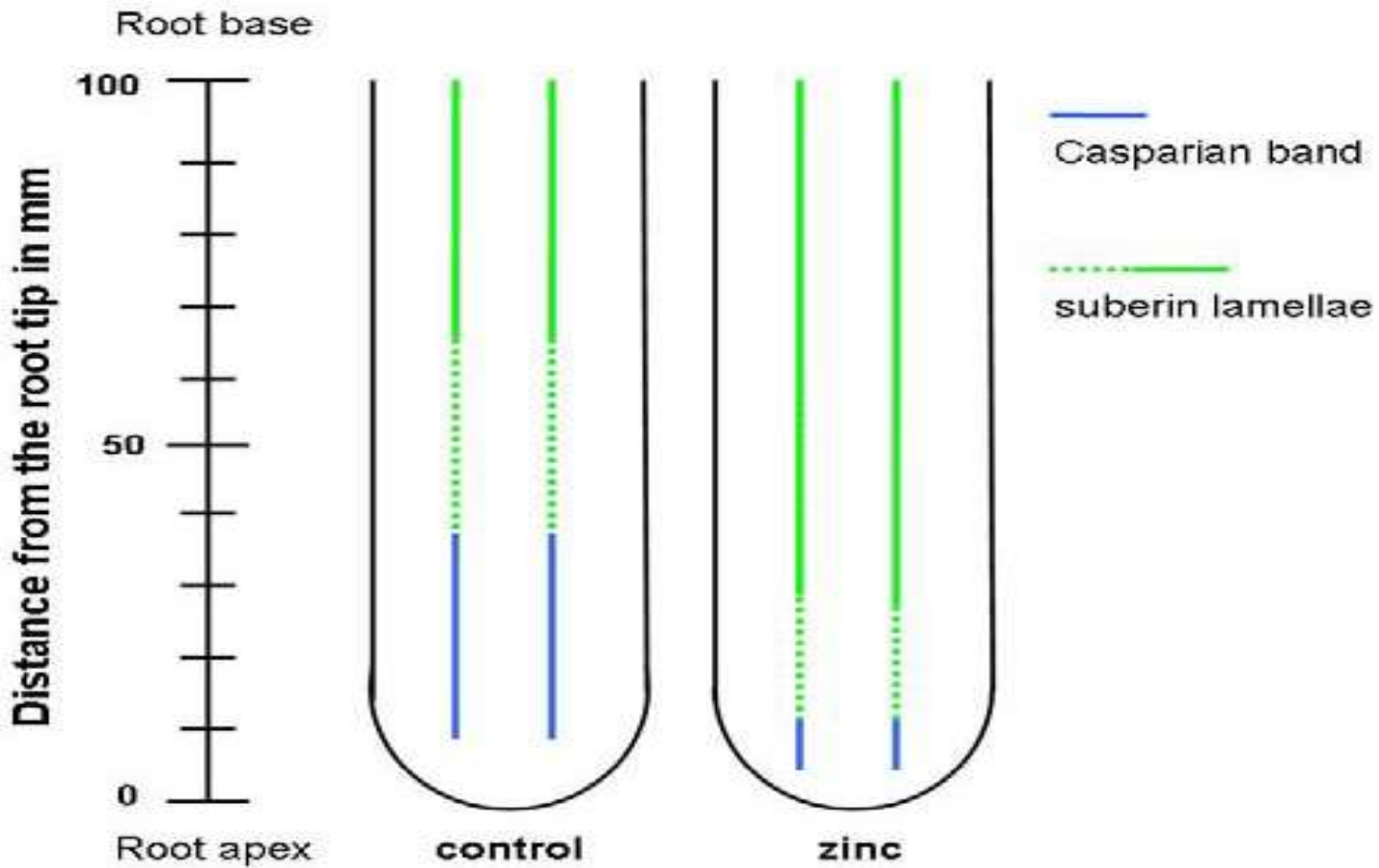
Modello sperimentale

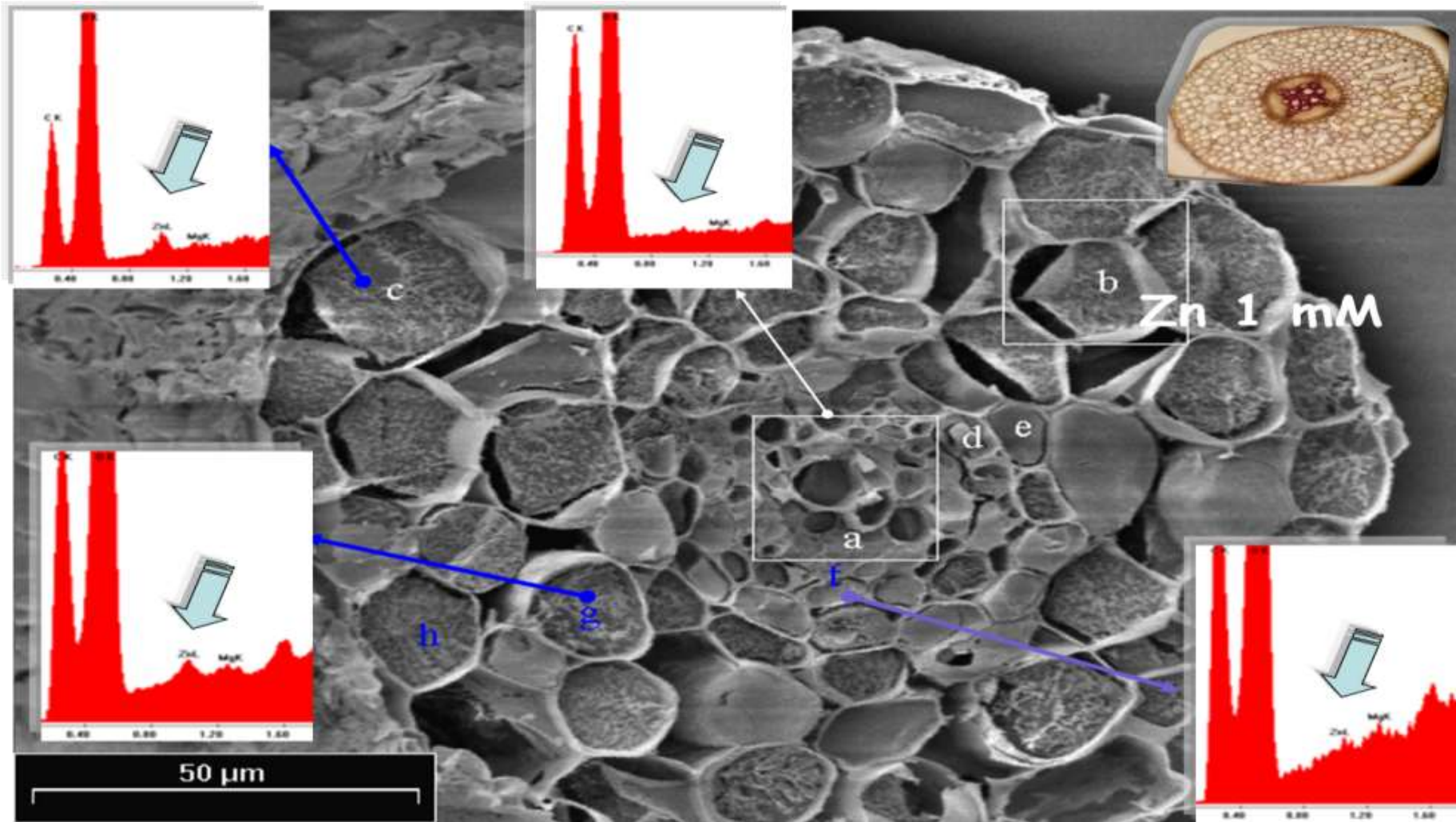


I-214

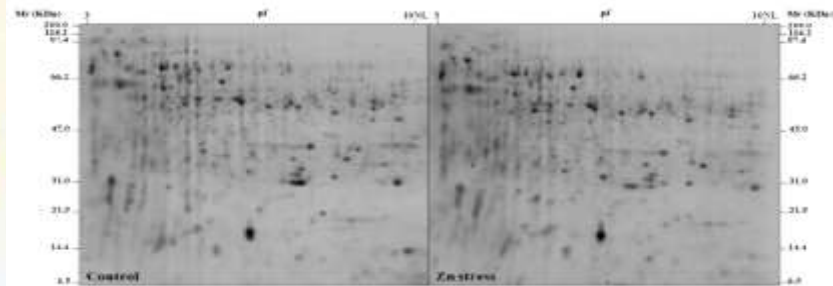
- Biomassa
- Fotosintesi
- Zn Uptake







Meccanismi molecolari coinvolti nell'omeostasi

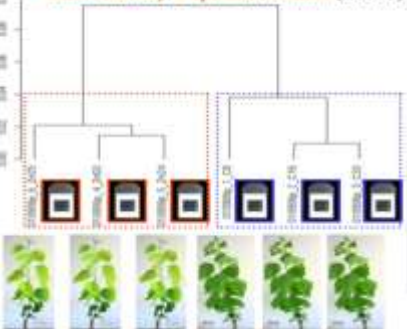


Transcriptome analyses of *Populus × euramericana* clone I-214 leaves exposed to excess zinc

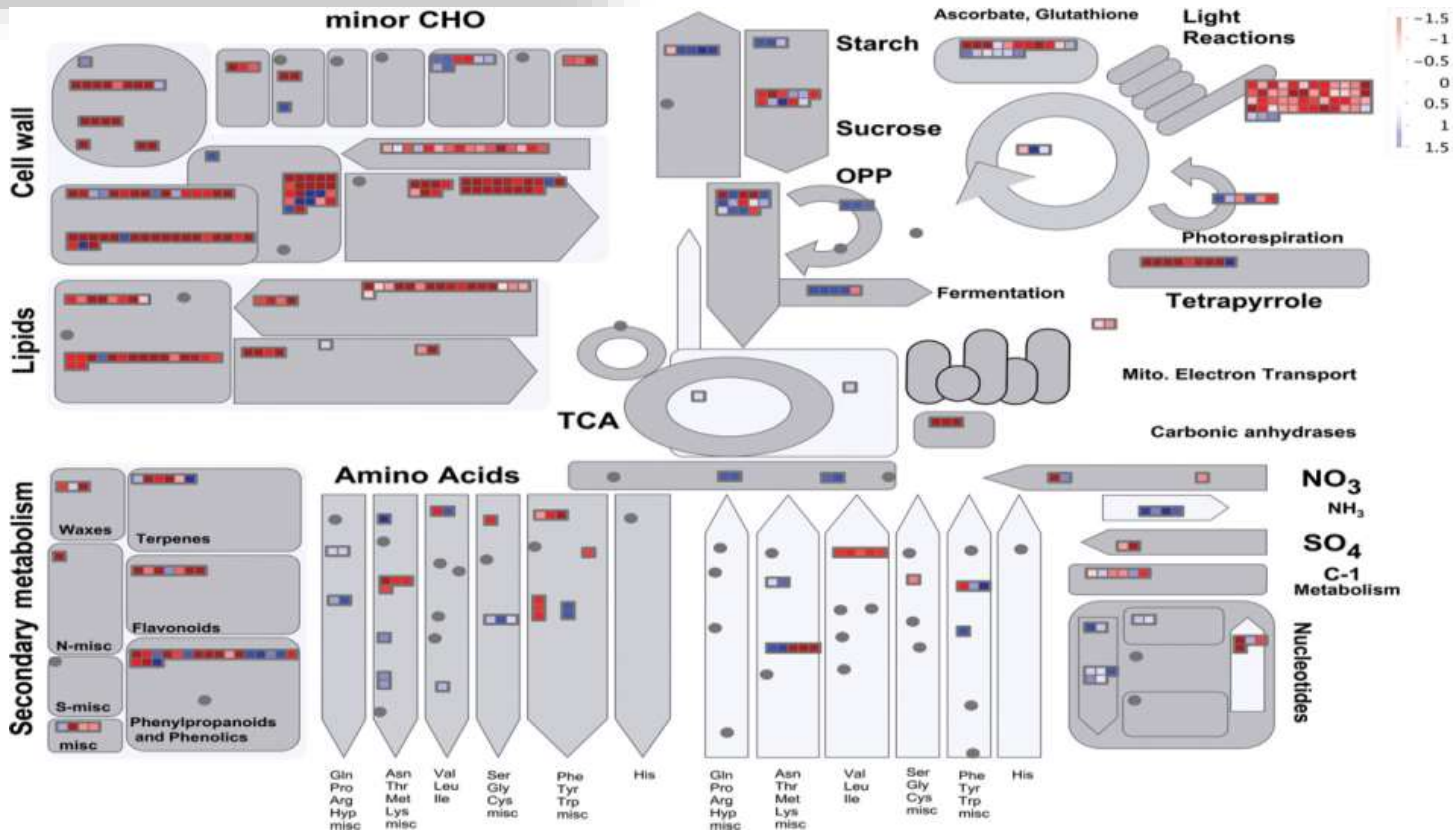
Daniela Di Baccio^{1,9}, Giulio Galla², Tania Bracci¹, Andrea Andreucci³, Gianni Barcaccia², Roberto Tognetti⁴ and Luca Sebastiani¹

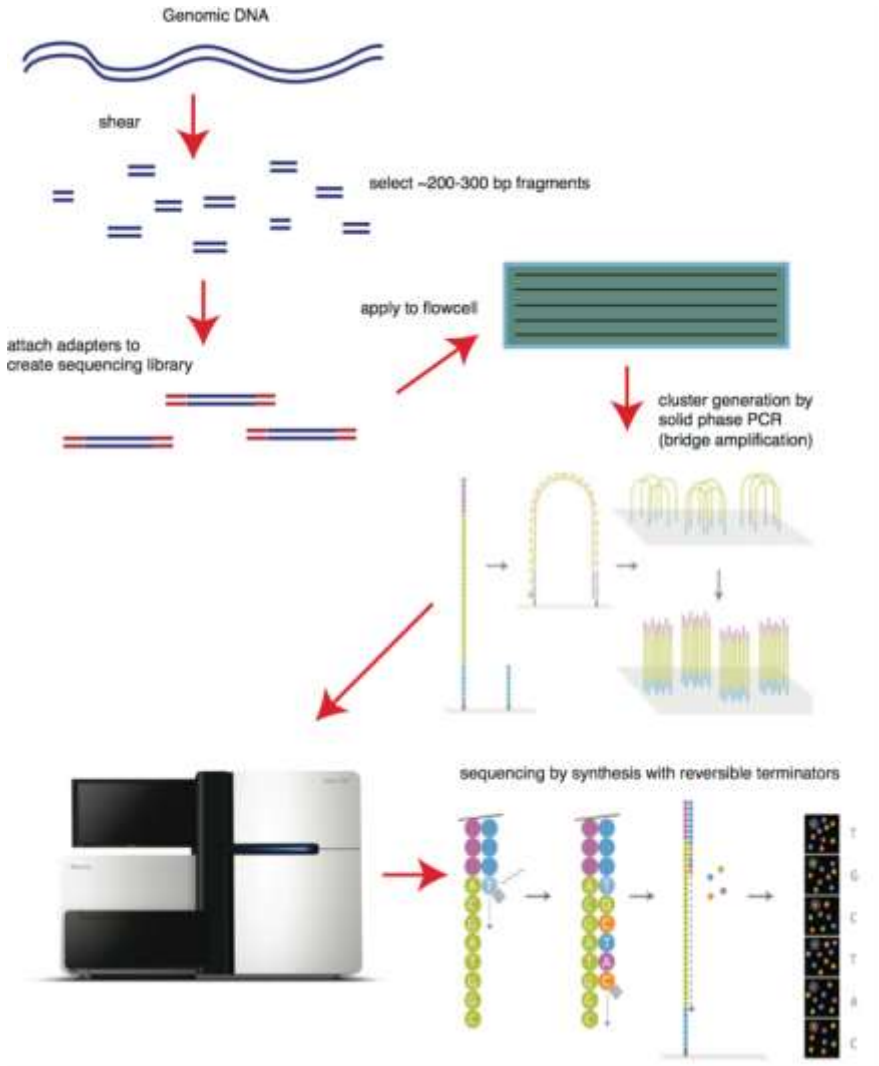
¹Biolab, Life Sciences Institute, Scuola Superiore Sant'Anna, Piazza Martiri della Libertà 33, I-56127 Pisa, Italy; ²Department of Environmental Agronomy and Crop Science, Università degli Studi di Padova, Viale dell'Università 16, I-35020 Legnaro (Padova), Italy; ³Department of Biology, Università di Pisa, Via L. Ghivetti, I-56100 Pisa, Italy; ⁴Coastal and Dune Lab—Dipartimento di Scienze e Tecnologia per l'Ambiente e il Territorio, Università degli Studi del Molise, Contrada Fonte Lappone, I-86090 Pesche, Italy; ⁹Corresponding author (dibaccio@ssnup.it)

Differentially Expressed Genes (DEGs)



AFFYMETRIX®



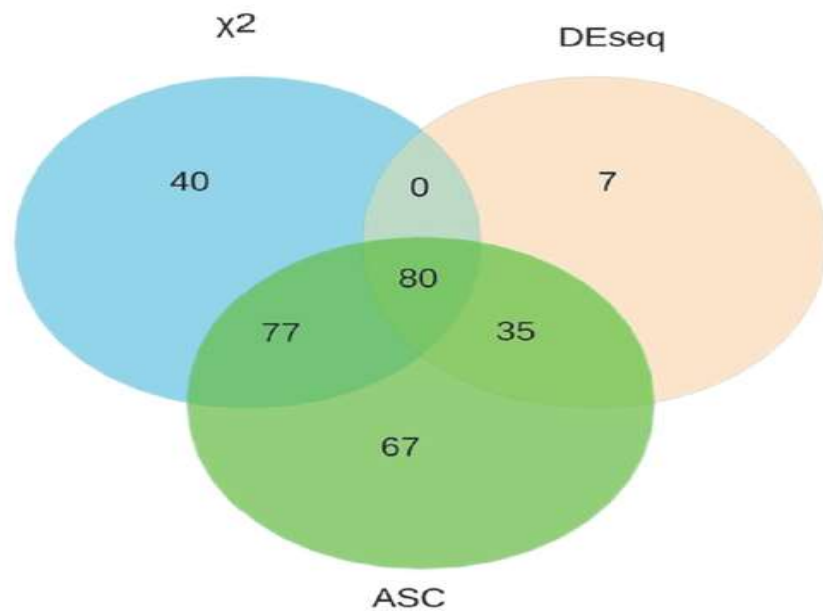


RESEARCH ARTICLE

RNA Sequencing of *Populus x canadensis* Roots Identifies Key Molecular Mechanisms Underlying Physiological Adaption to Excess Zinc

Andrea Ariani¹, Daniela Di Baccio^{1*}, Stefania Romeo¹, Lara Lombardi¹, Andrea Andreucci¹, Alexander Lux^{1,2,3}, David Stephen Horner⁴, Luca Sebastiani^{1*}

1 Biollato Institute of Life Sciences, Scuola Superiore Sant'Anna, I-56127 Pisa, Italy, **2** Department of Biology, Università degli Studi di Pisa, I-56126 Pisa, Italy, **3** Department of Plant Physiology, Faculty of Natural Science, Comenius University in Bratislava, Bratislava, Slovakia, **4** Institute of Chemistry, Slovak Academy of Sciences, Bratislava, Slovakia, **5** Department of Biotechnology, Università degli Studi di Milano, Milano, Italy



Control



Stressed



Protein extraction and sample preparation
in TCA/acetone

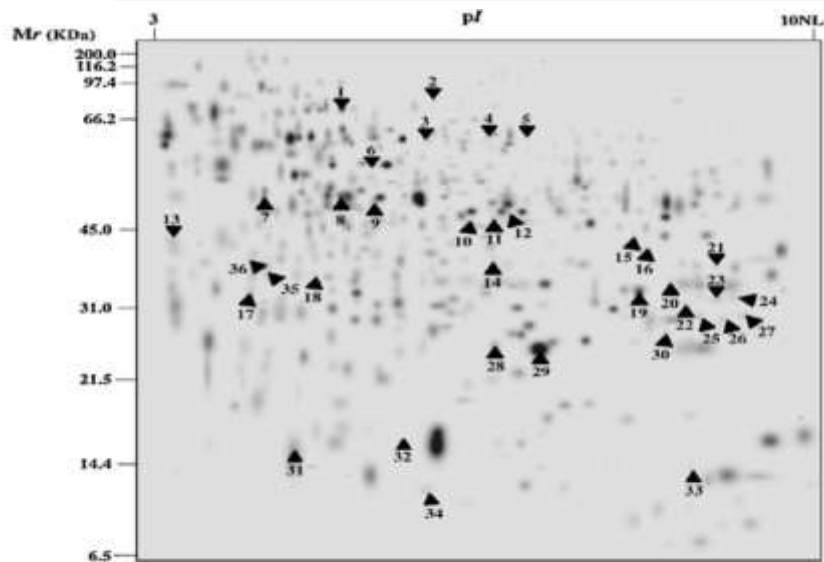


Fig. 3. Filtered image of the two-dimensional electrophoresis proteome reference map of *Populus x euramericana* (I-214 clone) root. Differentially represented protein spots between Zn-treated and control roots are indicated, Spot numbering corresponds to the proteins listed in Table 2.



Physiology

Proteomic analysis of *Populus x euramericana* (clone I-214) roots to identify key factors involved in zinc stress response

Stefania Romeo^{a,1}, Dalila Trupiano^{b,1}, Andrea Ariani^d, Giovanni Renzone^c,
Gabiella S. Scippa^b, Andrea Scaloni^c, Luca Sebastiani^{a,*}



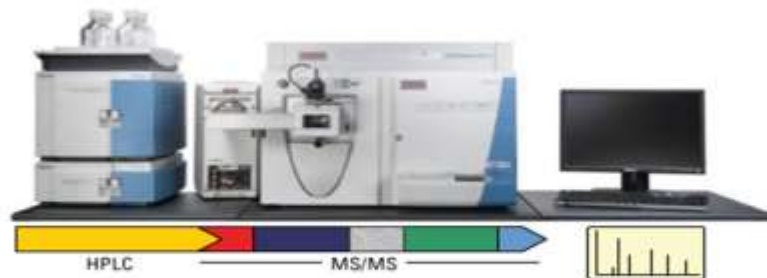
Spot identification and gel protein digestion

Spot numbering (1, 2, 3...x), excision and in-gel protein digestion (with trypsin etc.)

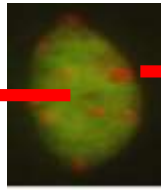
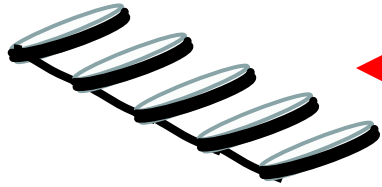


Protein identification

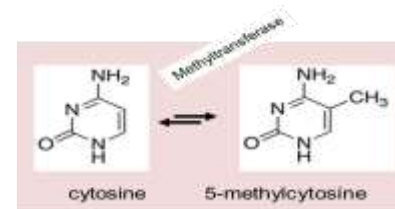
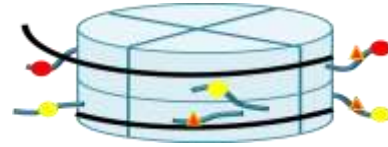
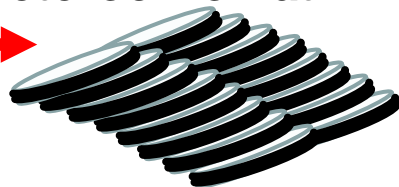
nanoLC-ESI-LIT-MS/MS and protein identification



euchromatin



heterochromatin



Epigenetic modifications of chromatin structure are extremely important in mediating stress responses in plants. Epigenetic modifications are especially important in perennial species such as trees, where they contribute to phenotypic plasticity and adaptation to unfavorable environments.

Environmental and Experimental Botany 132 (2016) 16–27



Contents lists available at ScienceDirect

Environmental and Experimental Botany

journal homepage: www.elsevier.com/locate/envexpbot

Comparative epigenomic and transcriptomic analysis of *Populus* roots under excess Zn

Andrea Ariani^{a,*}, Stefania Romeo^a, Andrew T. Groover^{b,c}, Luca Sebastiani^d

^a BioLabs, Institute of Life Sciences, Scuola Superiore Sant'Anna, Piazza Martiri della Libert , 33, 56127 Pisa, Italy

^b US Forest Service, Pacific Southwest Research Station, Davis, CA 95618, USA

^c Department of Plant Biology, University of California, Davis, CA 95618, USA



Variabilità genetica ...

Phytoremediation of Zn: Identify the Diverging Resistance, Uptake and Biomass Production Behaviours of Poplar Clones Under High Zinc Stress

Stefania Romeo • Alessandra Francini •
Andrea Ariani • Luca Sebastiani



I-214

Populus x euramericana



Jean Pourtet

Populus nigra



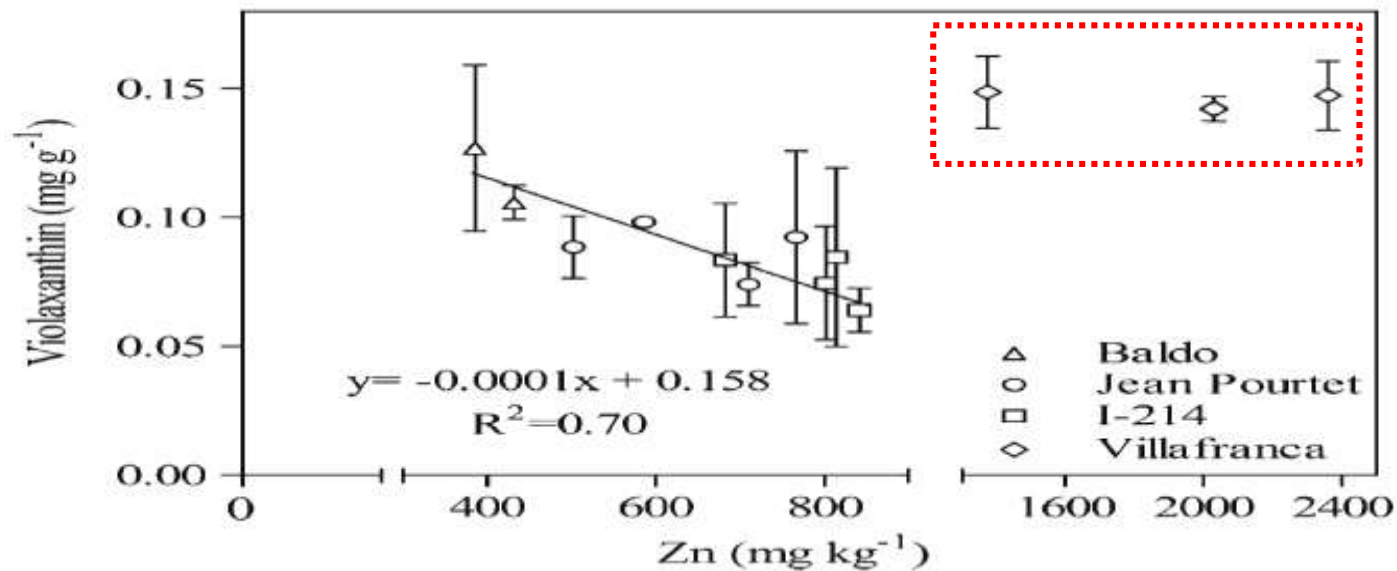
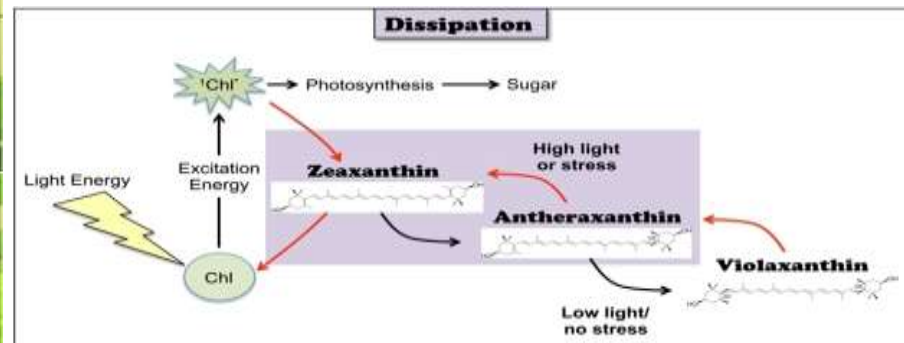
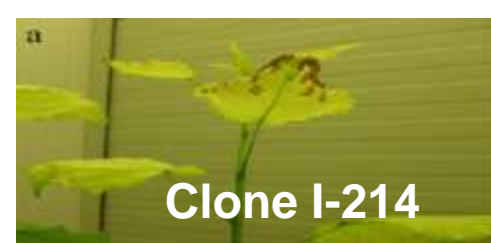
Baldo

Populus deltoides



Villafranca

Populus alba

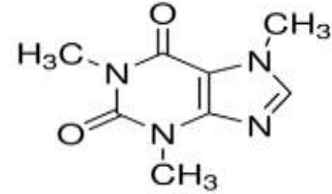


Inquinamento Acqua



Caffeine

- ❖ One of the most consumed drug worldwide
- ❖ Persistent in the aquatic environment



Espresso (30 mL)
47-64 mg

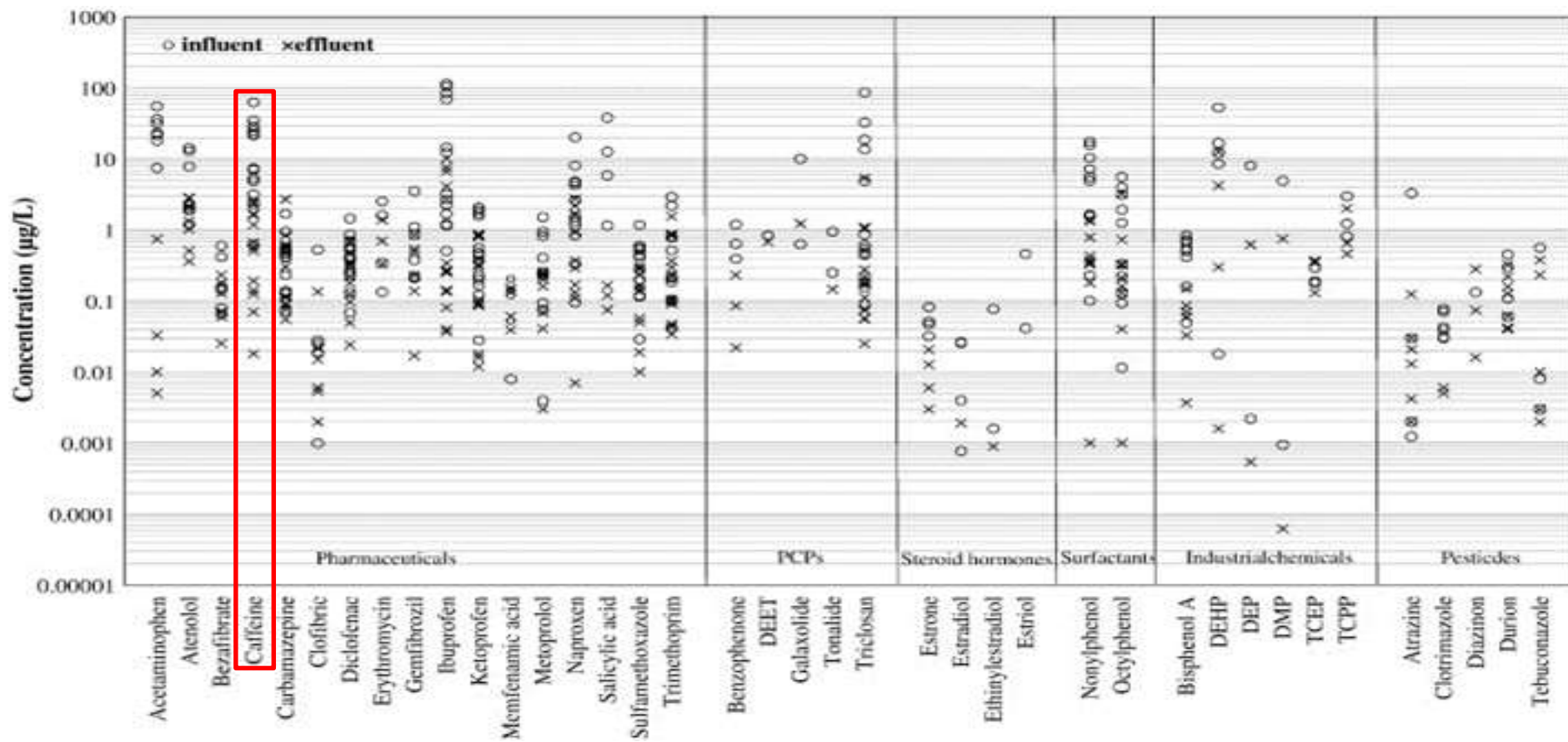


Black Tea (237 mL)
25-48 mg



1.22 $\mu\text{g ml}^{-1}$ \pm 2.45 $\mu\text{g ml}^{-1}$

Int J Sports Med. 2005 Nov;26(9):714-8.
Van Thuyne W1, Roels K, Delbeke FT.





Pioppo e Caffeina?

Environ Sci Pollut Res (2016) 23:7298–7307
DOI 10.1007/s11356-015-5935-z



RESEARCH ARTICLE

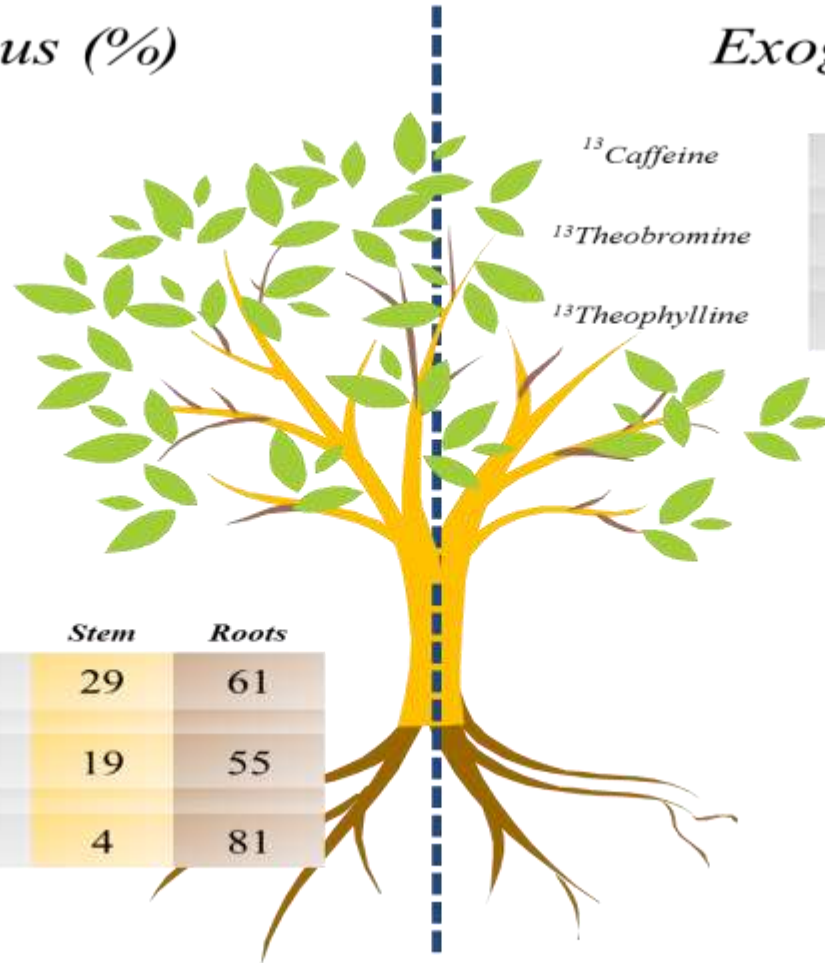
Degradation of exogenous caffeine by *Populus alba* and its effects on endogenous caffeine metabolism

Erika C. Pierattini¹ • Alessandra Francini¹ • Andrea Raffaelli² • Luca Sebastiani¹



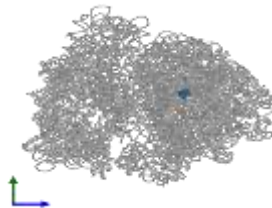
Endogenous (%)

Exogenous (%)



	<i>Leaves</i>	<i>Stem</i>	<i>Roots</i>
¹³ Caffeine	99	1	0
¹³ Theobromine	98	0	2
¹³ Theophylline	59	17	24

	<i>Leaves</i>	<i>Stem</i>	<i>Roots</i>
<i>Caffeine</i>	10	29	61
<i>Theobromine</i>	26	19	55
<i>Theophylline</i>	15	4	81



Erythromycin bound
to
E. coli ribosome

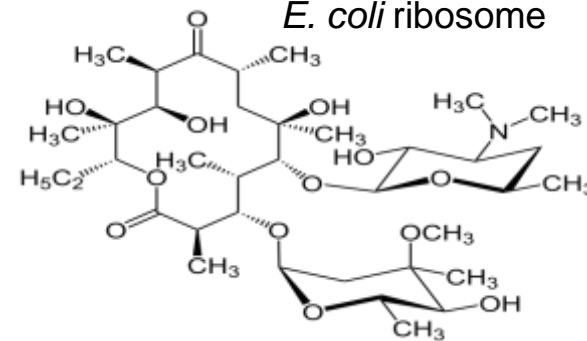


Morpho-physiological response of *Populus alba* to erythromycin: A timeline of the health status of the plant

Erika Carla Pierattini ^a, Alessandra Francini ^{a,*}, Andrea Raffaelli ^b, Luca Sebastiani ^a

^a Institute of Life Sciences, Scuola Superiore Sant'Anna, Piazza Martiri della Libertà 33, I-56127 Pisa, Italy

^b CNR - Istituto di Fisiologia Clinica, Via Moruzzi 1, I-56124 Pisa, Italy



- ❖ Antibiotic inhibitor of protein synthesis in bacteria
- ❖ Persistence in the aquatic environment is a risk for the **rising and spreading of antibiotics resistance mechanisms**
- ❖ Stable and doesn't undergo photodegradation processes

Erythromycin



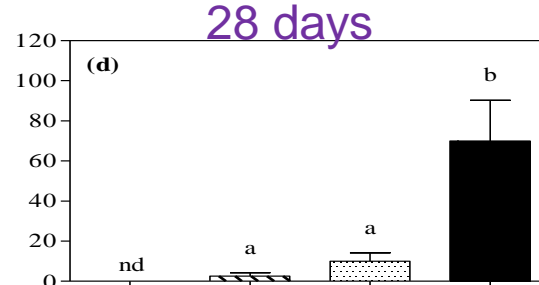
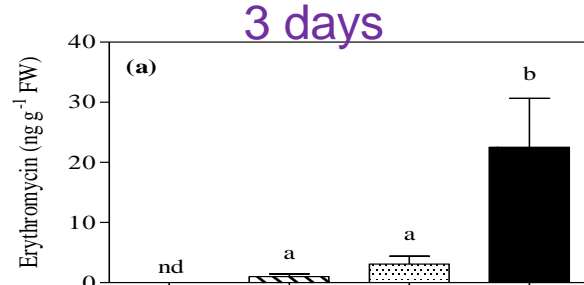
0 mg L⁻¹

0.01 mg L⁻¹

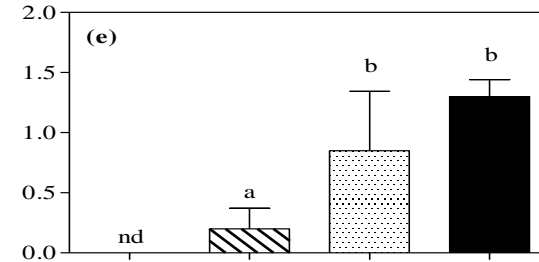
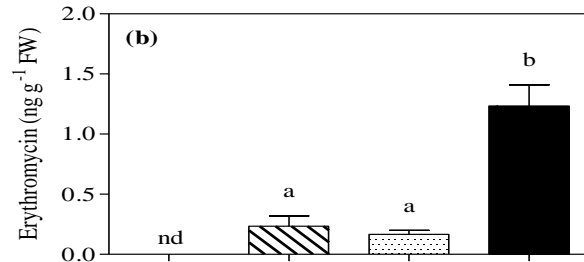
0.1 mg L⁻¹

1 mg L⁻¹

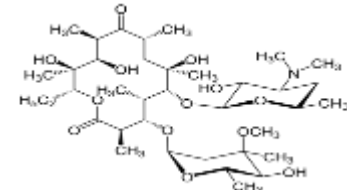
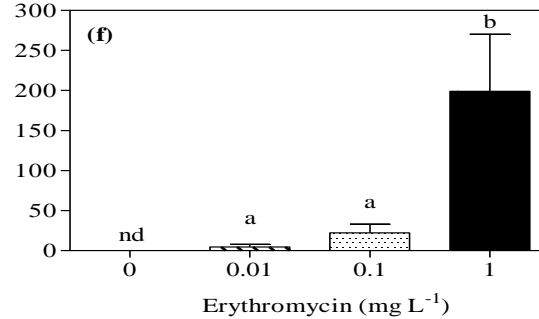
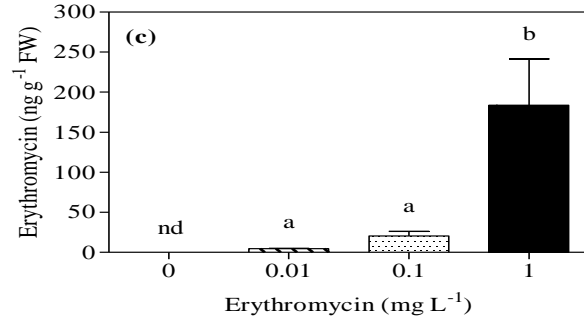
Leaves



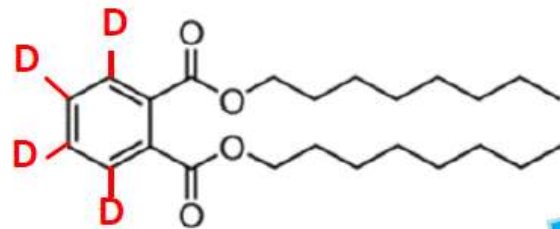
Stem



Roots

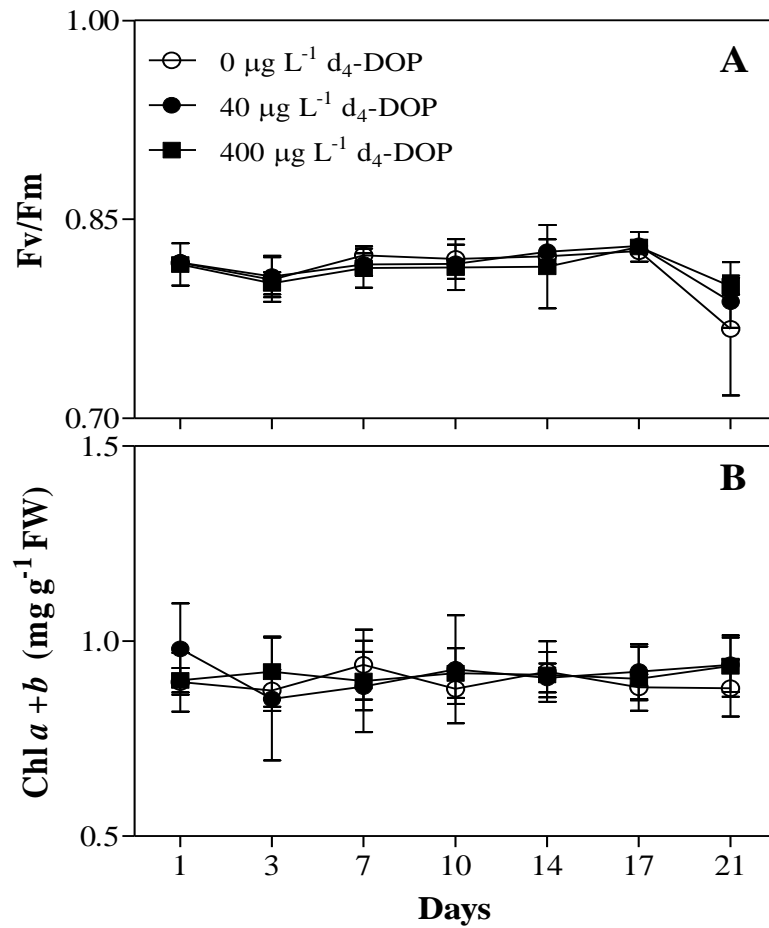
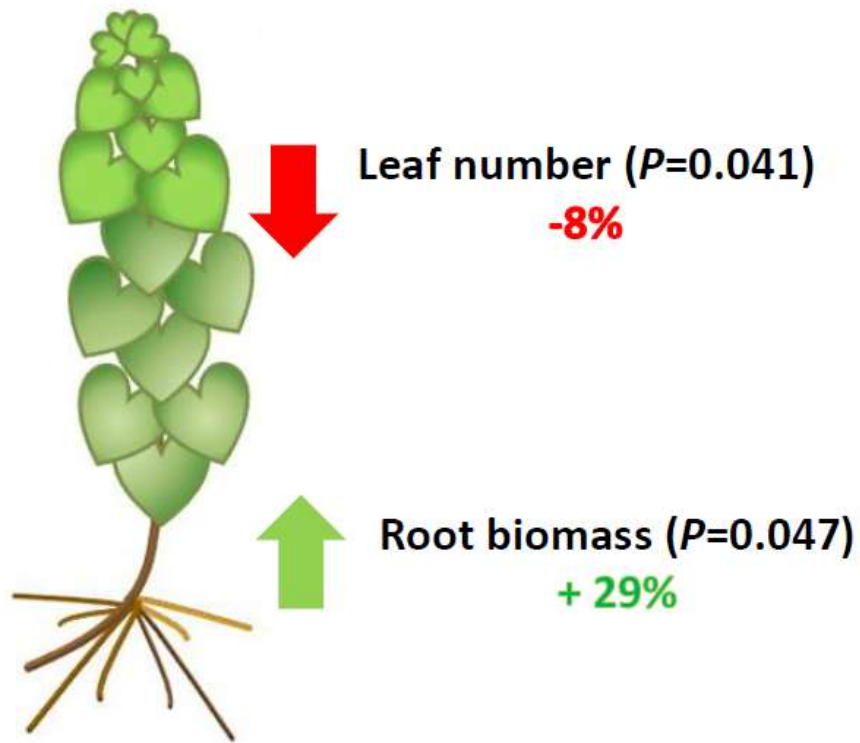


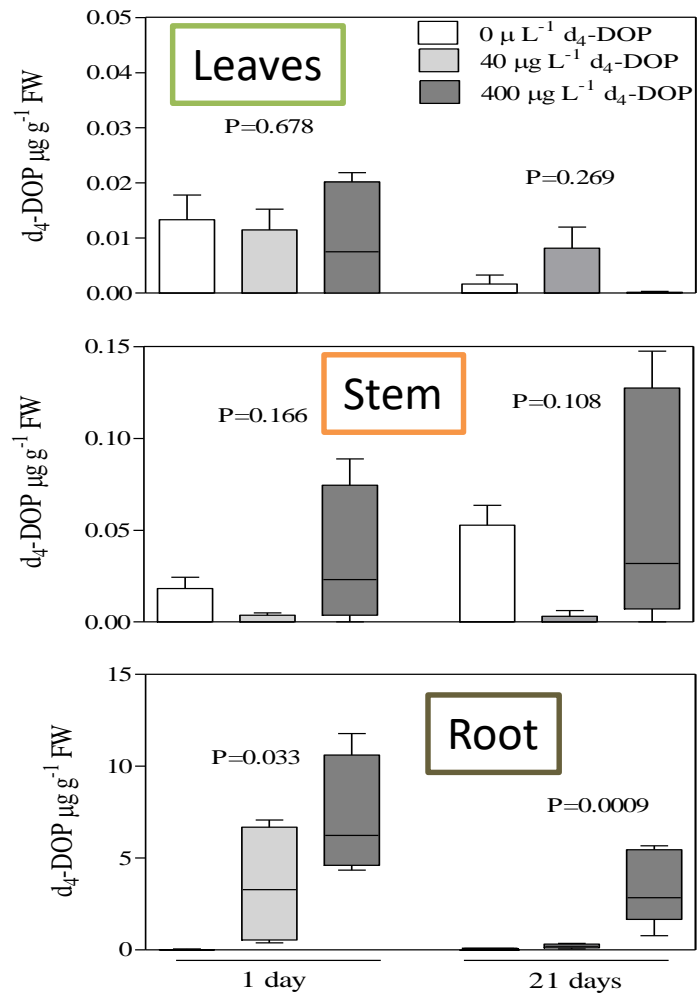
Application of **d₄-DOP** allows to differentiate such compound from one present in **laboratory environment**



Sampling
time:
1 and 21 days



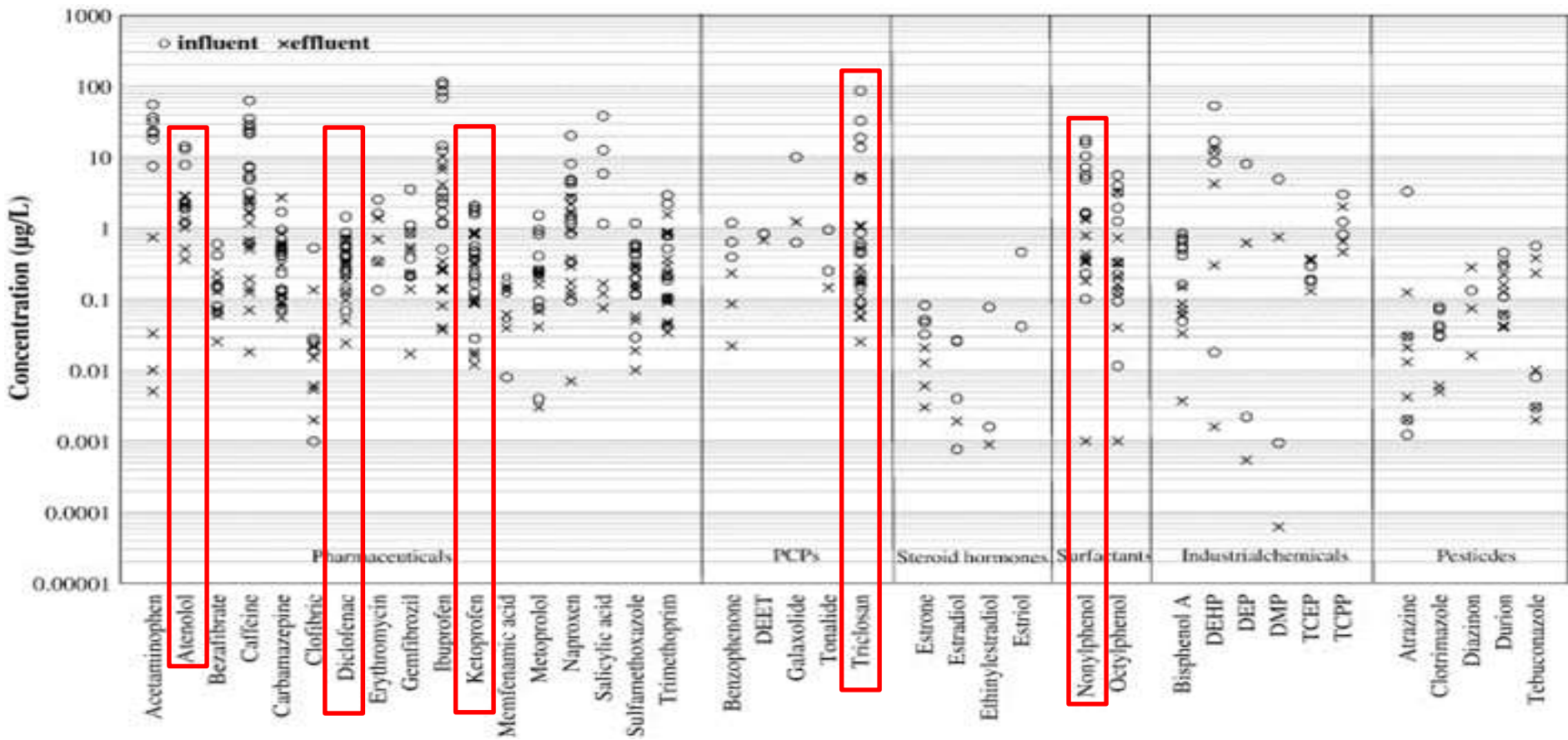


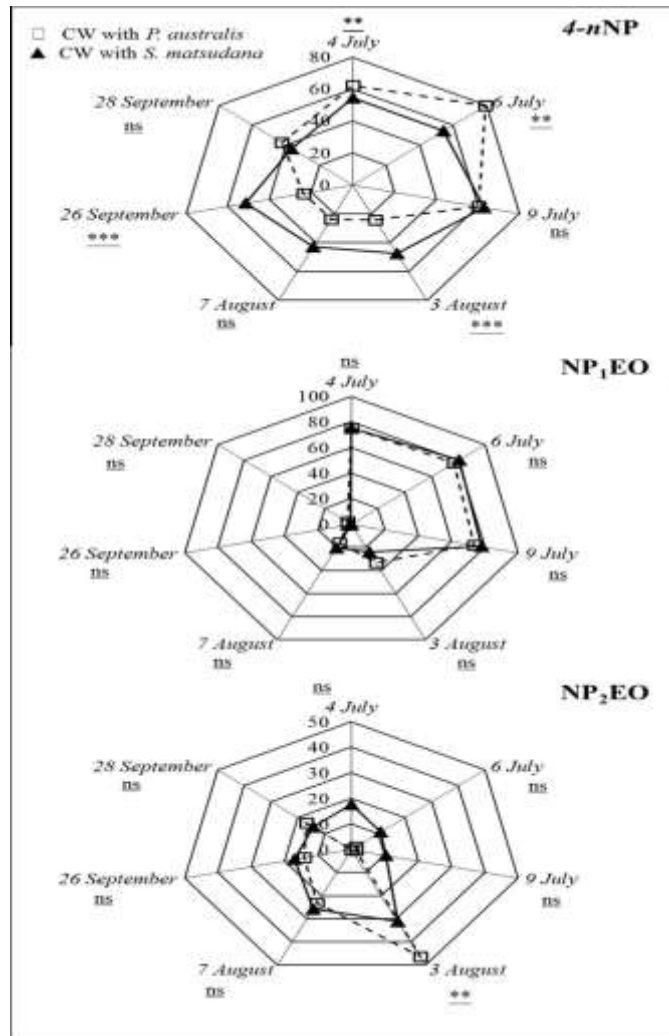
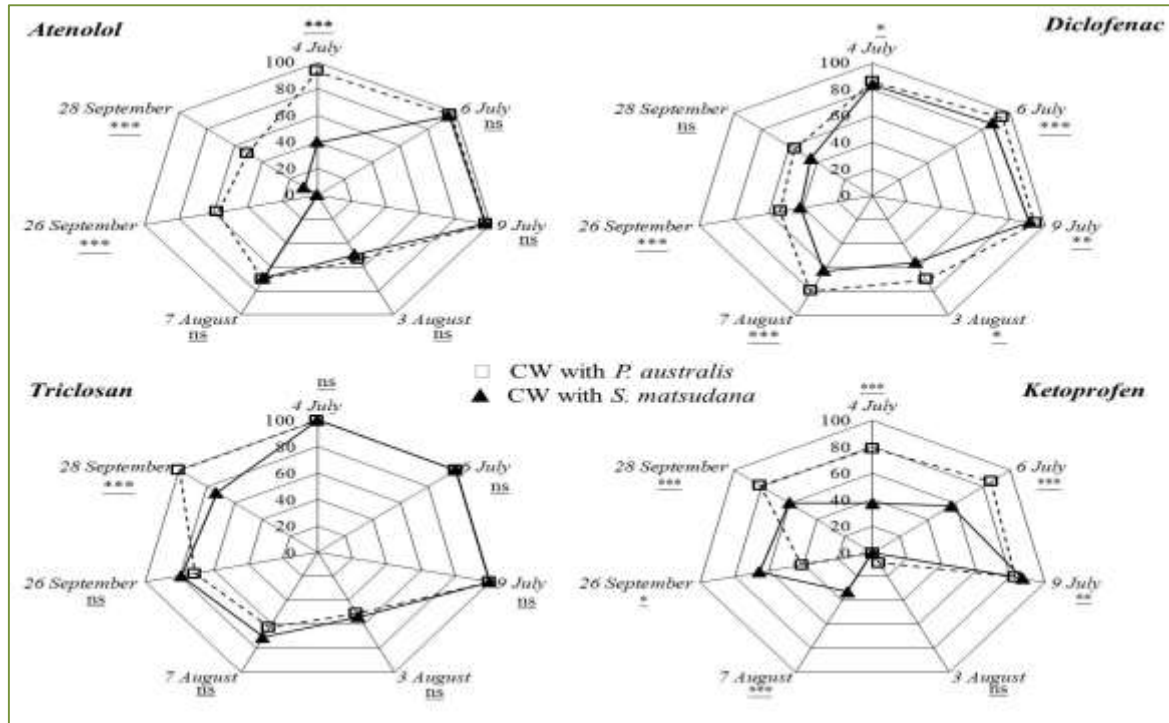


Funzionano ?



Removal of micro-pollutants from urban wastewater by constructed wetlands with *Phragmites australis* and *Salix matsudana*





Removal efficiency of micro-pollutants was evaluated by means of Eq. (1): Removal efficiency (%) = $(M_{inf} - M_{eff}) / M_{inf} \times 100$, where M_{inf} is the load of micro-pollutant in CW influent and M_{eff} is the load of micro-pollutant in CW systems effluent. Removal efficiencies were calculated assuming that no evapotranspiration took place since losses by evaporation in a CWs are considered negligible compared with the great volumes of water treated.