Development of Climate Smart Forestry (CSF) concept in the Republic of Serbia through mycorrhizal modulation of polyamine metabolism in pedunculate oak (*Quercus robur* L.) trees

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Crosstalk between climate change (CC) and foresty

Detrimetal effects of CC on forests

Climate change with a frequent heat waves and altered precipitation patterns increases abiotic (drought and heat stress etc) and biotic stress (pathogens, insects…) in trees.

CSF is "from science to policy" based concept that strives to implement new measurable, accurate parameters to track and monitor forests' adaptability to CC.

Beneficial effects of forest on mitigation of CC

"Climate smart forestry (**CSF**)" concept aims to reduce GHGs emission and to increase understanding of complex patterns of abiotic and biotic stress resilience of different forest tree species in order to build more adaptable forests to CC through practice of adaptive forest management.

CLIMATE SMART FORESTRY

“WOOD WIDE WEB”

Mycorrhizal fungi affects abiotic stress tolerance in plants

- Ectomycorrhiza (EcM) and arbuscular mycorrhiza fungi (AMF) are key players in carbon dynamics and carbon fluxes among plants, soil and the atmosphere
- They deliver water, nitrogen and phosphorus to the plant in exchange to photosynthetically produced carbohydrates from host trees
- Profuse hyphae system have 60 times more absorptive area than fine roots and higher efficacy in provision of inaccessible water

Hypothesis 1. to test

a) whether presence of mycorrhizal fungi affects oaks’ tolerance mechanisms to heat and drought

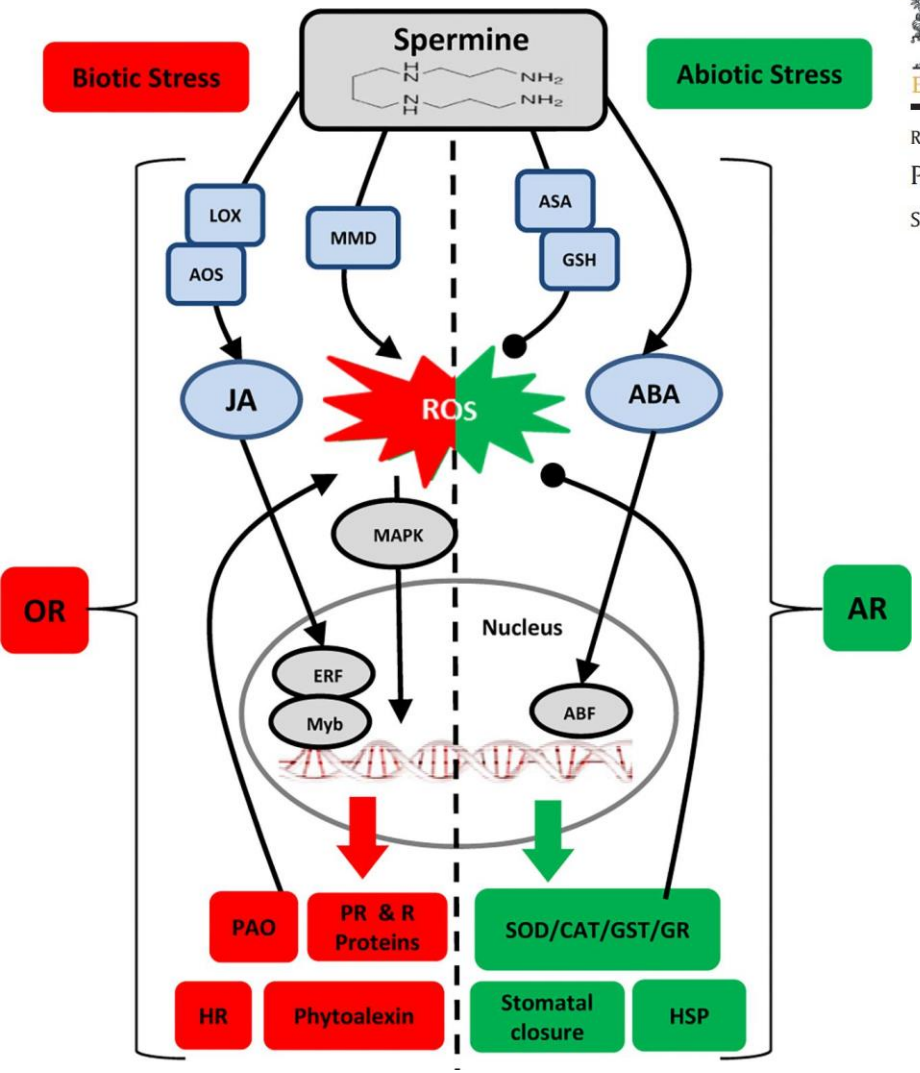
Mycorrhizal fungi alleviate biotic stress in plants

- Mycorrhizal fungi orchestrates all below- and aboveground multitrophic interations through modulation of VOCs
- Mycorrhiza modulate plant defense responses through molecular reprogramming
- MIR-mycorrhiza induced resistance
- Problem of powdery mildew disease triggered by *Erysiphe alphitoides Erysiphe quercicola and Erysiphe hypophylla*

Hypothesis 2. to test

b) whether mycorrhizal fungi modulates oak resistance mechanism against powdery mildew fungi

POLYAMINES (PAs) ALLEVIATE ABIOTIC AND BIOTIC STRESS IN PLANTS



from Seifi and Shelp, 2019. *Front. Plant Sci.,*

Biotechnology Advances 29 (2011) 300–311

Contents lists available at ScienceDirect

Biotechnology Advances

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

Research review paper

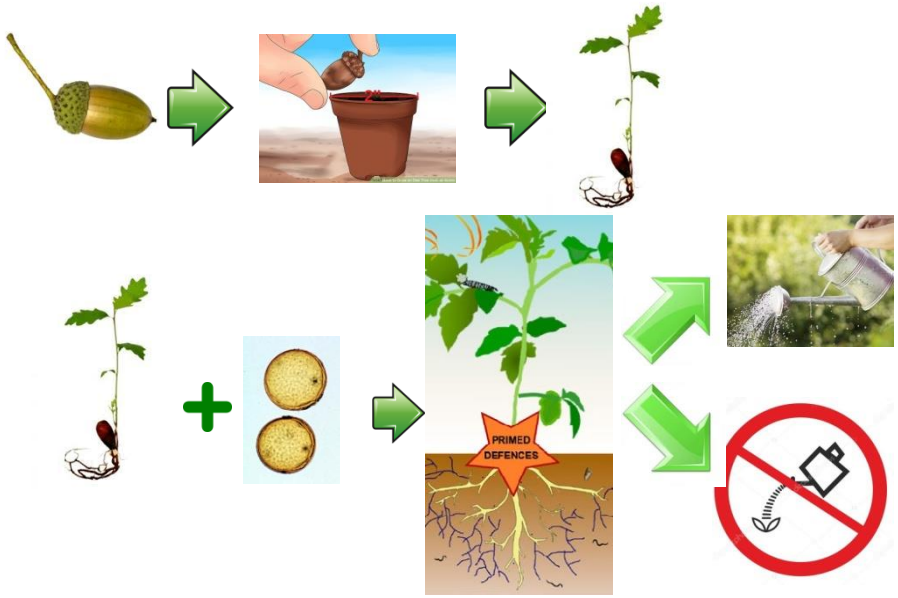
Polyamines: Natural and engineered abiotic and biotic stress tolerance in plants

Syed Sarfraz Hussain [a,*], Muhammad Ali [b], Maqbool Ahmad [c], Kadambot H.M. Siddique [d,e]

- Universal multifunctional regulators of physiological processes (DNK replication, transcription, translation, membrane stabilization and enzyme activity modulation, cell division and expansion, plant growth and development embryogenesis, leaf senescence etc)
- Low molecular-weight organic polycations displaying high biological activity
- The common PAs in plants are spermidine (Spd), spermine (Spm) and putrescine (Put).
- Modulate abiotic and biotic plant stress responses
- Regulation of oxidative homeostasis and phytohormone signaling
- PAs exhiibit antioxidant and prooxidative effects
- Osmoprotective and antimicrobial activity

Tripartite experiments: plant-mycorrhiza-environmental stresses or plant-mycorrhiza-pathogen

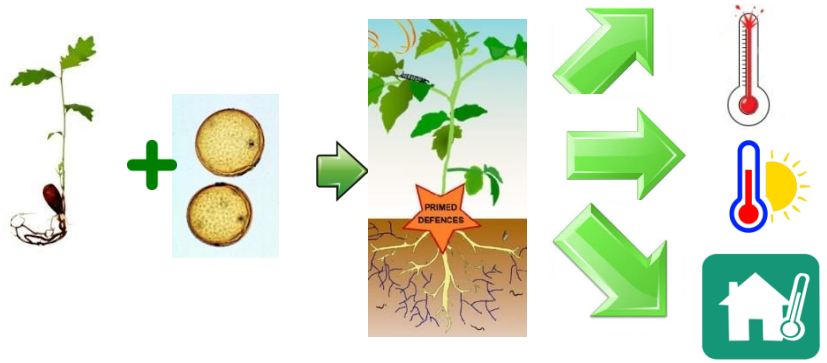
Experiment 1. Examination of mycorrhizal effects on biological response of oak seedling to drought



Mycorrhized plants			Non -mycorrhized plants		
Timely watered	Mild drought	Severe drought	Timely watered	Mild drought	Severe drought

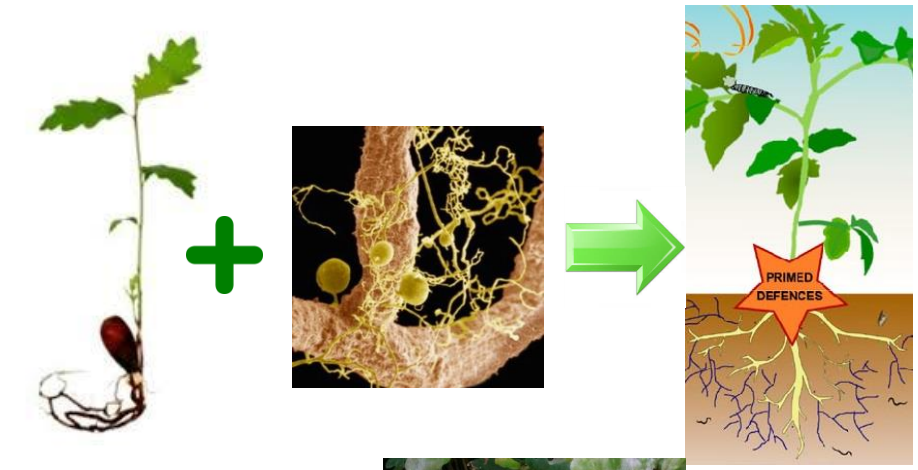
MILD DROUGHT => 50% FIELD WATER CAPACITY
SEVERE DROUGHT => <30% FIELD WATER CAPACITY

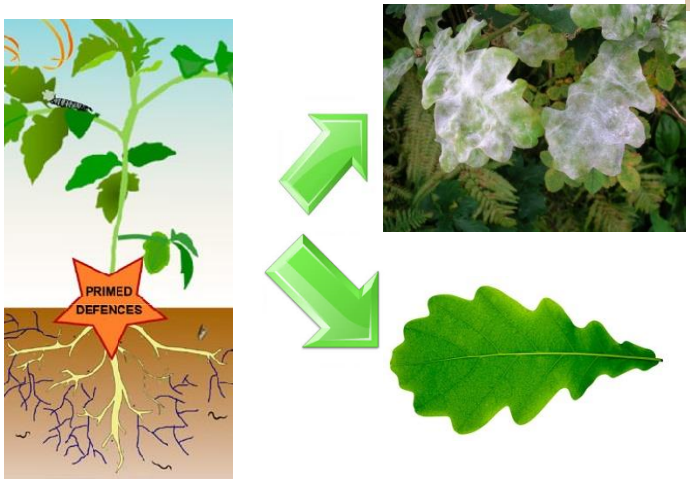
Experiment 2. Examination of mycorrhiza effects on biological response of oak seedling to temperatures



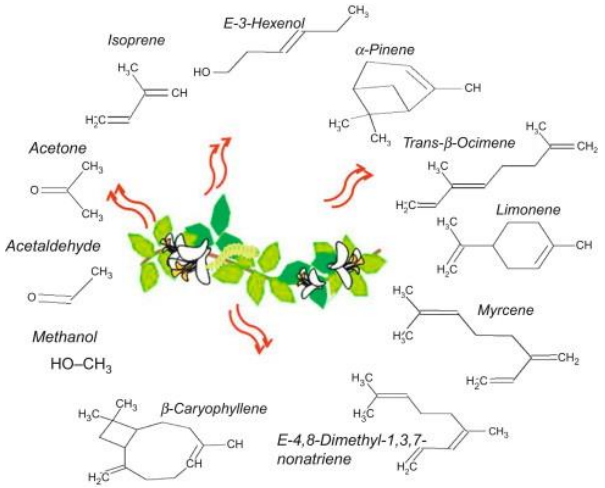
Mycorrhized plants			Non -mycorrhized plants		
RT	T1	T2	RT	T2	T3

Experiment 3. Powdery mildew in presence and absence of mycorrhiza

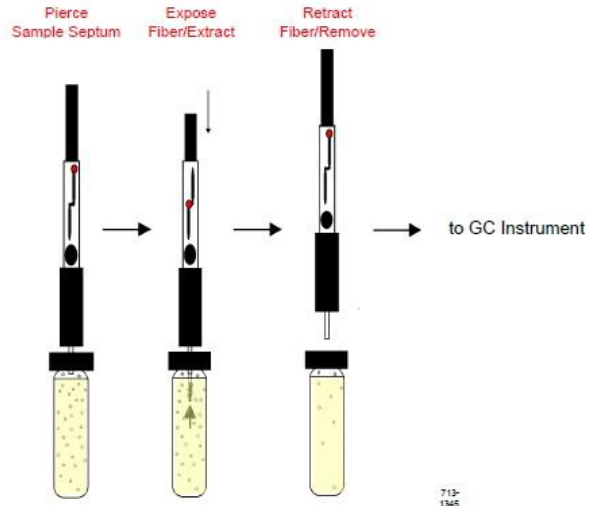




Mycorrhiza-induced VOCs modulation



Extraction Procedure for SPME



Mycorrhized plants		Non-mycorrhized plants	
No infection	Powdery mildew	No infection	Powdery mildew

MYCOCLIMART project objectives:

Specific objectives:

Objective 1.

To evaluate the effects of mycorrhyza on oaks' (*Quercus robur* L.) modulation of biochemical defense responses to abiotic stress factors (drought and heat) and its effects on PA metabolism

Objective 2.

To explore bioprotective properties of mycorrhyza in suppression of *Quercus robur* L. disease caused by powdery mildew (*Erysiphe alphitoides* (Griffon & Maublanc 1912)).

Objective 3.

Introduction of PAs as new reliable parameters in concept of CSF as markers of abiotic and biotic stress resilience in tress

General objectives:

- **to form a multidisciplinary research TEAM**
- **to foster know-how exchange among participants with different expertise**
- **to implement innovative techniques such as HPLC-DAD (high performance liquid chromatography coupled with diode array detection)**
- **to establish a cutting-edge laboratory that will be able to perform broad spectrum of metabolic, transcriptomic, enzymatic analysis regarding polyamine metabolism**

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Transcriptomic, metabolomic and spectrophotometric analysis

1) TRANSCRIPTOMIC ANALYSIS

ODC, ADC, DAO, PAO, CAT and PAL

2) METABOLIC ANALYSIS

a) TARGETED ANALYSIS of PAs HPLC/DAD

b) NON-TARGETED ANALYSIS OF VOCs by GC/MS

3) SPECTROPHOTOMETRICS

a) ENZYMES ACTIVITIES ODC, ADC, DAO, PAO, CAT, SOD and PAL

b) OSMOLYTES PROLINE AND GLYCINE BETAINE

4) PHYSIOLOGICAL MESURMENTS

a) Chlorophyll florescence

b) Gas exchange (Net Photosynthesis transpiration rate stomatal conductance)

PROJECT IMPACT AND DELIVERABLES

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- Establishment of the unique multidisciplinary scientific team.
- Strengthening of the young researchers capacities (boosting of their academic and laboratory skills)
- Improvement of the laboratory through introduction of new methodologies and equipment.
- Generation of new knowledge and launching research in new areas
- Fostering the development of the concept of Climate Smart forestry in the Republic of Serbia.
- Use of mycorrhizae in seedlings and nursery to improve reproductive material quality.
- Improvement of reforestation success under changing climate.
- Strengthening of management, supervision and coordination capabilities of PI.

Deliverables:

- Two publications in peer-review Internation journal
- Two publications in national journals
- “Step by step” guidelines for nursery production of mycorrhiza enriched seedlings
- Web page dedicated to nursery producers and nursery plant suppliers that will facilitate their communication with experts from science through this “forum based” web page

DISSEMINATION AND COMMUNICATION

a) Peer-reviewed international scientific journals
b) International scientific conferences
International Conference on Plant Physiology and Plant Science: **ICPPPS 2021**
International Conference on Plant protection, Crop Production and Field crops **ICPPCPFC 2021**
c) WORKHOPS at National Parks Fruška Gora and Kopaonik, the meeting with stakeholders
d) E-MEDIA (Word Press, Facebook, Researchgate etc)
e) Through teaching process at Faculty of Agricultuere
f) Promotional 10 min long video, Youtube