EUROLEGUME 2014-2017

Enhancing of legumes growing in Europe through sustainable cropping for protein supply for food and feed

FP7 Research Project Nº 61378

Agricultural University of Tirana

Could the artificial inoculation of AM fungi improve the benefits of using pea (*Pissum sativum* L) plants for soil amendment purposes in greenhouses?

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Salinity - a common and most severe environmental stressor in agriculture

- reduces the osmotic potential of the soil solution and consequently reduces the amount of water available to the plant causing physiological drought;
- induce cell toxicity due to excessive Na and Cl accumulation;
- induce nutrient imbalance;
- induce progressive oxidative damage and ultimately cell death due to production of reactive oxygen species (ROS);

Pea (*Pisum sativum* L.) - the role in sustainable cropping systems

- grown essentially for its protein-rich seeds;

But also, ...

- essential component of sustainable cropping systems (symbiotic nitrogen fixation; break crop for pest and pathogen reduction),
- increase the soil reserves of organic matter and stimulate soil biological activity,
- improve soil structure, water-holding capacity and aeration,

Pea – a very sensitive crop to salinity

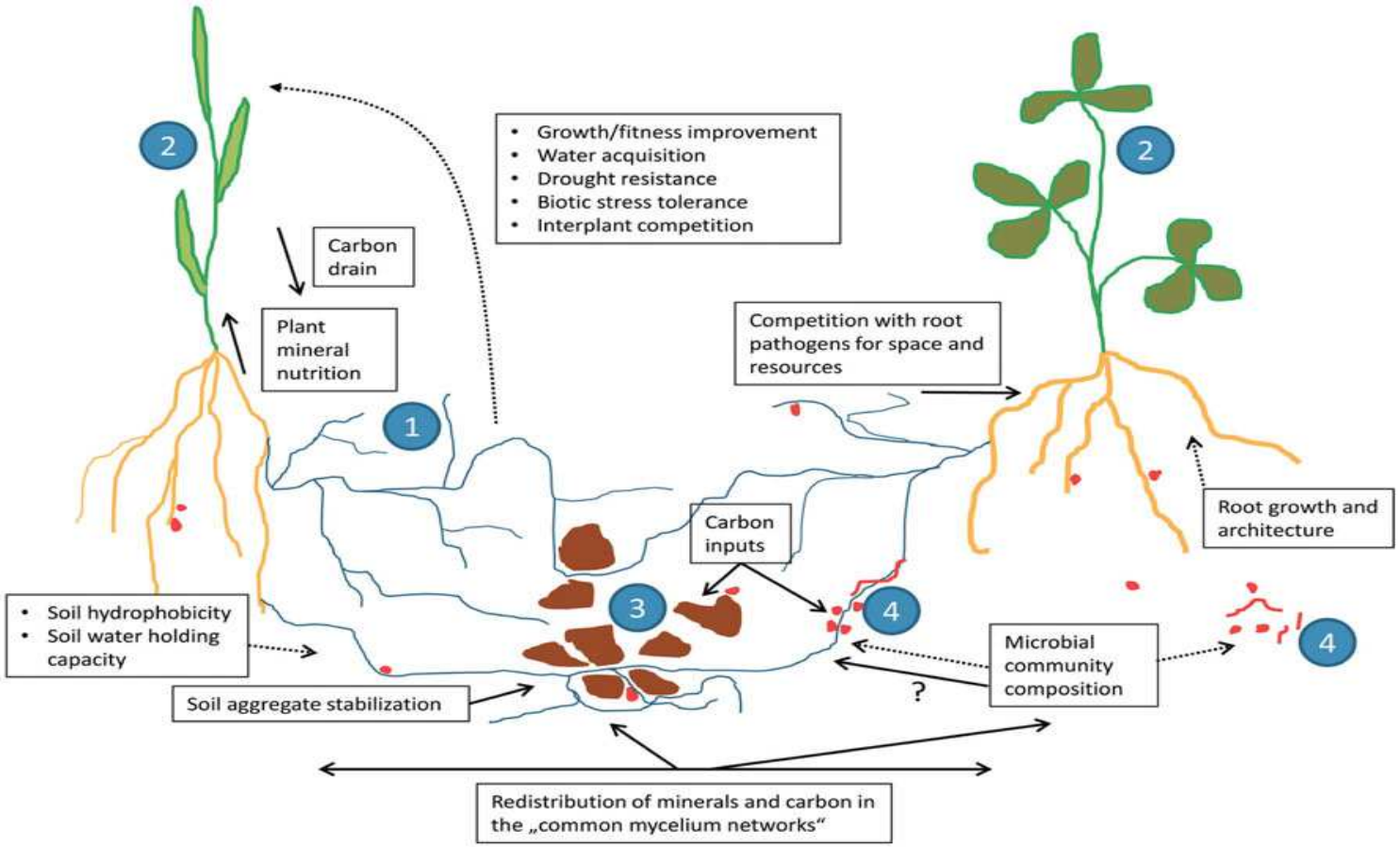
… two-phase growth response to salinity (Munns 2002);

- the first phase; essentially a water stress or osmotic phase, and the growth reduction is presumably regulated by hormonal signals coming from the roots.
- the second phase; salts accumulation in leaves exceeding the ability of the cells to compartmentalize salts in the vacuole, and finally causing leaf dieback.

Arbuscular mychorrhizae fungi (AMF);

environmentally friently
alternative to salinity stress alleviation

Schematic representation of the different functions played by the arbuscular mycorrhizal (AM) fungi (1) in the physiology and ecology of their host plants (2)
(Jansa, Bukovská and Gryndler, 2013**)**



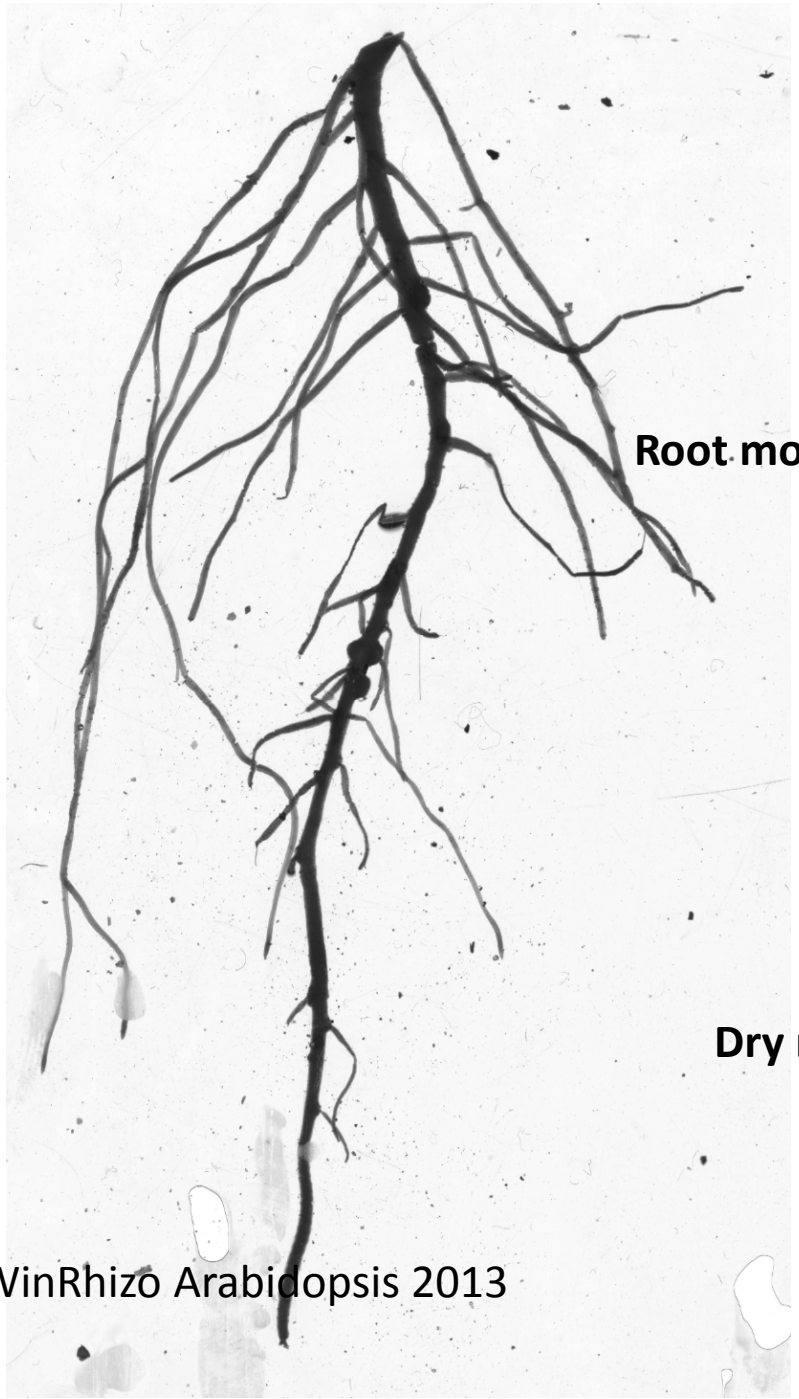
Mycorrhizal hyphae interconnect roots with soil particles (3), provide direct connections of root systems of different plant individuals (2), and interact with a number of soil microbes (4).

Objective

… to assess the impact of a commercial mixture of AM fungi (*Glomus intraradices*, *Glomus etunicatum*, *Glomus mosseae*,*Glomus geosporum*, and *Glomus clarum*) on plant growth parameters and nutrient absorption capacity of pea plants.

Materials and Methods

Parameters evaluated;



WinRhizo Arabidopsis 2013

Root morphology parameters

- **Root length (cm)**
- Root projected area (cm^2)
- **Root surface area (cm^2)**
- Average root diameter (mm)
- **Root volume (cm^3)**
- Tips
- Forks

Dry matter

- DM_{Roots}
- DM_{Shoots}

Materials and Methods

Analyses conducted;	N, P, K (on mixed root+shoot plant material)
Total nutrient uptake (N, P, K);	X accumulation (mg) = X concentration (mg g^{-1}) × plant dry weight (g)

Materials and Methods

Specific absorption rate (SAR; $mg^{-1}g\ d^{-1}$) (Martinez et al. 2008);

$$SAR = \frac{Xt2 - Xt1}{t2 - t1} x \frac{\ln wt2 - \ln wt1}{wt2 - wt1}$$

Specific utilization rate (SUR; $g\ mg^{-1}\ d^{-1}$) (Martinez et al. 2008);

$$SUR = \frac{Wt2 - Wt1}{t2 - t1} x \frac{\ln Xt2 - \ln Xt1}{Xt2 - Xt1}$$

X1 and X2 is plant nutrient content (mg) at the start and at the end of analyzed period,
W is plant dry matter (DM_{Plant}) and w is the root dry weight (DM_{Root}),
t2 = end of analyzed period (DAS 18), and t1 = start of analyzed period (DAS 52)

Results

Salinity effects pea plants

Root length (cm) of pea plants with/without arbuscular mycorrhizal fungi (amf+/amf-) and under two levels of salinity (0 and 50 mM NaCl).

Factors		Day after sowing (DAS)		
AMF	Salinity	18	42	58
AMF-	0 mM	82.3±13.8a	171.1±17.2a	180.1±33.7a
	50 mM	57.5±14.8b	120.2±22.3b	124.8±10.3b
AMF+	0 mM	81.5±14.0a	155.7±23.1a	162.5±22.0a
	50 mM	67.6±13.1b	121.1±13.1b	126.8±5.94b
Significance				
AMF		ns	ns	ns
Salinity		**	***	***
AMFxSalinity		ns	ns	ns

Root surface (cm^2) of pea plants with/without arbuscular mycorrhizal fungi (amf+/amf-) and under two levels of salinity (0 and 50 mM NaCl).

Factors		Day after sowing (DAS)		
AMF	Salinity	18	42	58
AMF-	0 mM	23.0±2.9a	35.9±5.4a	42.8 ±9.3a
	50 mM	14.9±1.8b	24.4±5.4b	26.3±4.9b
AMF+	0 mM	20.6±5.1a	31.6±5.3a	36.1±5.3a
	50 mM	19.9±2.5b	26.5±1.9b	27.0±3.2b
Significance				
AMF		ns	ns	ns
Salinity		**	***	***
AMFxSalinity		ns	ns	ns

Rot volume (cm^3) of pea plants with/without arbuscular mycorrhizal fungi (amf+/amf-) and under two levels of salinity (0 and 50 mM NaCl).

Factors		Day after sowing (DAS)		
AMF	Salinity	18	42	58
AMF-	0 mM	0.47±0.08a	0.57±0.10a	0.76±0.19a
	50 mM	0.31±0.04b	0.40±0.08b	0.42±0.07b
AMF+	0 mM	0.43±0.08a	0.50±0.08a	0.66±0.09a
	50 mM	0.39±0.05b	0.42±0.06b	0.42±0.09b
Significance				
AMF		ns	ns	ns
Salinity		**	***	***
AMFxSalinity		ns	ns	ns

Dry matter (gr) of aboveground part ($DM_{Shoots+Leaves}$) of pea plants with/without arbuscular mycorrhizal fungi (AMF+/AMF-) and under two levels of salinity (0 and 50 mM NaCl).

Factors		Day after sowing (DAS)		
AMF	Salinity	18	42	58
AMF-	0 mM	0.058±0.005 a	0.297±0.042b	2.140±0.533ab
	50 mM	0.047±0.004b	0.265±0.032b	1.767±0.377b
AMF+	0 mM	0.054±0.008ab	0.434±0.046a	2.744±0.265a
	50 mM	0.046±0.007b	0.269±0.023b	1.341±0.057b
Significance				
AMF		ns	***	*
Salinity		***	***	***
AMFxSalinity		ns	***	**

Nitrogen (N) specific absorption rate in regard to root length (**g cm^{-1} d^{-1}**), root surface (**g cm^{-2} d^{-1}**) and root volume (**g cm^{-3} d^{-1}**) of pea plants with/without arbuscular mycorrhizal fungi (AMF+/AMF-) and under two levels of salinity (0 and 50 mM NaCl).

Factors		**SAR_N**		
AMF	Salinity	**g cm^{-1} d^{-1}**	**g cm^{-2} d^{-1}**	**g cm^{-3} d^{-1}**
AMF-	0 mM	0.39±0.16b	0.30±0.11b	0.47±0.16b
	50 mM	0.31±0.15b	0.15±0.08b	0.43±0.09b
AMF+	0 mM	1.04±0.22a	0.65±0.25a	0.82±0.20a
	50 mM	0.75±0.15a	0.49±0.08a	1.22±0.29a
Significance				
AMF		***	***	***
Salinity		*	*	*
AMFxSalinity		ns	ns	*

Phosphorus (P) specific absorption rate in regard to root length (**g cm^{-1} d^{-1}**), root surface (**g cm^{-2} d^{-1}**) and root volume (**g cm^{-3} d^{-1}**) of pea plants with/without arbuscular mycorrhizal fungi (AMF+/AMF-) and under two levels of salinity (0 and 50 mM NaCl).

Factors		**SAR_P**		
AMF	Salinity	**g cm^{-1} d^{-1}**	**g cm^{-2} d^{-1}**	**g cm^{-3} d^{-1}**
AMF-	0 mM	0.11±0.04c	0.09±0.02c	0.15±0.07b
	50 mM	0.09±0.04c	0.04±0.02d	0.13±0.02b
AMF+	0 mM	0.31±0.06a	0.19±0.07a	0.24±0.07a
	50 mM	0.16±0.03b	0.11±0.01b	0.26±0.06a
Significance				
AMF		***	***	***
Salinity		***	**	*
AMFxSalinity		**	ns	*

Potassium (K) specific absorption rate in regard to root length (**g cm^{-1} d^{-1}**), root surface (**g cm^{-2} d^{-1}**) and root volume (**g cm^{-3} d^{-1}**) of pea plants with/without arbuscular mycorrhizal fungi (AMF+/AMF-) and under two levels of salinity (0 and 50 mM NaCl).

Factors		**SAR_K**		
AMF	Salinity	**g cm^{-1} d^{-1}**	**g cm^{-2} d^{-1}**	**g cm^{-3} d^{-1}**
AMF-	0 mM	0.21±0.06b	0.16±0.04b	0.28±0.15b
	50 mM	0.16±0.08b	0.08±0.04a	0.23±0.05b
AMF+	0 mM	0.63±0.13a	0.39±0.15a	0.51±0.15a
	50 mM	0.22±0.04b	0.15±0.02b	0.36±0.08ab
Significance				
AMF		***	***	**
Salinity		***	***	ns
AMFxSalinity		***	*	ns

Specific absorption rate (mg $g^{-1}d^{-1}$) of N (SAR_N), P (SAR_P) and K (SAR_K) of pea plants with/without arbuscular mycorrhizal fungi (AMF+/AMF-) and under two levels of salinity (0 and 50 mM NaCl).

Factors		Day after sowing (DAS)		
AMF	Salinity	SAR_N	SAR_P	SAR_K
AMF-	0 mM	9.21±3.75d	2.64±1.07d	4.61±1.87d
	50 mM	14.0±1.88c	4.15±0.55c	6.91±0.94c
AMF+	0 mM	21.8±3.78b	6.55±1.13b	10.7±1.84b
	50 mM	41.2±10.6a	8.57±2.21a	13.4±3.47a
Significance				
AMF		***	***	***
Salinity		***	**	*
AMFxSalinity		**	**	**

Specific utilization rate (g mg^{-1} d^{-1}) of N (SUR_N), P (SUR_P) and K (SUR_K) of pea plants with/without arbuscular mycorrhizal fungi (AMF+/AMF-) and under two levels of salinity (0 and 50 mM NaCl).

Factors		Day after sowing (DAS)		
AMF	Salinity	SUR_N	SUR_P	SUR_K
AMF-	0 mM	0.0025±0.00b	0.0090±0.00b	0.0051±0.00
	50 mM	0.0027±0.00b	0.0091±0.00b	0.0053±0.00
AMF+	0 mM	0.0030±0.00a	0.0103±0.00a	0.0058±0.00
	50 mM	0.0016±0.00c	0.0080±0.00b	0.0053±0.00
Significance				
AMF		**	ns	ns
Salinity		***	**	ns
AMFxSalinity		***	**	ns

Conclussions

- Saline irrigation water strongly diminishes the growth of pea plants and strongly reduces the absorption capacity of its root system.
- The inoculation of AM fungi in the growing substrate could alleviate salinity stress effects by increasing plant biomass and improving the absorption capacity of plants' root system.
- The artificial inoculation of AM fungi could be considered as an effective and environmentally friendly alternative to significantly increase the volume and improve the quality of biomass in pea plants grown for greenhouses soil amendment (green manure) purposes

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FP7 Research Project № 61378

This results are part of the work conducted in the framework of EUROLEGUME Project (Enhancing of legumes growing in Europe through sustainable cropping for protein supply for food and feed), founded by 7th Research Framework Programme of the European Union (FP7 Research Project No. 61378)

Dry matter (gr) of root system (DM_{Root}) of pea plants with/without arbuscular mycorrhizal fungi (amf+/amf-) and under two levels of salinity (0 and 50 mM NaCl).

Factors		Day after sowing (DAS)		
AMF	Salinity	18	42	58
AMF-	0 mM	0.016±0.003b	0.084±0.008a	0.209±0.038a
	50 mM	0.020±0.004ab	0.043±0.008b	0.131±0.011b
AMF+	0 mM	0.023±0.004a	0.065±0.018a	0.134±0.019b
	50 mM	0.019±0.005ab	0.037±0.007b	0.073±0.009c
Significance				
AMF		*	*	***
Salinity		ns	***	***
AMFxSalinity		**	ns	ns