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Innovative approaches to increase biodiversity in forest plant production

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Environmental
technologies





Artificial regeneration

- **Enhances biodiversity of species** (seed-derived seedlings)
- **Among species** (regenerate increased number of forest species)
- **Within the ecosystems** (Species that occur at the regenerated sites, suitable, based on the climatic and edaphic conditions of the area).



Challenges in forest plant production :

- Increased biodiversity from the increased number of diverse forest species for regeneration
- A more cost efficient way to produce seedlings to serve in regeneration purposes
- Increased demand for high quantity of produced seedlings
- Improved quality of the produced seedlings to enhance regeneration success
- The need to restore forest ecosystems, when possible.

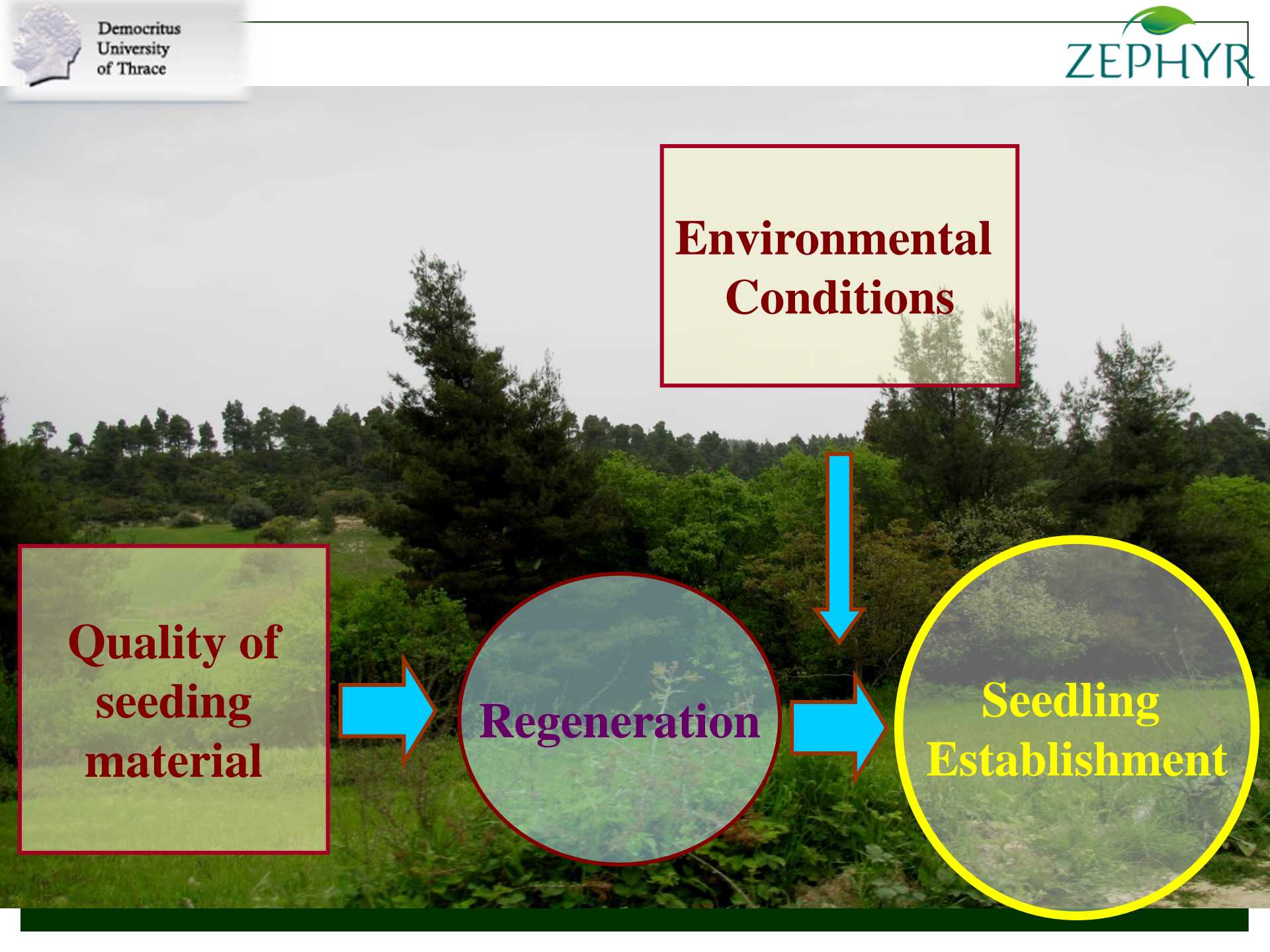
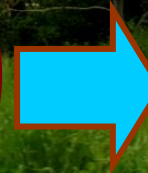


**Environmental
Conditions**

**Quality of
seeding
material**

Regeneration

**Seedling
Establishment**





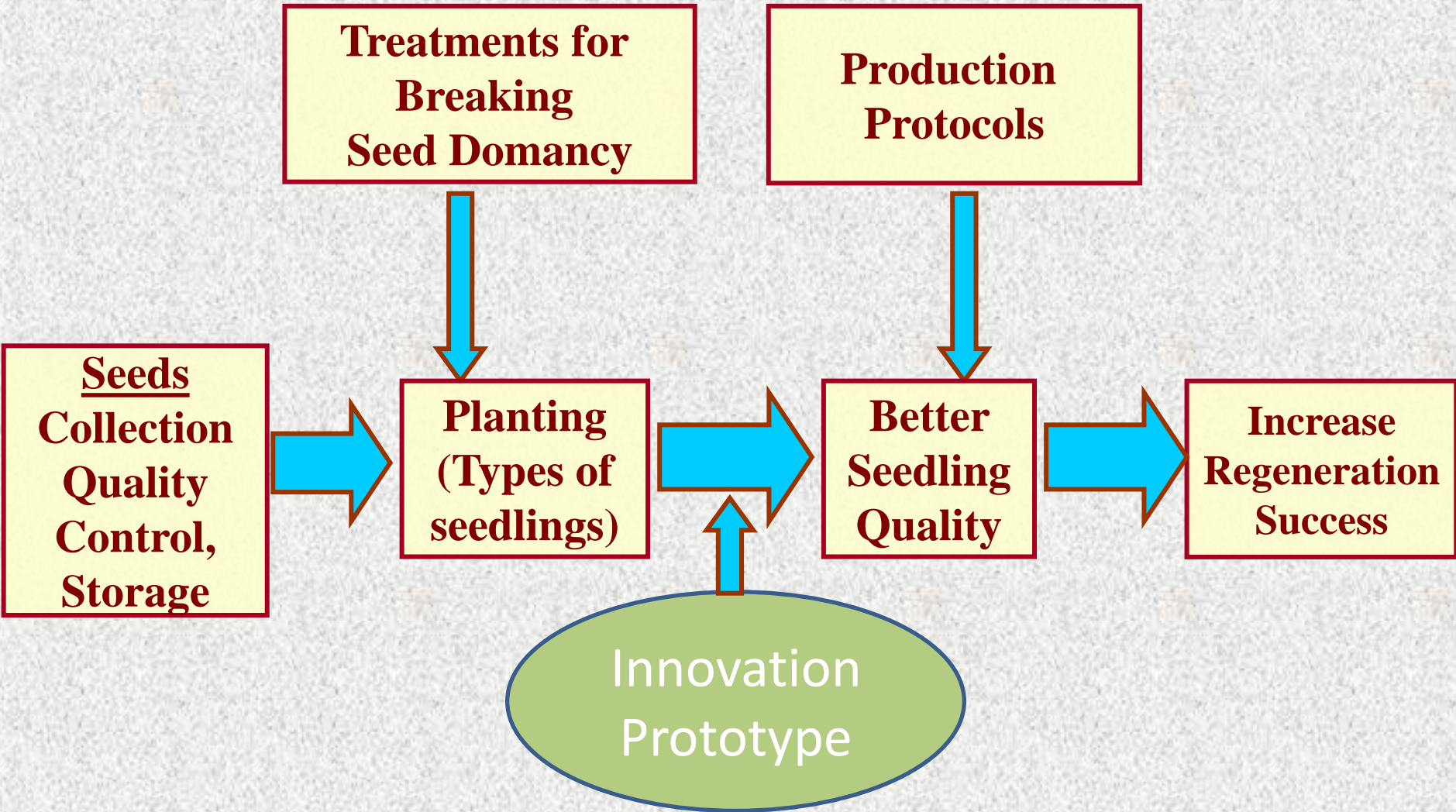
Needs for increasing biodiversity at regenerated sites

- Collection-quality & quantity of seeds
- Seed treatment protocols
- Production of high quality seedlings for many species
- Different species (Seedling production protocols)
- Higher % survival at the regenerated sites



Use innovative technology

- Cost efficient and environmental friendly production
- not affected by outdoors climate (controlled conditions)
- LED Labs
- New robot arm
- Integration of the technology into a functional prototype
- Provide cultivation protocols for the prototype





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Seed Collection



**Mediterranean
forest species**

Prior to Collection



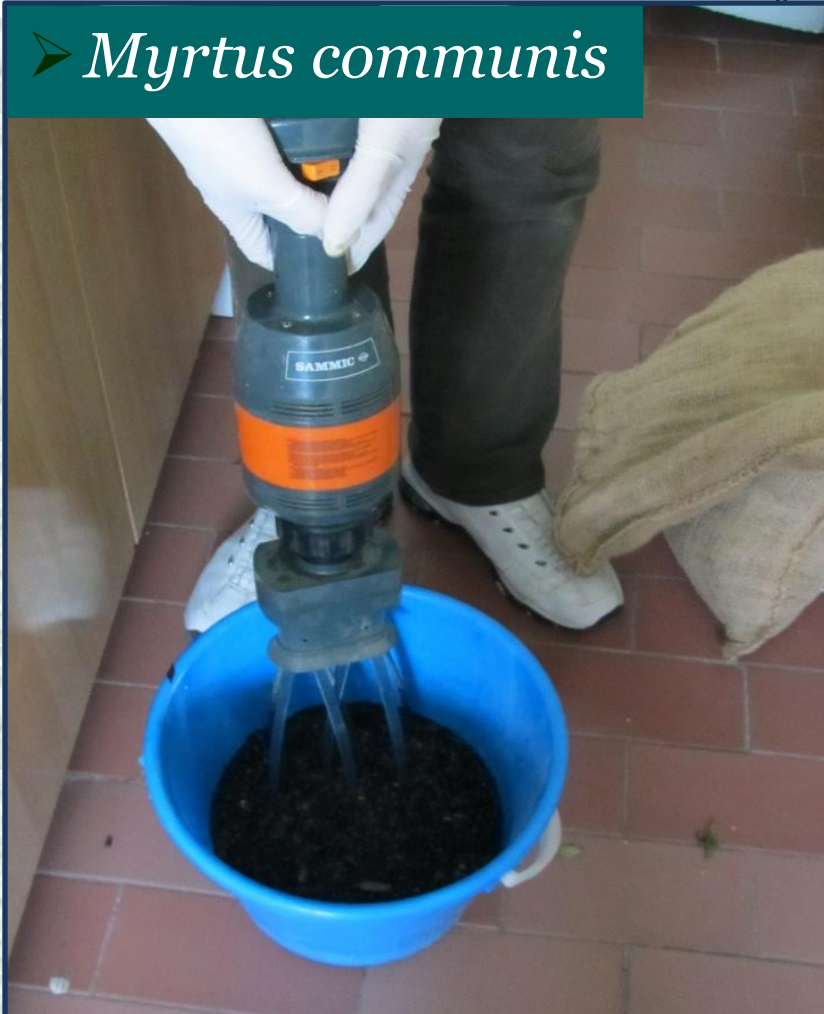
Criteria of tree selection

Criteria of tree selection	
1	Vigorous trees
2	Open crown trees
3	Middle age healthy trees
4	Collection from low crown part
5	Avoid isolated trees
Criteria of seed selection	
	Color of the fruit

Data sheet

DATE	SPECIES	PARENT	AREA	COLLECTOR	POSITION OF PLANT (GPS)	ALTITUDE (m)	CROWN WIDTH (m)	PLANT HEIGHT (m)	CROWN AREA OF SELECTION	SEEDS COLLECTED (kg)
EXAMPLE BELOW										
5/11/2010	Acer pseudoplatanus	1	X	X	465361, 4568095	1477	2	20	Whole crown	5
	Acer pseudoplatanus	2	X	X	X	1400	3	25	Base of crown	3

➤ *Myrtus communis*



➤ *Rosa canina*



Cleaning the seeds based on the species



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Seed Tests recommended by ISTA

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Clean seeds



Purity test



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Seed Tests

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Viability test



Celtis australis live



Moisture content and seed weight



Placement in the oven
 103 ± 2 °C for 17 ± 1 hrs

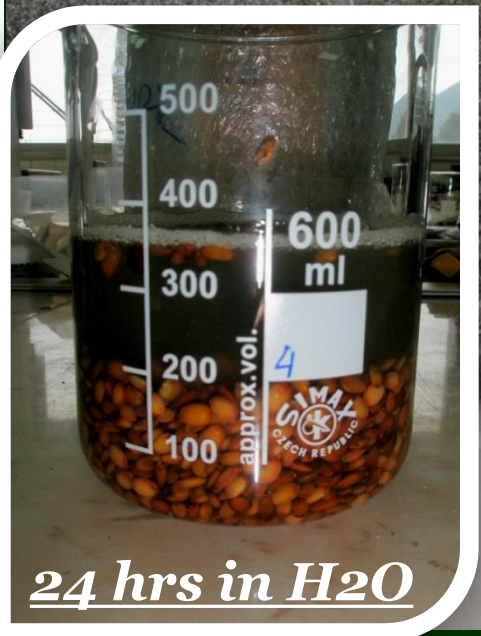
A close-up photograph of several small, young plant seedlings growing out of a dark, granular substrate in a clear petri dish. The seedlings have thin, white, curved stems and small, yellowish-green cotyledons. In the background, a white label with some illegible text is visible, and other similar petri dishes are partially seen.

➤ **Development of seed germination protocols of the 26 species**

Breaking dormancy-Protocols



Germination protocols



24 hrs in H₂O



Germination protocols suggested for each of the tested species

- **Hydration (24h in the water)**
- *Pinus nigra* Arn.:
- *Platanus orientalis* L.:
- *Pinus sylvestris* L.:
- *Picea abies* Karst.:
- *Quercus ilex* L.:
- *Quercus ithaburensis* var. *macrolepis*:
- *Ocimum basilicum* :
- **Cold stratification**
- *Arbutus unedo* L.: 1 month
- *Fraxinus excelsior* L.: 3 months
- *Myrtus communis* L.: 3 months
- *Cornus sanguinea* L. : 9 months
- *Punica granatum* L.: 2 months
- **Warm stratification (20 C)**
- *Prunus avium* L.:
- Still working on trts



Castanea sativa

- a) removing bur-like involucre that consist of numerous elongate, slender spines,
- b) hydration of seeded nuts,
- c) use polyethylene bags with several holes and plastic containers,
- d) stratification in wet sand for months in refrigerator at 3-5°C.

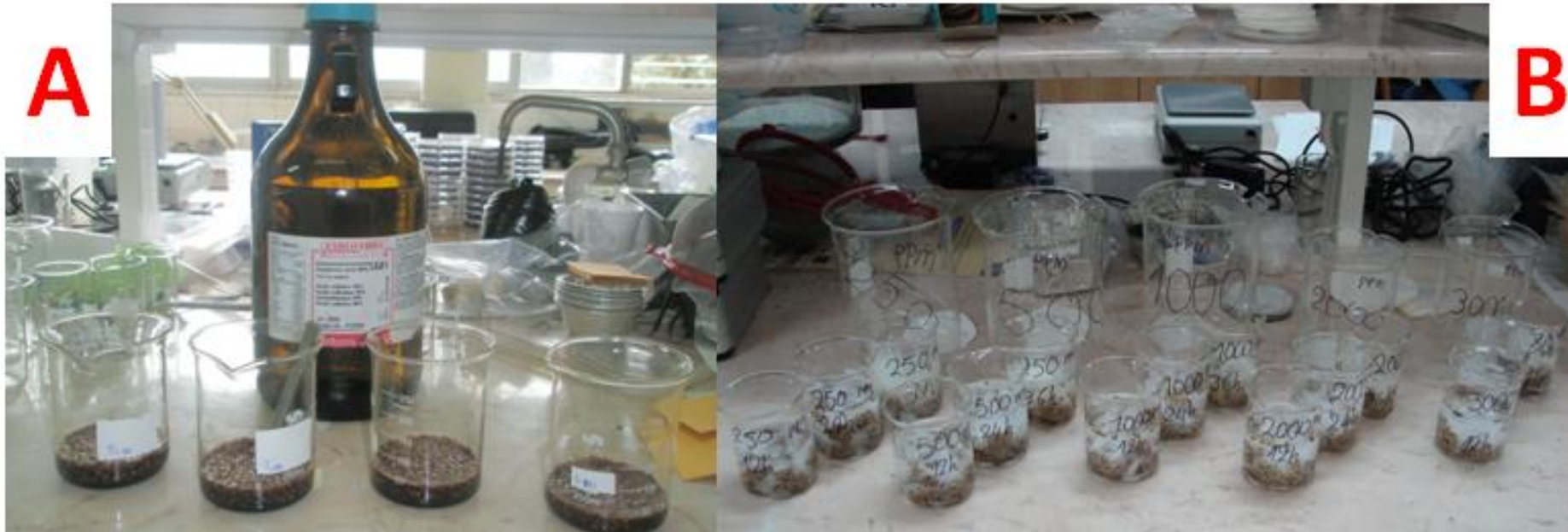


Quercus ithaburensis var. macrolepis

- a) acorns,
- b) removal of pericarp,
- c) cut of 1/3 of cotyledons,
- d) pre-germinated acorn



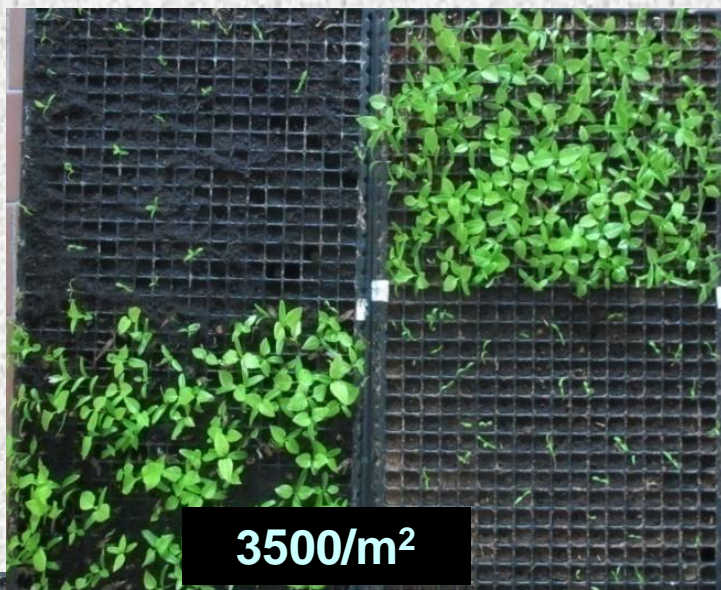
Chemical treatments



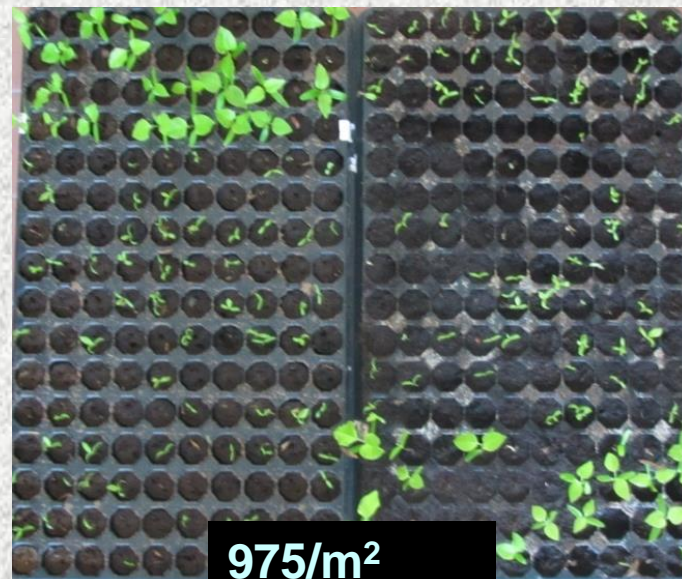
- Seed treatment of *Cercis siliquastrum* seeds with H_2SO_4 (A) and *Phillyrea latifolia* with GA_3 (B).

New way: Mini-plugs

Production of large seedling quantities through the use of «mini-plugs».



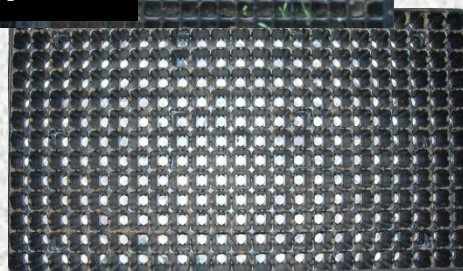
3500/m²



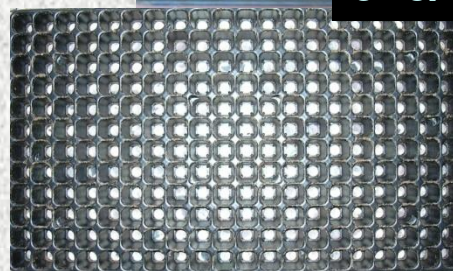
975/m²



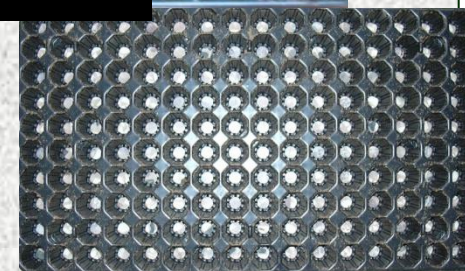
3500/m²



1820/m²



1460/m²



975/m²



QPD104 RW

cell dimensions: 33X33X45 mm
distance between: 40 mm
volume: 27 cc.



(QuickPot®, Herkuplast-Kubern GmbH, Ering, Germany)

Soil substrate: stabilized medium (SM)

ph: 3.7-4.3 rich in Br

(Preforma PP01, Jiffy Products International AS, Stange, Norway).





LED Lamps



metal halide



fluorescent (FL)



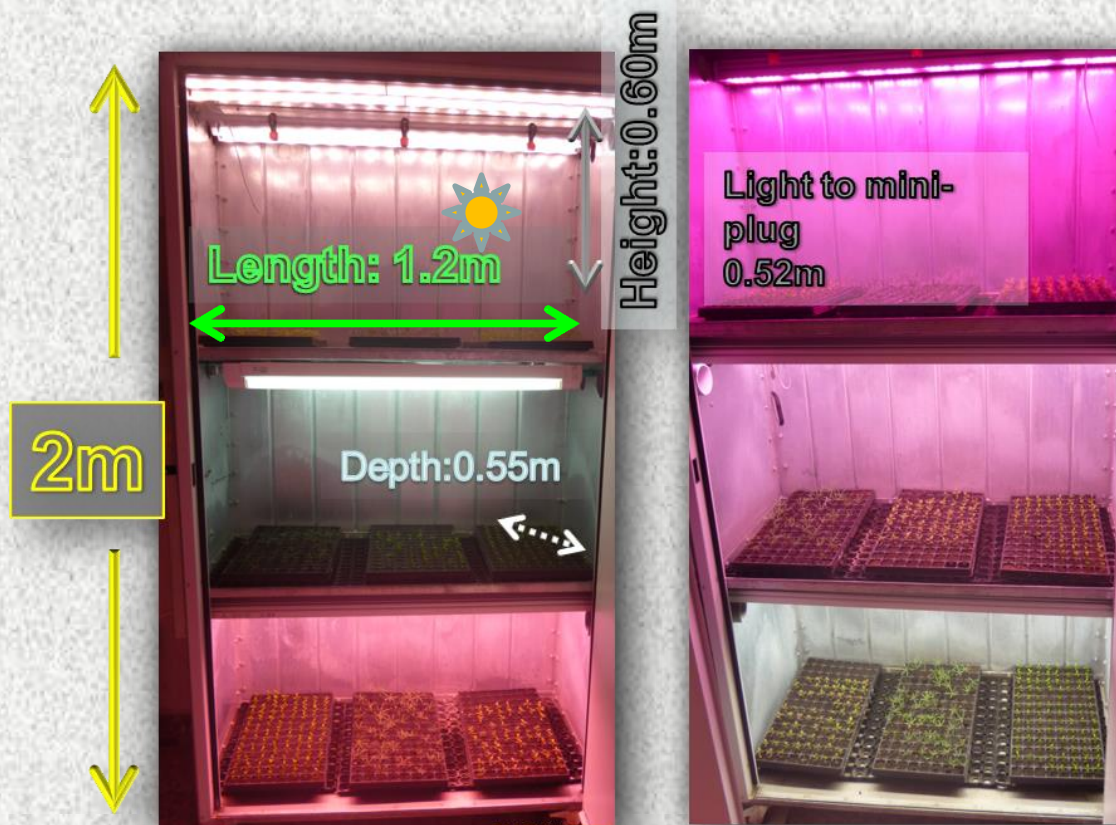
high-pressure
sodium

(LEDs)





Facilities and Growth conditions



✓ Automated watering

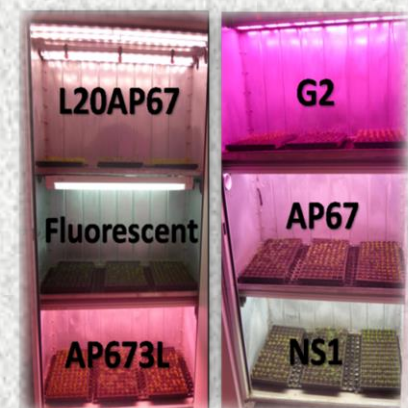
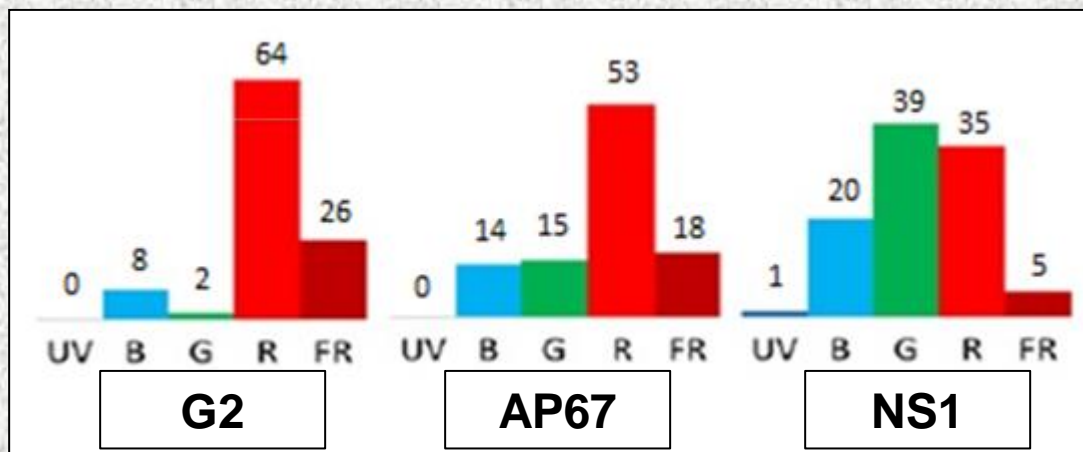
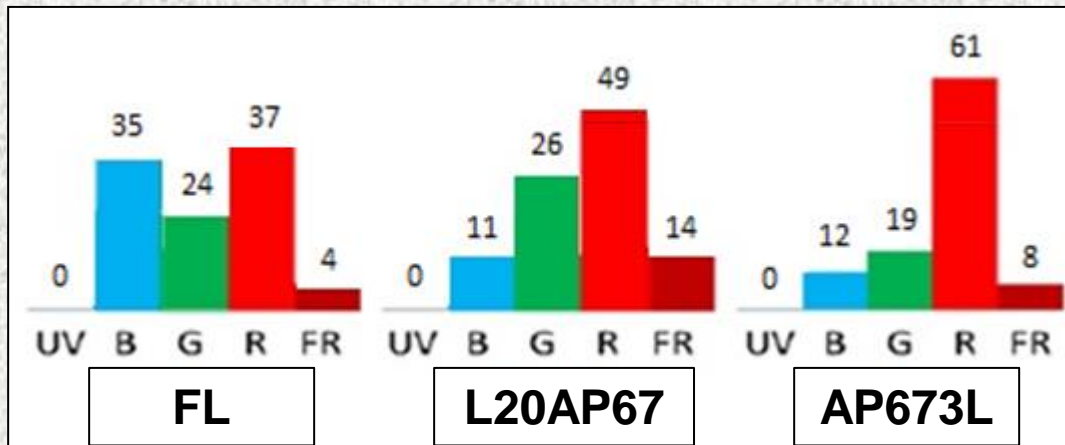
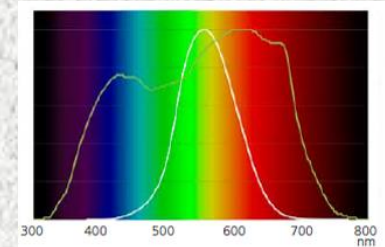
✓ Automated lights

- **Diurnal cycle** : 14 hrs day/8 hrs night of 20/15 °C
- **PPFD** : $140 \mu\text{mol m}^{-2} \text{s}^{-1}$
- **Relative humidity (RH)** : $80 \pm 10\%$
- **Watering** : Twice a day with automatic sprinklers



Materials and Methods

Studied Lights – Growth Chambers





Growth Chamber Measurements

Morphological & Physiological variables

✓ **Seedling evaluation: every week for a total of 7 weeks**

- ✘ Seedling growth rates
- ✘ Evaluation of needle color & number

✓ **At the end of the 7th week**

- ✘ Seedling shoot height, root length
- ✘ Seedling dry weights (leave shoot & roots) & root/shoot ratio

✓ **Physiological quality testing of seedlings**

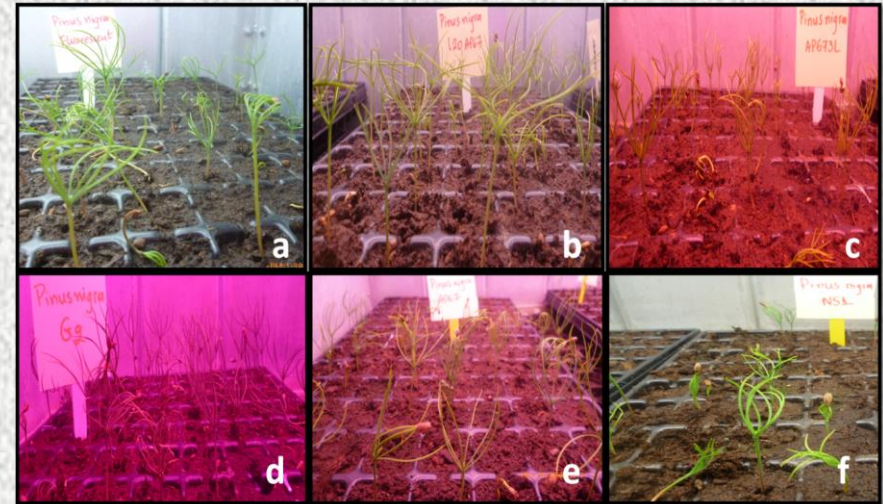
- **RGP** of each seedling was assessed by measuring the new root length (**NRL**) and new root dry weight (**NRDW**).





Seedlings of ***Castanea sativa Mill.*** into the growth chambers under

- a) L20AP67 LED,
- b) Fluorescent conventional light,
- c) AP673L LED,
- d) G2 LED,
- e) AP67 LED,
- f) NS1 LED,
- g) RGP light environment (FL & sodium lamps).



Seedlings of ***Pinus nigra L.*** into the growth chambers under

- a) Fluorescent conventional light,
- b) L20AP67 LED,
- c) AP673L LED,
- d) G2 LED,
- e) AP67 LED,
- f) NS1 LED.



Optimal light qualities suggested for some tested species

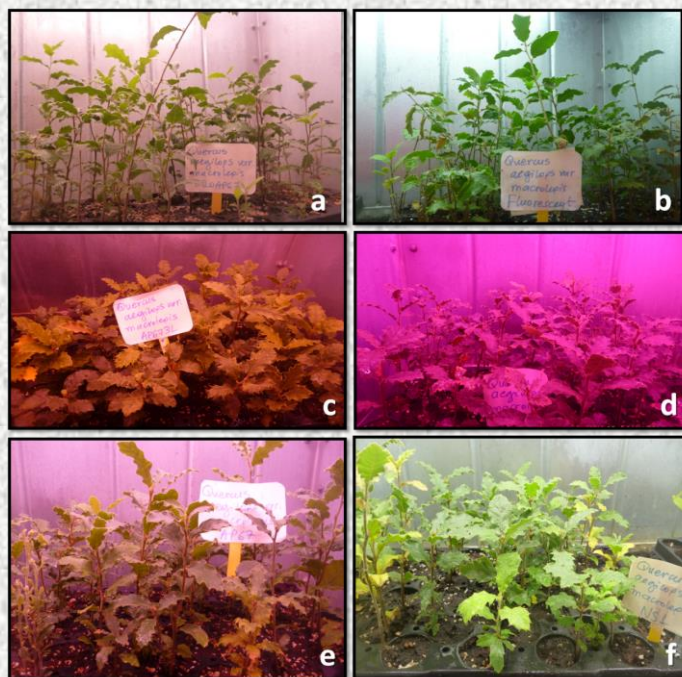
- *Pinus sylvestris* L.: AP67, AP673L, G2
- *Picea abies* Karst.: AP67, AP673L
- *Pinus nigra* Arn.: AP67, AP673L
- *Quercus ithaburensis* var. *macrolepis*: NS1, AP673L
- *Castanea sativa* Mill: AP673L, G2
- *Myrtus communis* L.: AP673L, NS1
- *Ocimum basilicum* L.: AP67, G2, NS1
- *Ocimum basilicum* RR hybrid: AP67, G2, NS1
- *Cornus sanguinea* L. : AP673L, G2, AP67
- *Prunus avium* L.: AP673L, G2, AP67
- *Punica granatum* L.: L20AP67,
- *Platanus orientalis* L.: AP67



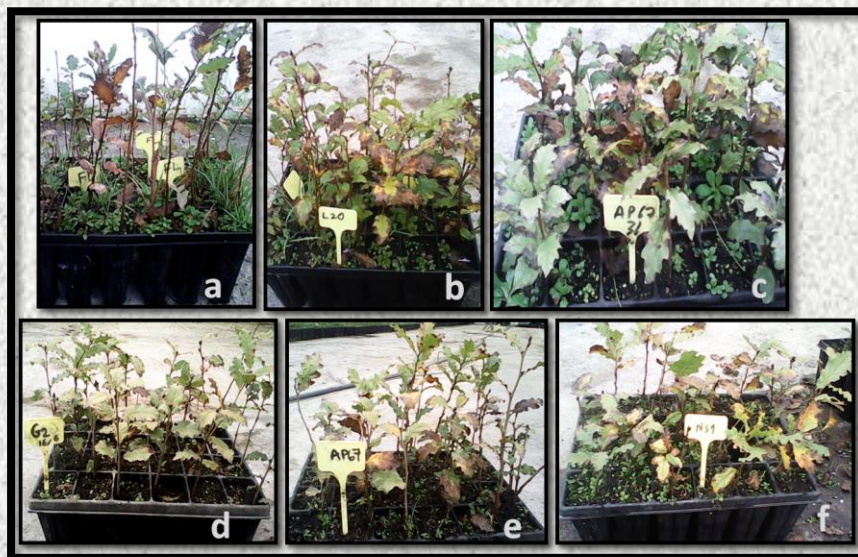
- Transplant from mini-plugs LED's to traditional nursery containers



Growth chambers



Nursery performance



Seedlings of Quercus ithaburensis var. macrolepis

a) *for the Fluorescent light,*

b) *L20AP67 LED,*

c) *AP673L LED,*

d) *G2 LED,*

e) *AP67 LED,*

f) *NS1 LED,*

after 6 month period at the nursery of Chalkidona.

Seedlings of Q. ithaburensis var. macrolepis in growth chambers under

a) *L20AP67,*

b) *Fluorescent conventional light,*

c) *AP673L*

d) *G2 LED,*

e) *AP67 LED,*

f) *NS1 LED.*



Field Performance



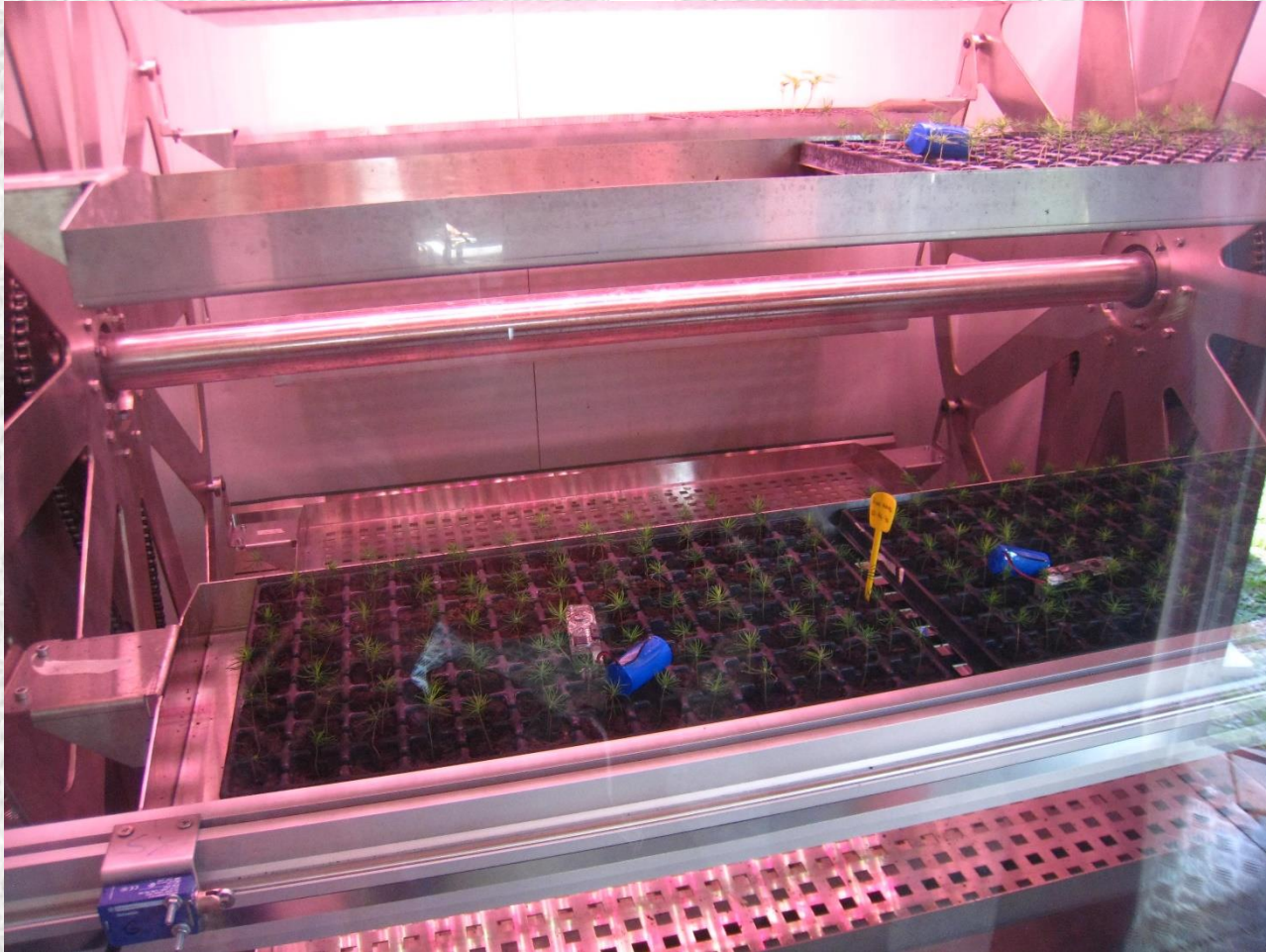
Seedlings grown both in LED lights and traditional nursery containers transplanted in the field area of nursery.



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Prototype

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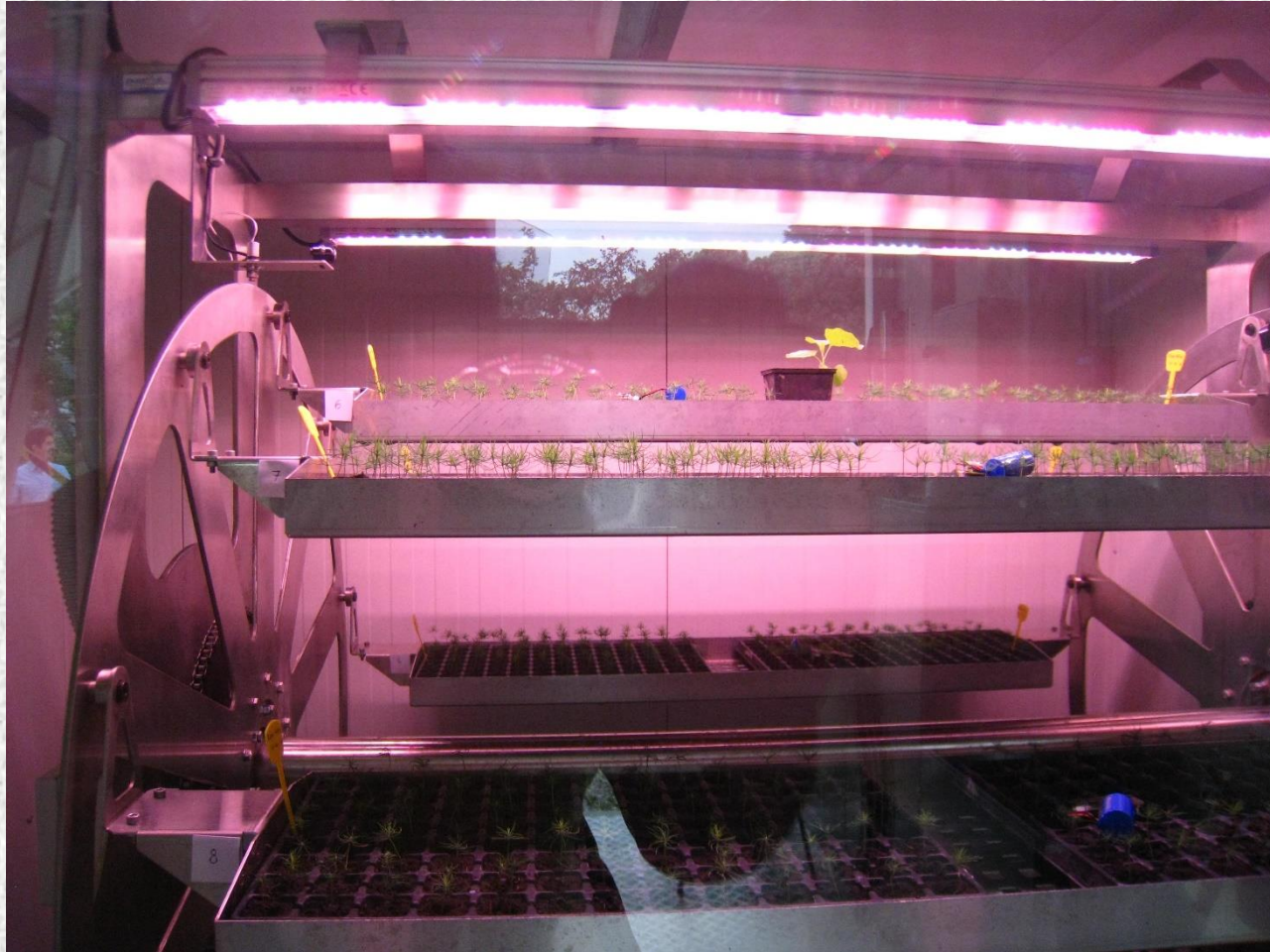




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Technical validation of the prototype with Mediterranean seedlings

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Energy independent prototype

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Conclusions

- Seedling production under **LEDs** resulted in the production of more **balanced seedlings** (regarding morphological & physiological attributes) for **Mediterranean species**.
- Industrial implementation of the prototype demands further cultivation protocols for more Mediterranean species.

Furthermore:

- **Seedling outdoor performance is needed to predict the chances for survival within a forest restoration program with high number of species, produced in the prototype**
- **Thus Nurseries can improve their production by increasing the number of species they produce by using the innovative proposed approaches**





Thank you for your attention