Are priming agents a step forward to enhance soft fruit yield efficiency?

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Outline

#1 Soft fruit production:
Introductory note

#2 The concept of priming as a novel agricultural approach
Prospects and future perspectives

#3 Why priming agents (PAs) in soft fruits?
PRIMESOFT project in a snapshot

#4 Experimental data on application of PAs

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Academic positions

Post-doctoral Research Associate
University of California at Davis
Department of Plant Sciences, USA
[01/07/06-31/12/06]

Post-doctoral Research Associate
Recipient of an MC-IEF Fellowship
University of Padova, Italy
[01/01/07- 30/04/08]

Cyprus University of Technology
Full professor
Associate Professor
Assistant Professor
Lecturer
[01/01/24- present]
[03/01/18- 31/12/23]
[01/01/13- 28/02/18]
[01/01/09- 31/12/12]

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The CUT Fruit Sciences Group

www.frutsciences.eu

- Objectives
- Members
- Projects
- Facilities
- Protocols
- Courses
- Conferences
- Publications

- ✅ To optimize production systems practices
- ✅ To incorporate postharvest applications in the supply chain of fresh produce
- ✅ To characterize and valorize indigenous cultivars
- ✅ To identify market opportunities for the horticultural sector
- ✅ To create interest, raise awareness among policy makers and attract funding

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Soft fruit production: Introductory note

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Soft fruits [small berries]: a very diverse group

Table 1. Nomenclature of small berry fruits

Common name	Scientific name
Fragaria genus (family: Roseaceae)	
Cultivated strawberry	*Fragaria* × *ananassa*
Chilean (coastal) strawberry	*Fragaria chiloensis*
Wild strawberry	*Fragaria virginiana*
Alpine strawberry	*Fragaria vesca*
Musk strawberry	*Fragaria moschata*
Ribes genus (family: Glossulariaceae)	
Black currant	*Ribes nigrum*
Red currant	*Ribes rubrum*
White currant	*Ribes glandulosum*
Gooseberry	*Ribes uva-crispa*
Rubus genus (family: Rosaceae)	
Blackberry	*Rubus fruticosus*
Black raspberry	*Rubus occidentalis*
Red raspberry	*Rubus idaeus*
Boysenberries	*Rubus ursinus* × *idaeus*
Cloudberries	*Rubus chamaemorus*
Loganberry	*Rubus loganobaccus*
Vaccinium genus	
Highbush blueberry	*Vaccinium corymbosum*
Lowbush blueberry	*Vaccinium angustifolium*
Rabbit eye blueberries	*Vaccinium virgatum*
Velvet leaf blueberry	*Vaccinium myrtilloides*
Bilberry	*Vaccinium myrtillus*
Cranberry	*Vaccinium macrocarpon*
Other berries	
Mulberry	*Morus alba, Morus nigra*
Bayberry	*Myrica rubra*

Review

Berry antioxidants: small fruits providing large benefits

George A Manganaris,[a*] Vlasios Goulas,[a] Ariel R Vicente[b,c] and Leon A Terry[d]

ELSEVIER ScienceDirect Food Chemistry

Antioxidant capacity, phenol, anthocyanin and ascorbic acid contents in raspberries, blackberries, red currants, gooseberries and Cornelian cherries

G.E. Pantelidis, M. Vasilakakis, G.A. Manganaris *, Gr. Diamantidis

Continuous growth of raspberry production

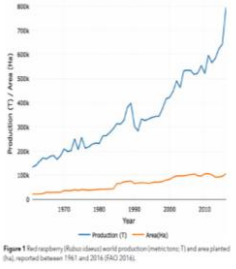

Figure 1 Red raspberry (Rubus idaeus) world production (metric tons; T) and area planted (ha), reported between 1961 and 2014 (FAO 2016).

Source: Manganaris et al. (2014)

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Washington Red Raspberry Commission Research Priorities 2023

#1 priorities
- Develop cultivars: summer bearing, high yielding, winter hardy, machine-harvestable, disease resistant, virus resistant and have superior processed fruit quality
- Control of the spotted wing drosophila, mites and foliar & cane diseases
- Labor saving practices

#2 priorities
- Fruit rot including pre harvest, post-harvest, and/or shelf life
- Understanding soil ecology (including biology, nutrient balance) and soil borne pathogens
- Soil fumigation techniques to control soil pathogens, nematodes, and weeds

#3 priorities
- alternative management systems – fruit yield– planting densities, row spacing, trellising
- season extension: improve viability of fresh marketing
- nutrient and irrigation management

Source: https://www.red-raspberry.org/research

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Raspberry cultivation in Europe: the case of Greece

Total cultivated area: 20 ha
Farm gate price: 7.0-8.0 €/kg - fruits for fresh consumption

Primocane cultivars (Kwanza, Adelitta) / soiless production (cocopeat)
1st production: Fall (October-December)
2nd production: Spring (April-May)

Ability to produce year round through a combination of cultivars and techniques with a gap during July and August

Source: Tsormpatsidis

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Strawberry versus Raspberry: a crash test

- Plant material cost: 0.18-0.27€/plant
- Labor efficiency: 150 kg/ working day
- Annual production cost: *ca.* 50,000€/ hectare

- Plant material cost: 1,0 €/plant
- Labor efficiency: 20 kg/ working day (60% of total cost)
- Annual production cost: *ca.* 125,000€/ hectare

Soft fruits are a capital- and knowledge-intensive cultivations

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Strawberry cultivation: the case of Greece

Fortuna

- Total Production: 1 kg/plant
- Ripening period: November-April
- Plant material: 0.3€/tray plant

- Early production: 0,15 kg (4-fold ↑ price)
- Excellent taste
- High sensitivity to *Phytophthora*
- Skilled farmer

Victory

- Total Production: 0.85 kg/plant
- Ripening period: February-May
- Plant material: 0.18€/bare root plant

- Excellent postharvest performance
- No claims
- Inferior taste compared to 'Fortuna'

key factors: breeding and capacity to optimize production protocols

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Strawberry cultivation: 'Fortuna' case study

- Annual production cost: *ca.* 50,000€/ hectare
  65,000 plants per hectare
- Farm gate price (mid November – early January): 4,0€
  marketable yield per plant: 150 g
- Farm gate price (mid January – early May): 1,4€
  marketable yield per plant: 850 g

- Early production: 0,6€
- Mid-late production: 1,2€
- Net profit: 67,000 €/ hectare

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The concept of priming: a novel agricultural approach

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Types of stress conditions

Abiotic stress (salinity, drought, heavy metals etc.)
Biotic stress (pathogenic microorganisms)

The plant receives multiple environmental stimuli

⬇

Several metabolic pathways are 'switched on' in response to accumulation of signaling molecules

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Solutions to combat stress conditions

- Genetic modification
  CRISPR-Cas movement: Targeted Genome Editing Technology
- Selection of tolerant cultivars
  Conventional breeding
- Plant priming

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Defining the term 'priming'

- The process of priming involves prior exposure to a biotic or abiotic stress factor making a plant more resistant/tolerant to future exposure.
- Priming can also be achieved by applying natural or synthetic compounds which act as signaling transducers, 'activating' the plant's defense system.
- Exposure to a stimulus allows a plant to respond in a more rapid and effective way to a later stimulus (the same or equivalent) compared with a non-primed plant.

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Classes of priming agents

Chemicals (including natural and synthetic molecules)
- Hormones (i.e. salicylic acid, jasmonic acid, strigolactones)
- Reactive Oxygen Nitrogen and Sulphur Species (RONSS: NO, H_2S, H_2O_2)
- Small organic molecules (i.e. melatonin, putrescine)

Microorganisms
- Arbuscular mycorrhizal fungi (AMF, the most types: *Funneliformis mosseae, Rhizophagus irregularis*)
- Plant growth-promoting bacteria (PGPR, soil bacteria living in the rhizosphere that are involved in promoting plant growth and development)

Nanomaterials
- Organic nanoparticles
- Inorganic nanoparticles
- Polymers

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Classes of priming agents: a meta-analysis report

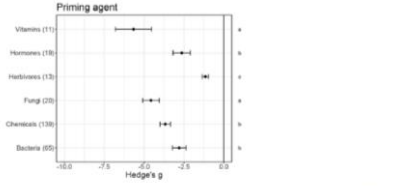


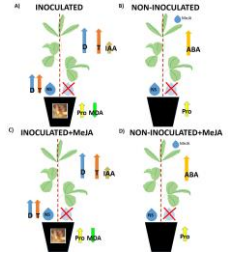
Figure 1. Resistance effects of priming Arabidopsis plants with indicated agents. Results of meta-analysis of data obtained from 267 experiments described in 77 publications. Negative values imply that primed plants were more resistant (less damaged or associated with lower pest fitness) than unprimed controls. Numbers of experiments are shown in brackets, and symbols specify means of Hedge's g ± SE bars, equivalent to effects of groups of priming agents (Vitamins, Hormones, etc.). Different letters along the right-hand axis indicate significant differences according to the Kruskal Wallis test ($\alpha = 0.05$) followed by Dunn's post-hoc test to rank differences ($\alpha = 0.05$).

Source: Westman et al. (2019) Sci Rep 9, 13309

Mixing it up

Combination of chemical and biological priming

Arbuscular mycorrhizal fungal inoculum + MeJa priming



Source: Irankhah et al. (2020) *Env Exp Bot* 176, 104096

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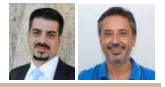
The effect of PAs on strawberry plants



Plant priming with signaling molecules (H_2O_2, NO, H_2S): a promising approach for alleviating abiotic stress devastating effects

Pretreatment was carried out in a hydroponic cultivation system and plants exposed to a multitude of abiotic stress factors and analyzed through a combinatorial physiological, biochemical and molecular approach

- **Plants (strawberry) pretreatment with H_2O_2 and NO effectively alleviated oxidative stress induced by salt exposure through redox homeostasis and induced antioxidant activity**
- **H_2S pretreatment enhanced osmotic and non-ionic osmotic stress tolerance, as well as thermotolerance of strawberry plants through the systemic activation of tolerance mechanisms (redox homeostasis, ROS pathway, heat shock proteins)**

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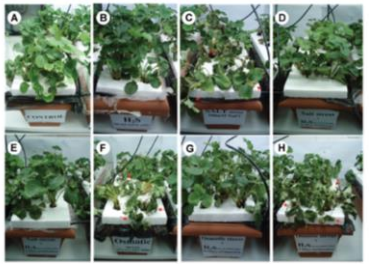
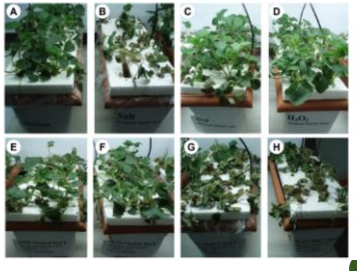
The effect of PAs on strawberry plants


Hydrogen sulfide induces systemic tolerance to salinity and non-ionic osmotic stress in strawberry plants through modification of reactive species biosynthesis and transcriptional regulation of multiple defence pathways




Environmental and Experimental Botany

Systemic mitigation of salt stress by hydrogen peroxide and sodium nitroprusside in strawberry plants via transcriptional regulation of enzymatic and non-enzymatic antioxidants



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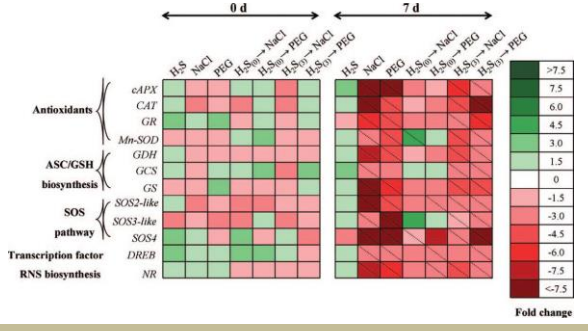
Exploring the mechanistic action of PAs



Defence-related genes are highly-expressed in primed plants

Source: Christou et al. (2013), *J Exp Bot 64*, 1953-1966

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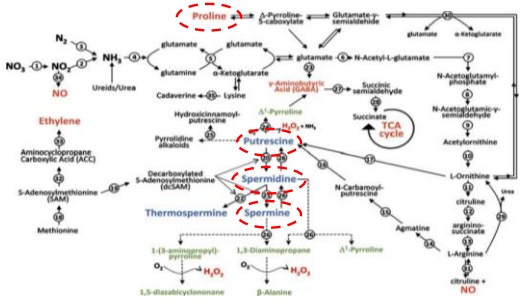
Polyamines: putrescine, spermidine, spermine

- Low molecular weight N-containing compounds
- Protective role to stress conditions
- Proline: a precursor molecule of poyamines with osmoprotective effect



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Melatonin

- The production and secretion of the hormone melatonin is linked with the light-dark cycle
- A naturally occurring hormone in plants, animals, and humans, has gained significant attention
- enhance biomass production and resistance to water stress

Source: Antoniou et al. (2017), *J. Pineal Res. 62, e12401*

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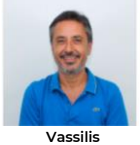
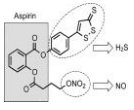
A new kid on the block

- NOSH and NOSH-aspirin
- Developed as a potent anticancer drug
- Hybrid donor of H_2S, NO and aspirin
- No more rotten egg smell!!!



Vassilis Fotopoulos



Method of Priming Plants Against Abiotic Stress Factors and Promoting Growth (Pub. No. WO/2015/123273).

NOSH: to promote growth
NOSH-aspirin: to ameliorate response to abiotic stress conditions

Source: Fotopoulos

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Advanced nanomaterials

another way to the future?

The concept: The application of nanomaterials as a means to enhance the efficiency of the priming agent (use as a carrier of the priming agent)

- Application of TiO_2 nanoparticles as plant growth promoters and stress protectors
- Engineered melatonin/chitosan conjugates
- Chitosan-Putrescine nanoparticles (CTS-Put NPs)
- Nanofertilisation with chitosan-selenium nanoparticles

Source: Ioannou et al. (2020), *Env Exp Bot 176*, 104048

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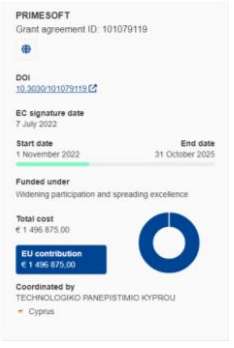
PRIMESOFT project in a snapshot

Development of innovative priming technologies safeguarding yield security in soft fruit crops through a cutting-edge technological approach

www.prime-soft.eu



Cyprus University of Technology

National Technical University of Athens

KATHOLIEKE UNIVERSITEIT LEUVEN

CSIC

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Overarching objective

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The increased demand of more sustainable and environmentally-friendly practices on soft fruit cultivations: evaluation of PAs from a range of perspectives

Eco-efficiency approaches
- Assessment of cultivation protocols (G x E effect)
- Life Cycle Cost Analysis
- Product development (i.e. encapsulation of PA and/or nanomaterial engineering)

-Omic tools
- Global transcriptomic analysis (RNAseq)
- Metabolomic analysis (Volatile organic compounds and phytochemicalS)

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Our experimental approach on application of PAs on soft fruits

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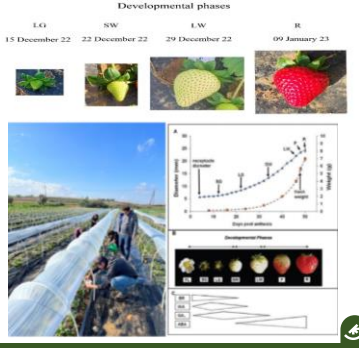
Working hypothesis
Pre-harvest application of priming agents on strawberry undergoing ripening processes on-vine under non-stressful conditions will result in enhanced qualitative and phytochemical properties

Priming agent treatments

- water-sprayed [Control]
- sodium alginate (0.5%)
- melatonin (100 μM)
- sodium alginate/melatonin (100 μM)
- putrescine (1 mM)

Sodium alginate: a biodegradable polymer applied in nano smart delivery systems



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Phytochemical analysis

Phenolics

Ascorbic acid

Anthocyanins

Flavonoids

Sugars

T.S.S.

Sucrose

Glucose

Fructose

Flavan-3-ols

Hydroxycinnamic acids

Ellagic acid and conjugates

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Working hypothesis

The potential use of bio-based carriers as a postharvest treatment to enhance strawberry fruit performance

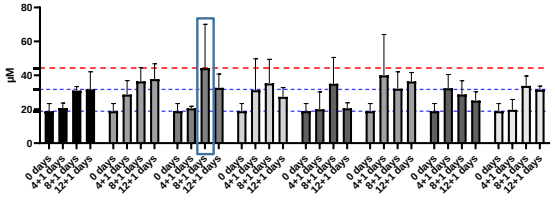
	Treatments
1	Untreated
2	Hydroprimed
3	NOSH-A
4	CTS
5	CTS-NOSH-A
6	Alg
7	Alg-NOSH-A
8	CTS-Alg-NOSH-A

Days
0
4+1
8+1
12+1

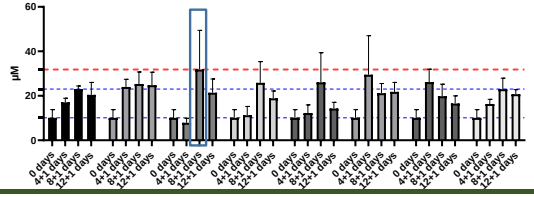
30

Ellagitannins

Ellagic acid



Ellagic acid rhamnoside



- Untreated
- Hydroprimed
- NOSH-A
- CTS
- CTS-NOSH-A
- Alg
- Alg-NOSH-A
- CTS-Alg-NOSH-A

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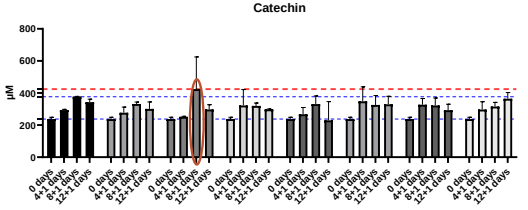
Catechin



Prof. M.I Gil

Prof. F.A.T.B

Garcia CJ, PhD

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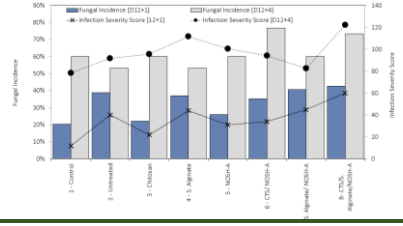
Priming agent efficacy towards biotic stressors?



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Working hypothesis

Priming to combat abiotic stress by simulation of control, moderate and severe conditions, such as salinity (0, 50 and 75 mM NaCl) and drought (regulated deficit irrigation (RDI) (0, 25 and 50%)

1. **Untreated**
2. **Water spray: water + tween 20 (0.1% v/v)**
3. **Sodium Alginate: 0.1% w/v)**
4. **Melatonin: 100μM + tween 20 (0.1% v/v)**
5. **Mel@Alg: 100μM + tween 20 (0.1% v/v)**
6. **Proline: 1mM + tween 20 (0.1% v/v)**
7. **Pro@Alg: 1mM + tween 20 (0.1% v/v)**

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Priming strategy to combat abiotic stress conditions

Plant material: potted strawberry plants (Cultivar 10-75)
Experimental Duration: Mid-October – End of December
Experimental Design: Randomized complete block design

Application timings:
- ✓ **First Application: Before Planting 14/10 (Soil)**
- ✓ **Second Application: 24/10 (Foliar)**
- ✓ **Third Application: 31/10 (Foliar)**

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Seed priming experiment

Aim of the study: Evaluation of strawberry seed priming on germination, seedling establishment and yield, under control and abiotic stress conditions.

Working Hypothesis: Seed priming is an effective approach, leading to fast and uniform germination, improved seedling vigor, and also increased crop yield and stress tolerance under abiotic stressors.

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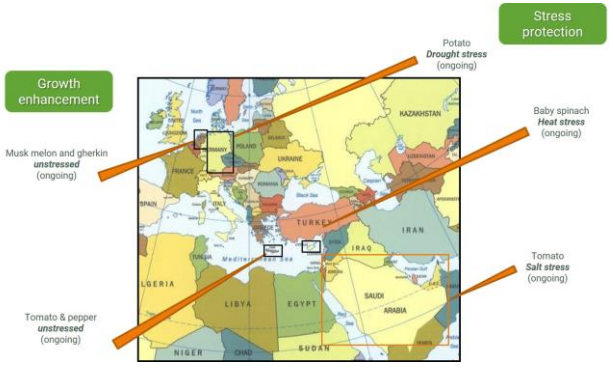
Prototype Field Trials in Process for TRL 7



6 crops, 5 countries, 3 climates, 4 conditions

PRIMESOFT International Workshop

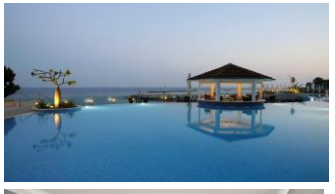
primesoft

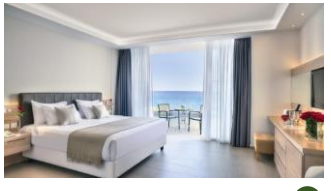
Application of cutting-edge technological approaches on value-added soft fruit crops

Save the dates (4-5 November 2024)

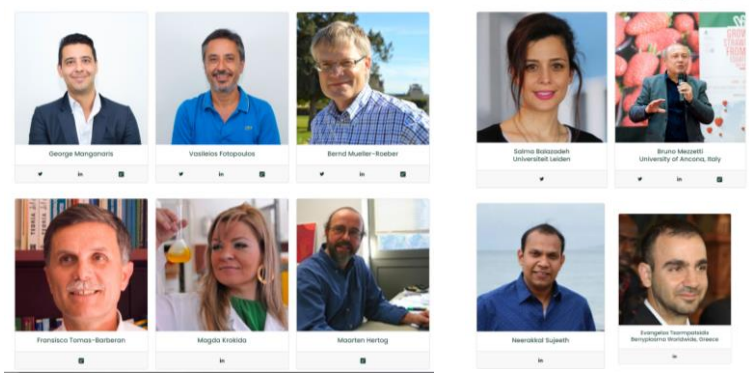
primesoft

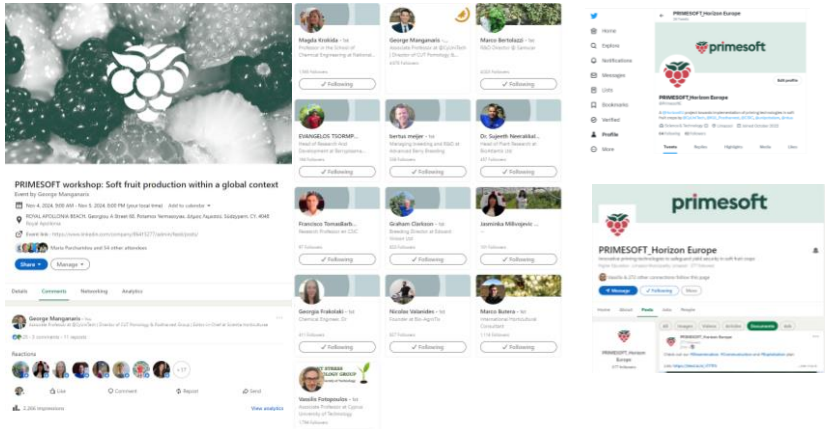
- Closed-type workshop (80 slots) on priming agents and soft fruits
- Registration-free (includes participant package, meals, coffee break)
- Apply through email (letter of interest)
- City: Lemesos, Cyprus
- Venue: Apollonia Royal Palace, Cyprus





Speakers





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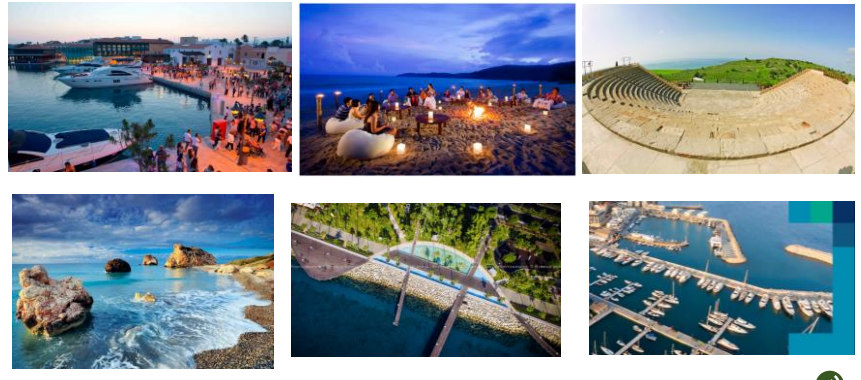
Lemesos at a glance



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The employment of priming agents as elicitors towards enhanced performance of soft fruits

Looking for partners

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Any questions?

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SCAN ME

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