

Phytoremediation assisted by arbuscular mycorrhizal fungi to clean-up dioxins/furans polluted soil : a microcosm experiment

Hacène MEGLOULI

Pr Anissa LOUNES-HADJ SAHRAOUI and Dr Joël FONTAINE

Université Littoral Côte d'Opale (ULCO) - FRANCE

Hacene.Meglouli@univ-littoral.fr

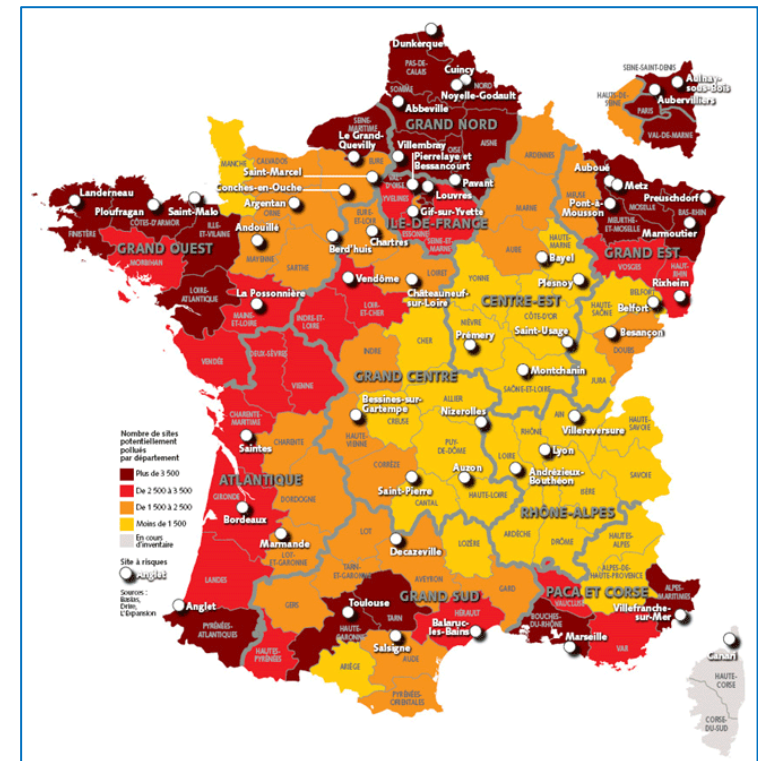
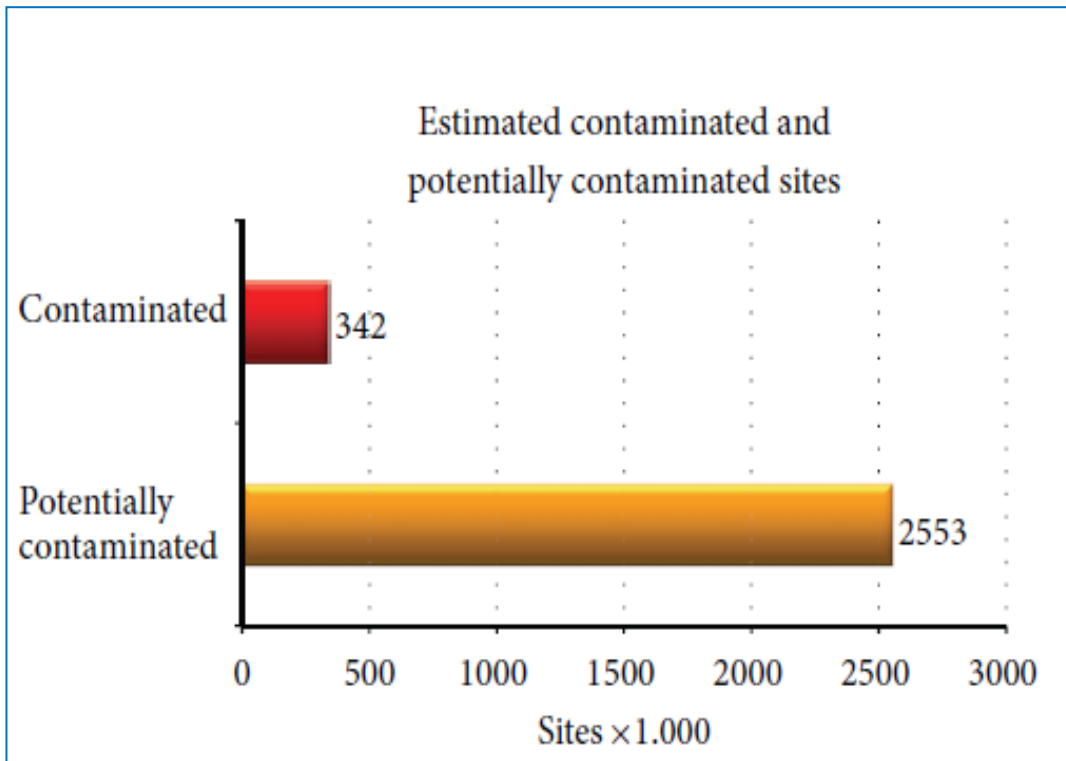


Lille 16 Mars 2016

CONTEXT AND PROBLEMATIC

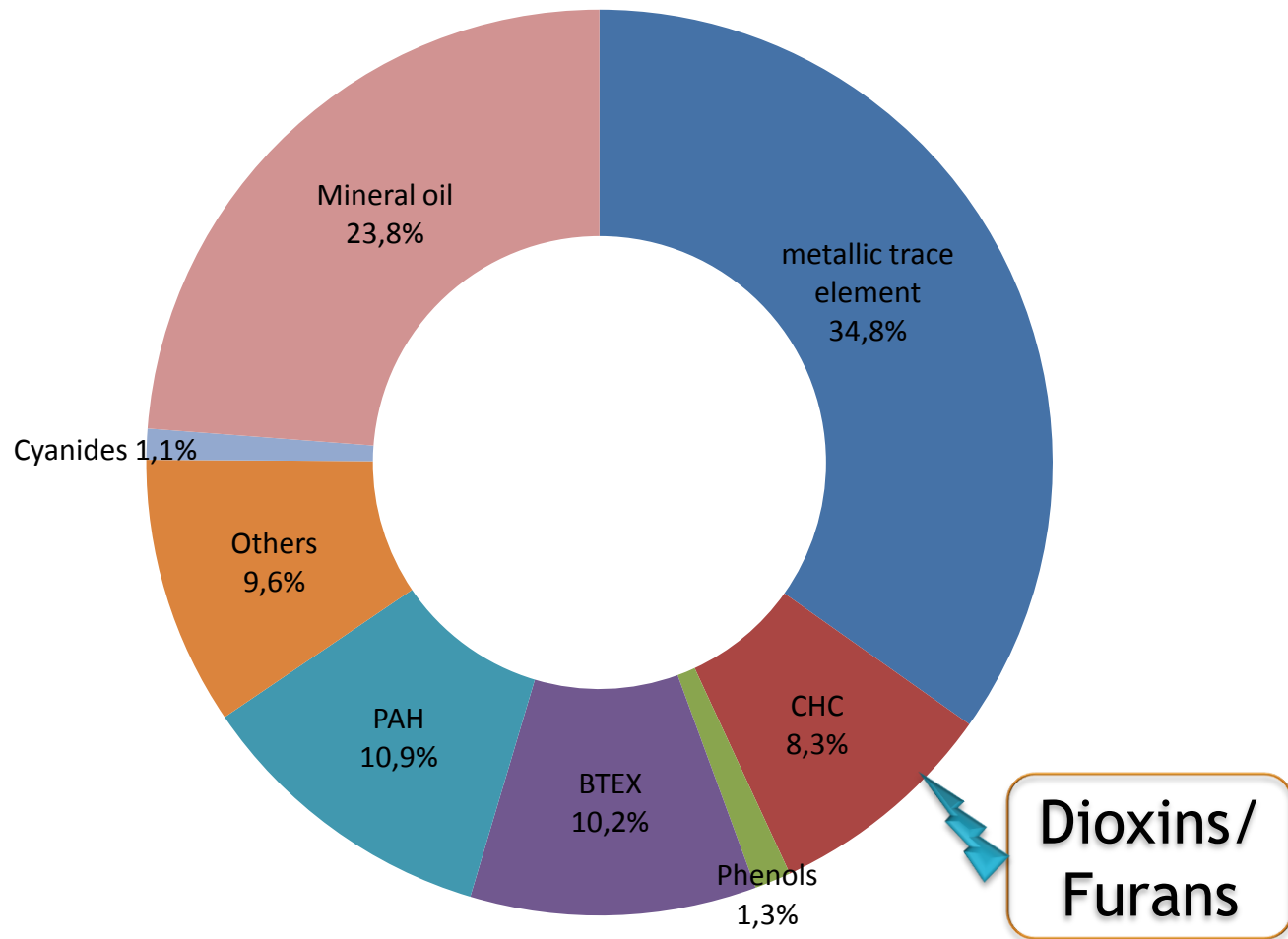
- In the world: between 5 to 10 million contaminated sites (www.siteremediation.com.au/contaminated-soil/contamination/).
- In Europe, 2 895 000 polluted sites listed

- In France, 6287 sites listed
- In Nord-Pas de Calais- Picardie 665 sites listed



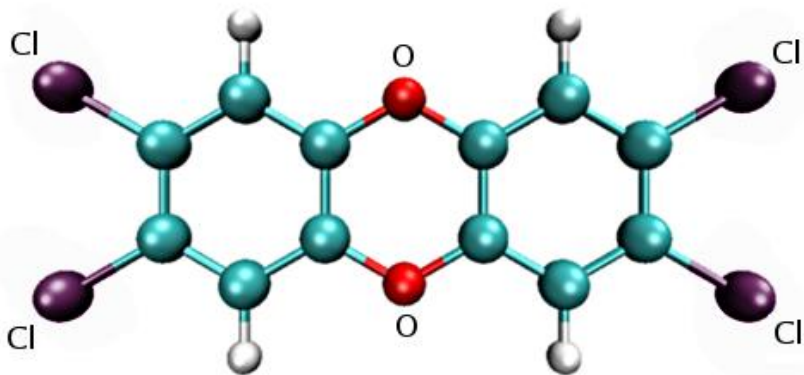
Panagos et al, 2013

CONTEXT AND PROBLEMATIC

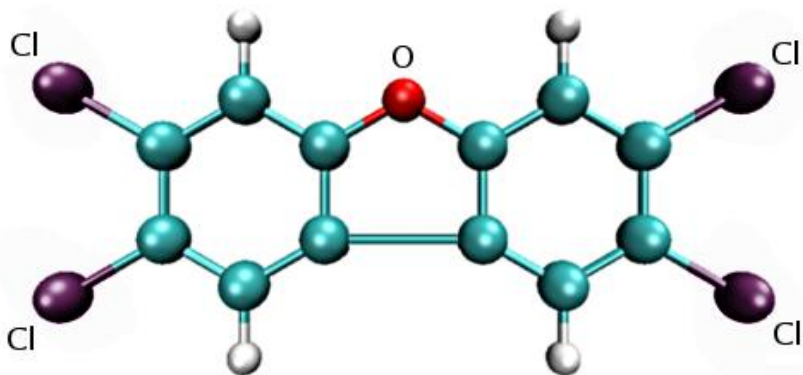


Panagos et al, 2013

DIOXINS AND FURANS



Dioxins 75 congeners



Furans: 135 congeners

- Lipophilic, semi-volatile
- Recalcitrant to biodegradation
- Half-life time : 10 - 100 years
- High toxicity : Cancer, neurological and immunological effects, ...

PHYTOREMEDIATION

Advantages :

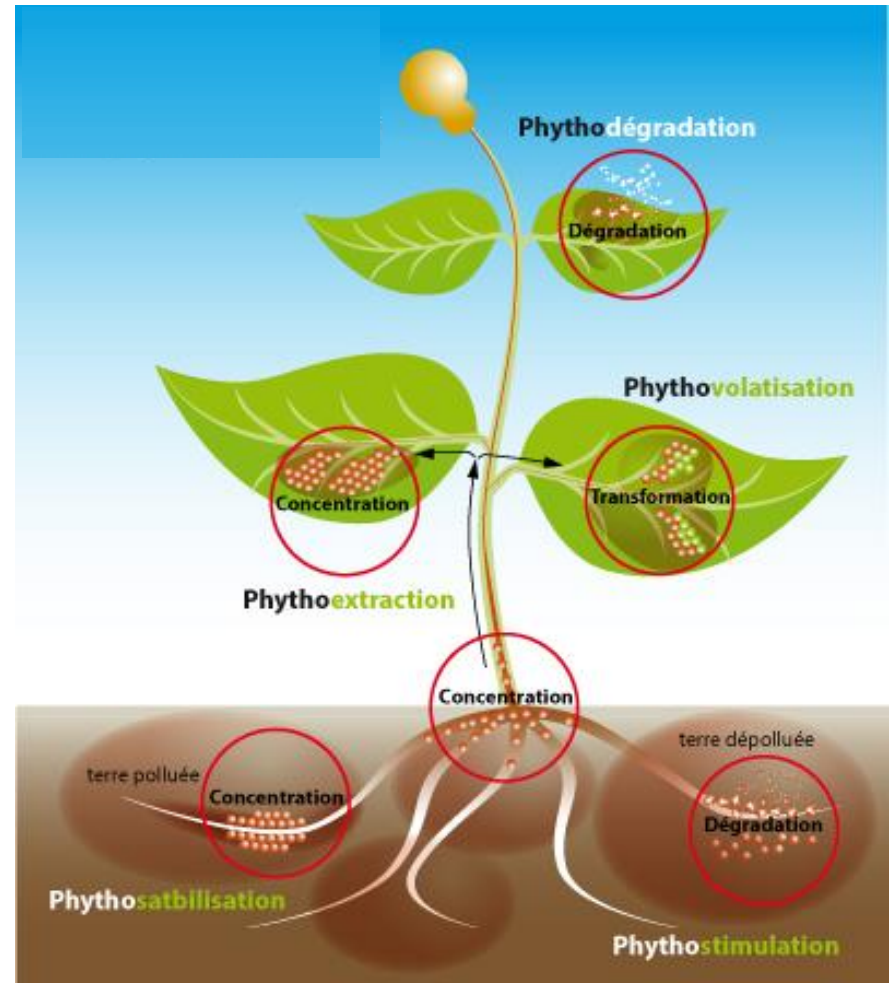
- Economic, Ecological
- Applicable over large areas
- Well accepted by the public
 - Easy to implement

But

Phytoremediation Efficiency

=

f (plant tolerance to pollutant toxicity)



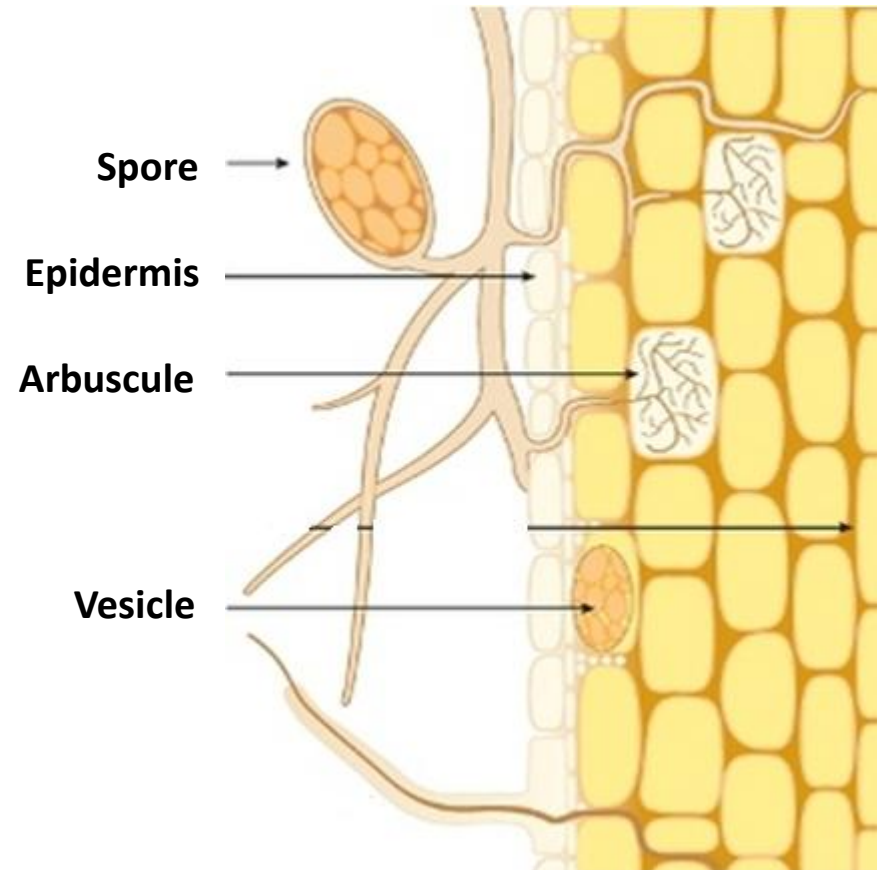
ARBUSCULAR MYCORRHIZAL FUNGI (AMF)

Mutualistic symbiosis association between a mycorrhizal fungus and plant roots

Arbuscular mycorrhizal fungi benefits:

- Better water and mineral nutrition
- Better stress tolerance (biotic + abiotic)
- Better dissipation of organic pollutants

Smith et Read (2010)



Bonfante & Genre (2013)

AMF-ASSISTED PHYTOREMEDIATION & DISSIPATION OF ORGANIC POLLUTANTS

Organic pollutants	plants	AMF	Degradation rates (%)	Experiment duration (Weeks)	References
Polycyclic Aromatic Hydrocarbons (PAH)	Clover, Ryegrass	<i>Funneliformis mosseae</i>	42-66	16	Joner et al, 2001
	Alfalfa	<i>Glomus caledonium</i>	57-86	13	Liu et al, 2004
	Leek	<i>Glomus intraradices</i> , and <i>G. versiforme</i>	43-88	12	Liu and Dalpé, 2009
	Alfalfa	<i>Funneliformis mosseae</i> and <i>G. etunicatum</i>	88-98	10	Gao et al, 2011
PolyChlorinated Biphenyl (PCB)	Alfalfa	<i>Glomus caledonium</i>	12	16	Teng et al, 2010
Dioxins/Furans (PCDD/F)	No studies available				

AIM OF THE STUDY

The efficiency of phytoremediation assisted by arbuscular mycorrhizal fungi to clean-up dioxins/furans polluted soil

PROJECT CONTEXT

research project "Dioxins: contamination and biodegradation"

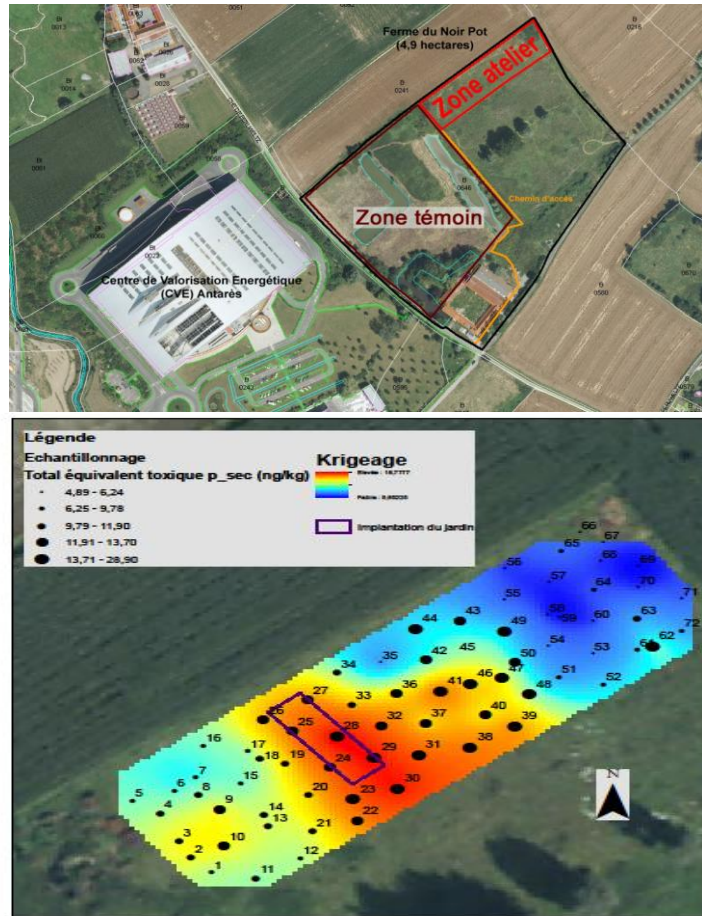
Coordination: Halluin Association 3R

Financial partners : PO FEDER, ADEME, LMCU and NPdCP Region

pollution origin: Atmospheric emissions of a garbage incinerator

EXPERIMENTAL SITE

- Experimental site: of 3500 m² historically contaminated with dioxins/ furans (PCDD/F)



PCDD/F (ng/kg)

200

PCDD/F (ng I-TEQ/kg)

16

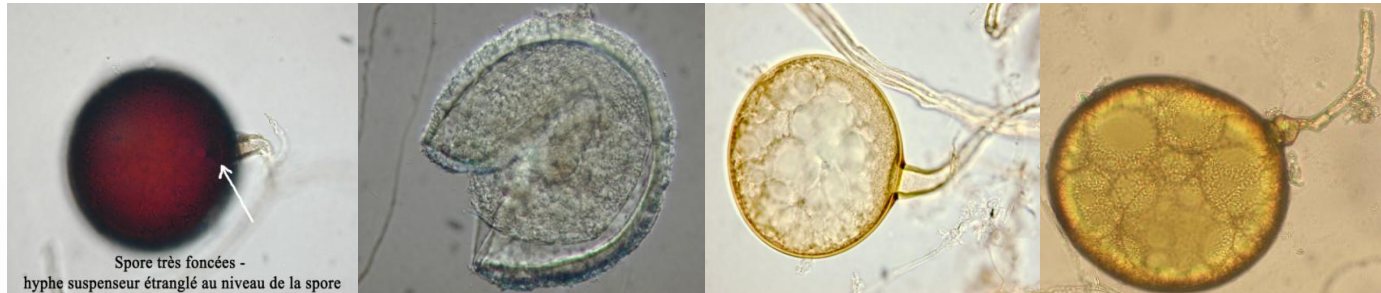
GC/HRMS (INRA d'Arras)

PRELIMINARY FEASIBILITY STUDY OF PHYTOREMEDIATION ASSISTED BY AMF

- **Isolation and identification of AMF spores from contaminated soil ?**
- **Germination of AMF spores in presence of dioxins / furans ?**
- **Germination of plant seeds on the contaminated soil ?**

PRELIMINARY FEASIBILITY STUDY OF PHYTOREMEDIATION ASSISTED BY AMF

➤ Isolation and identification of AMF spores from contaminated soil with dioxin/furan

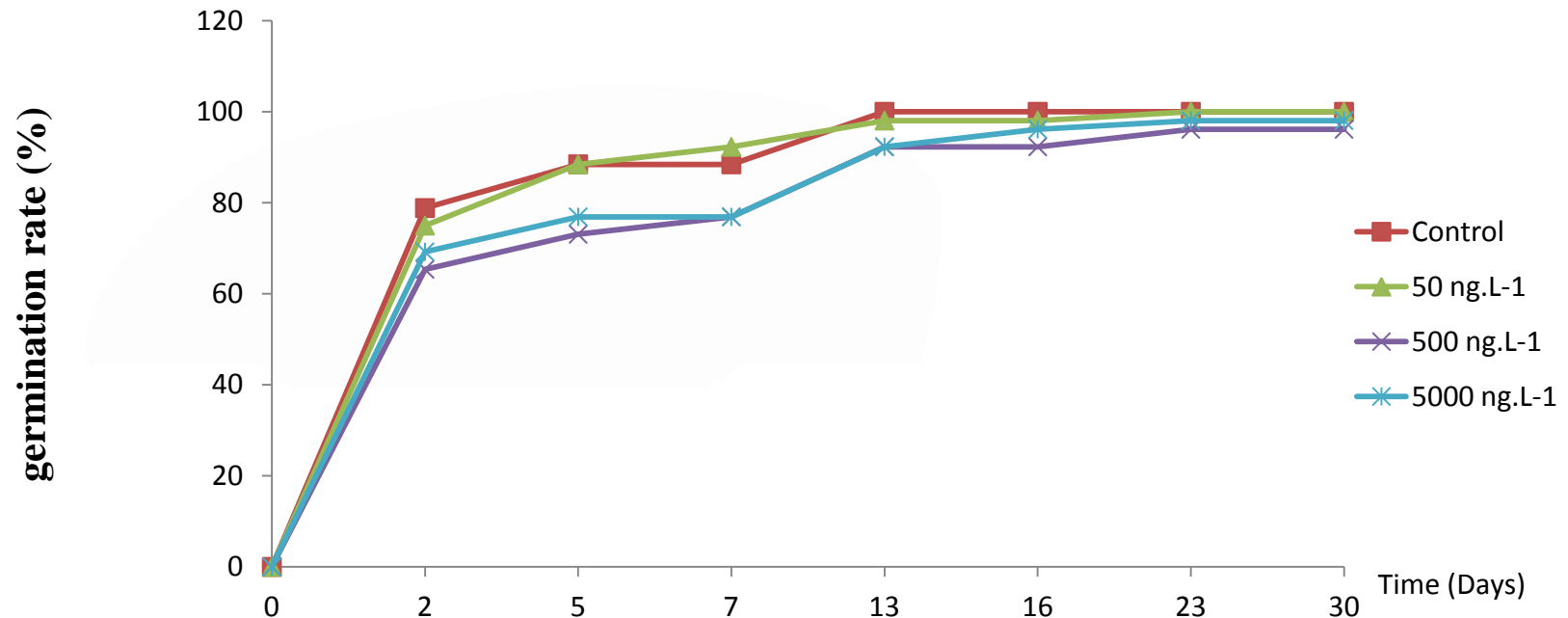


Species of spore	<i>Glomus Constrictum</i>	<i>Glomus lamellosum</i>	<i>Glomus geosporum</i>	<i>Funneliformis mosseae</i>	Total
Number of spores/ 100g of soil	10	20	200	230	460

- Presence of AMF spores on experimental site.

PRELIMINARY FEASIBILITY STUDY OF PHYTOREMEDIATION ASSISTED BY AMF

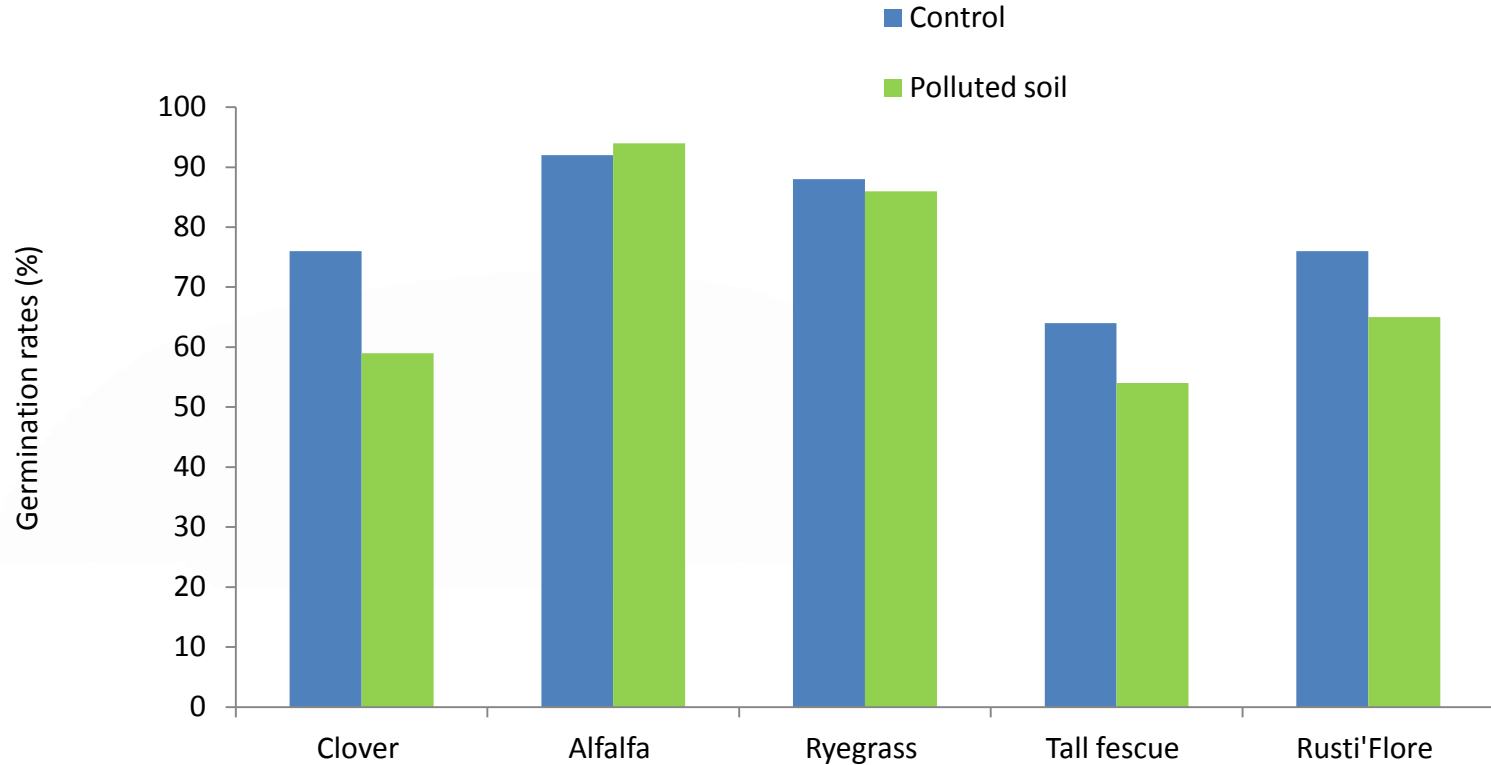
➤ Impact of dioxins/furans on the germination of spores *Rhizophagus irregularis*



- Delayed effect of PCDD/F on spore germination of *Rhizophagus irregularis*.

PRELIMINARY FEASIBILITY STUDY OF PHYTOREMEDIATION ASSISTED BY AMF

➤ Impact of dioxins / furans on the germination of plant seeds



- Slight effect of PCDD/F on the germination of clover seeds

PRELIMINARY FEASIBILITY STUDY OF PHYTOREMEDIATION ASSISTED BY AMF

Presence of AMF spores
in contaminated soil
with dioxins/furans

Ability of AMF spores
to germinate at high
concentrations of
dioxins/furans.

Capacity of plant seeds
to germinate on polluted
soil with dioxins/furans

Feasibility of phytoremediation assisted by AMF
possible on the polluted soil by dioxins / furans

EXPERIMENTAL SET-UP



Clover



Alfalfa



Ryegrass



Tall Fescue



Rusti'Flore

Non-vegetated
soil : Control



Sterilized soil
(Ionisation 45 kGy, IONISOS
Sablé sur Sarthe)



Commercial
AMF Inocula

Experimental Conditions

5 Replicats/condition

Inoculated sterilized soil + microflora

Non inoculated sterilized soil + microflora

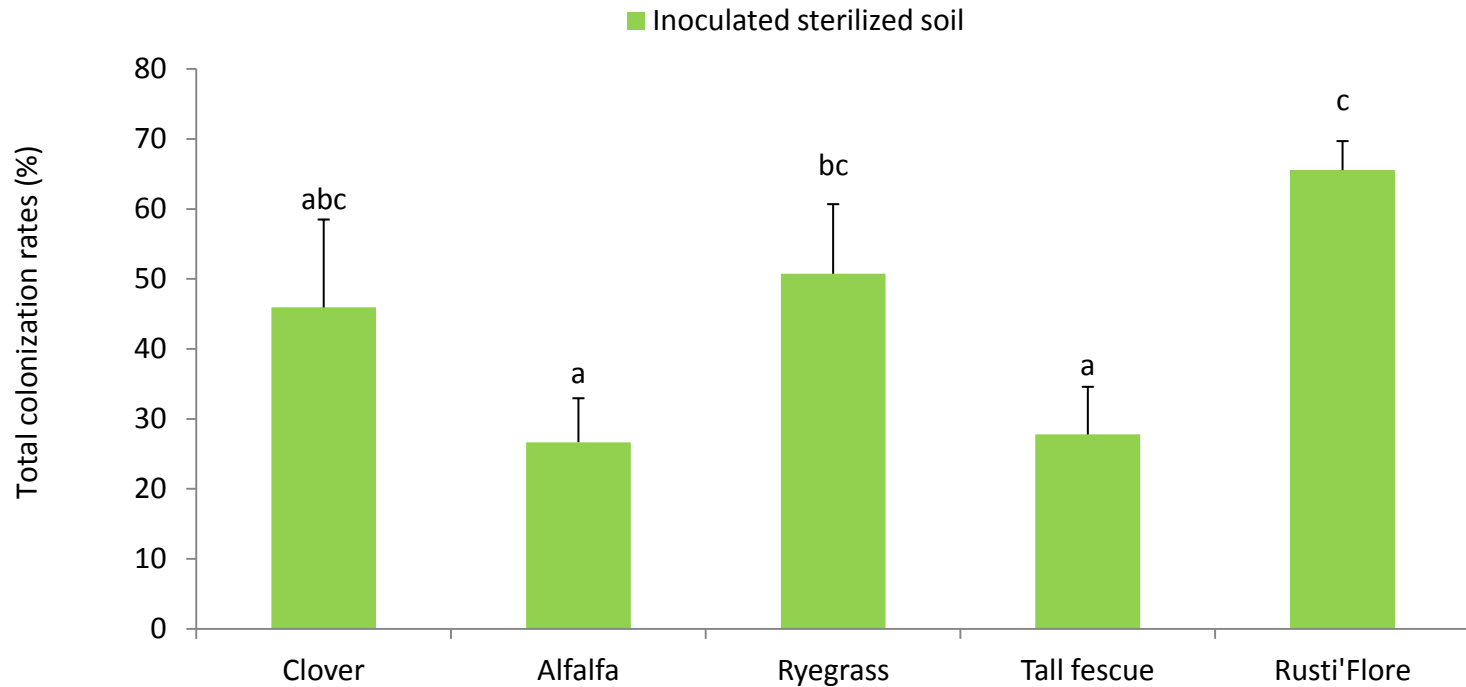
After 20 Weeks



- Mycorrhization rate (McGonigle *et al.*, 1990)
- Plant Biomass
- Residual dioxins/furans concentrations
- Quantification of telluric microbial biomass: specific Phospholipid fatty acid by GC/MS
 - Bacterial bioamass : Gram+ (i15:0, a15:0, i16:0, i17:0, a17:0), Gram- (cy17: 0, C18:1 ω 7, cy19:0)
 - Fungal biomass (saprotrophic and ectomycorrhizal) : c18:2 ω 6,9
- Microbial activity: Deshydrogenase (DHA) (adapted from Tabatabai, 1994)

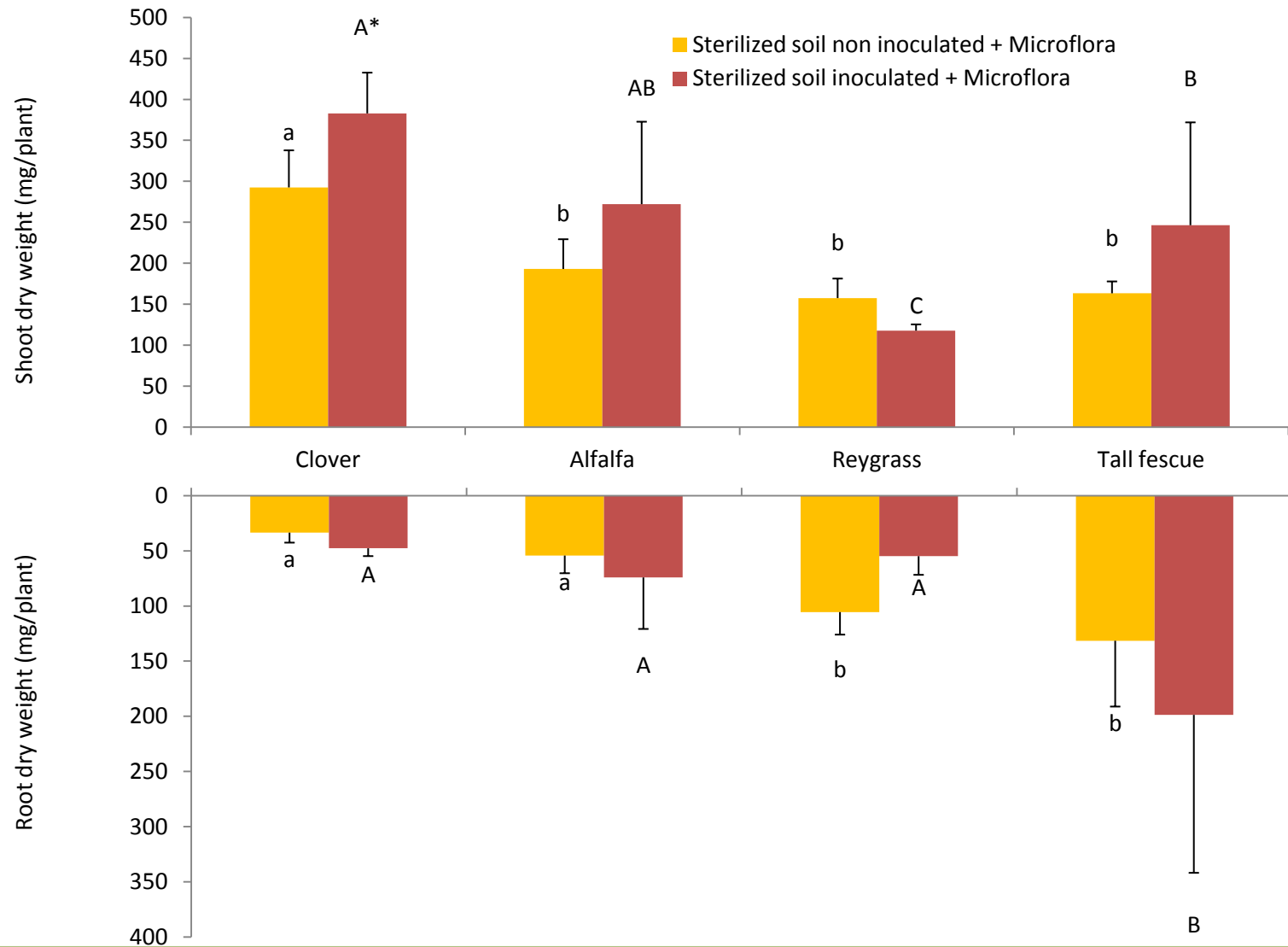
Results and discussion

MYCORRHIZAL COLONIZATION



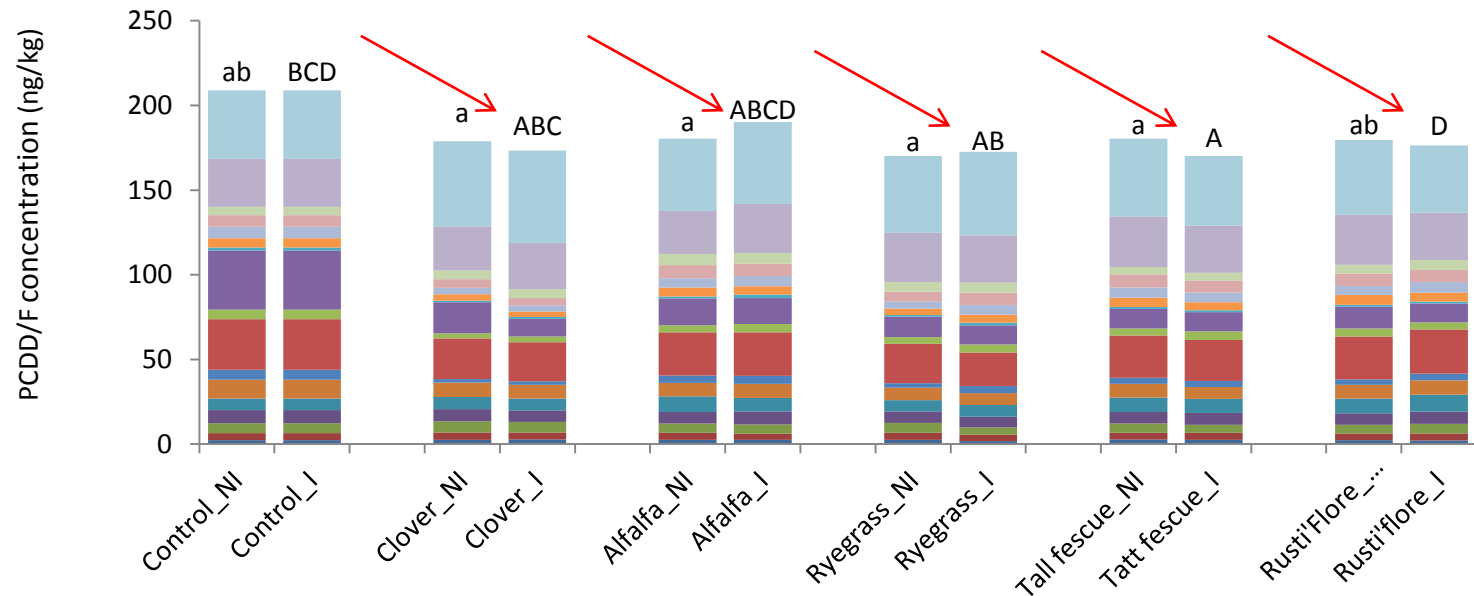
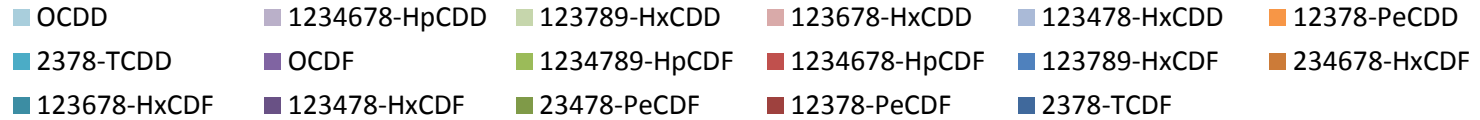
- Colonization of different plants with exogenous inoculum after 20 weeks of culture

PLANT GROWTH



- Beneficial effect of mycorrhiza on plant growth especially in clover
- Good growth of plants tested on contaminated soil after 20 weeks.

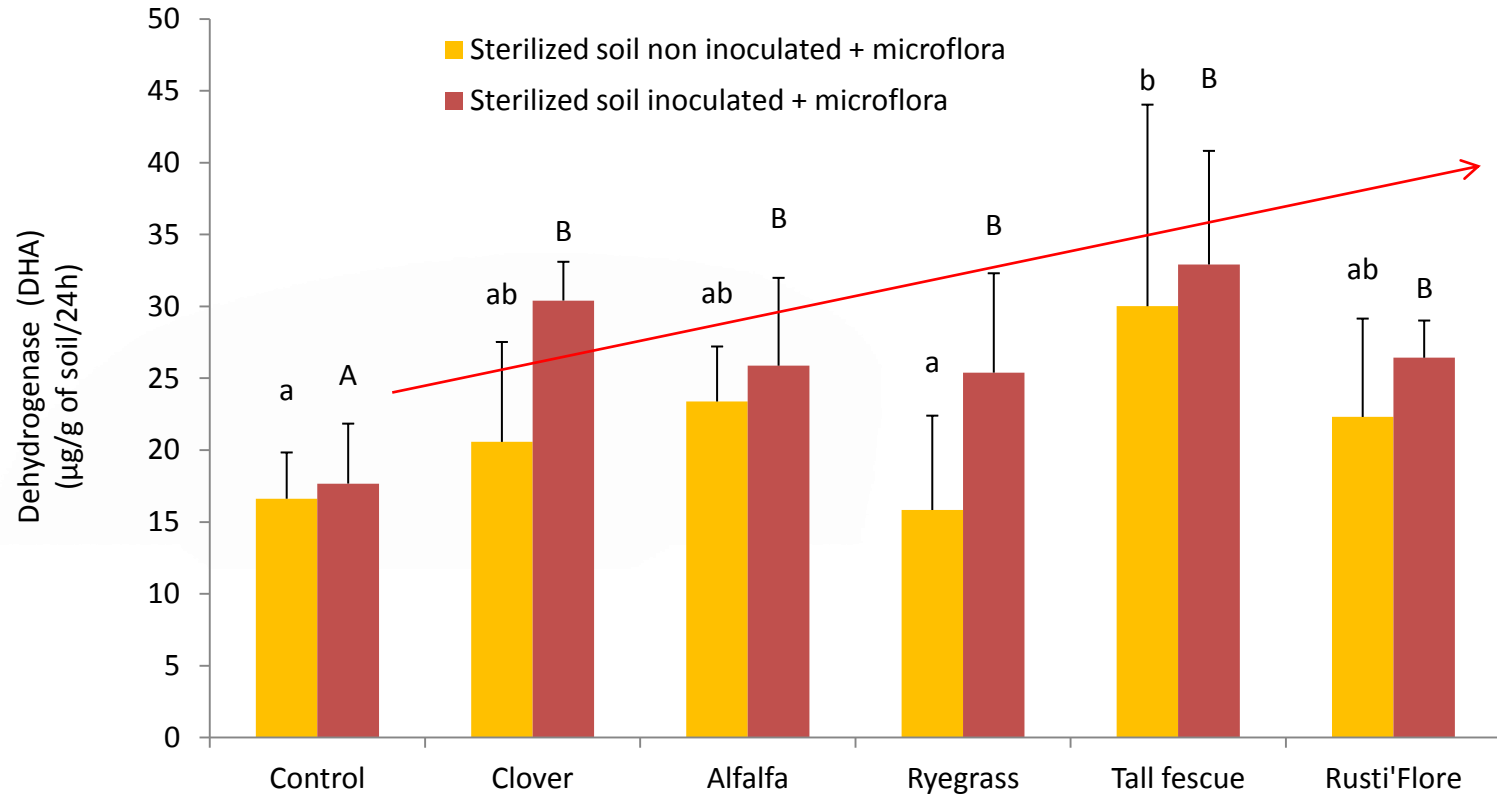
RESIDUAL DIOXINS/FURANS CONCENTRATIONS IN THE SOIL



- Reduced levels of PCDD/F in the vegetated soil after 20 weeks.

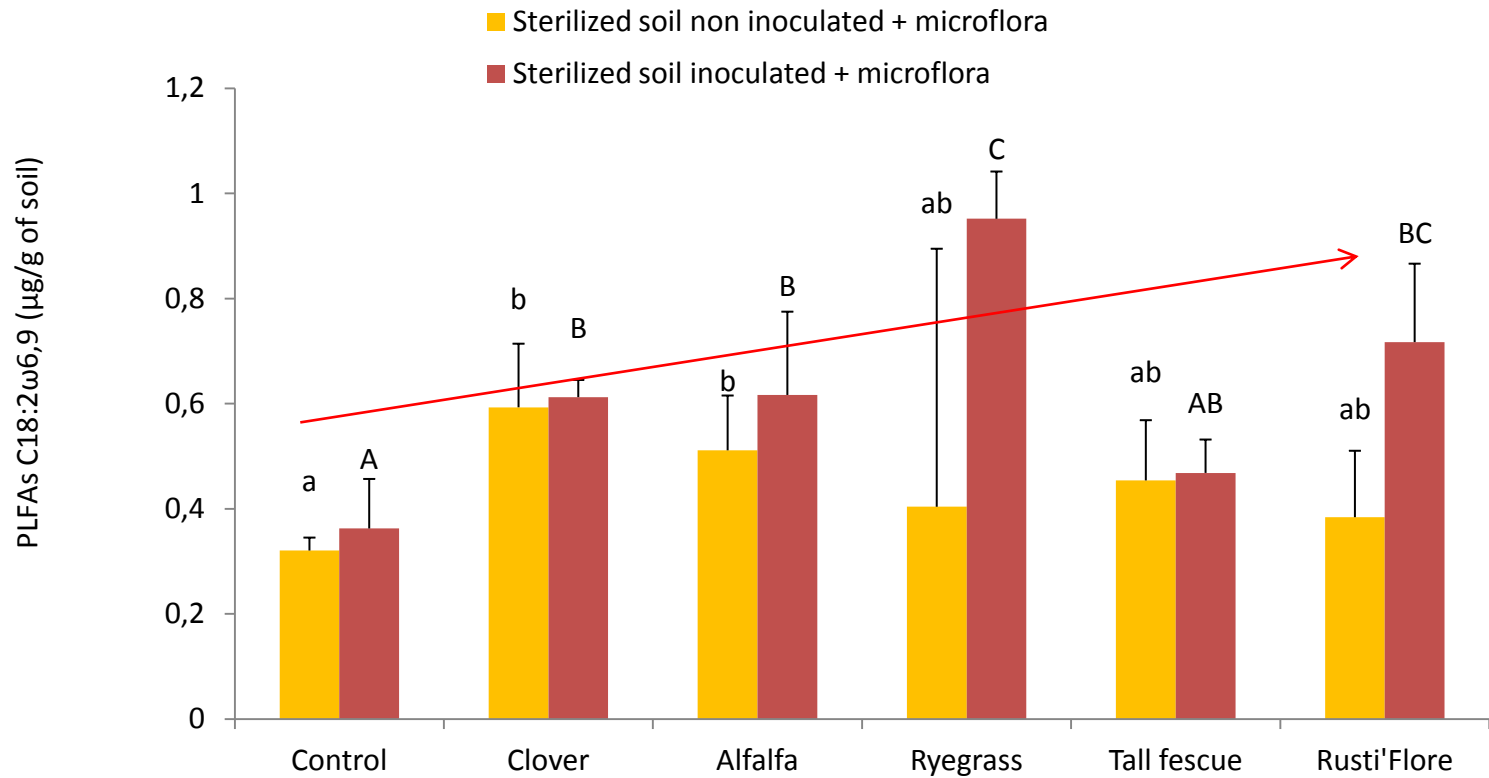
MICROBIAL ACTIVITY IN THE SOIL

➤ Dehydrogenase (DHA)



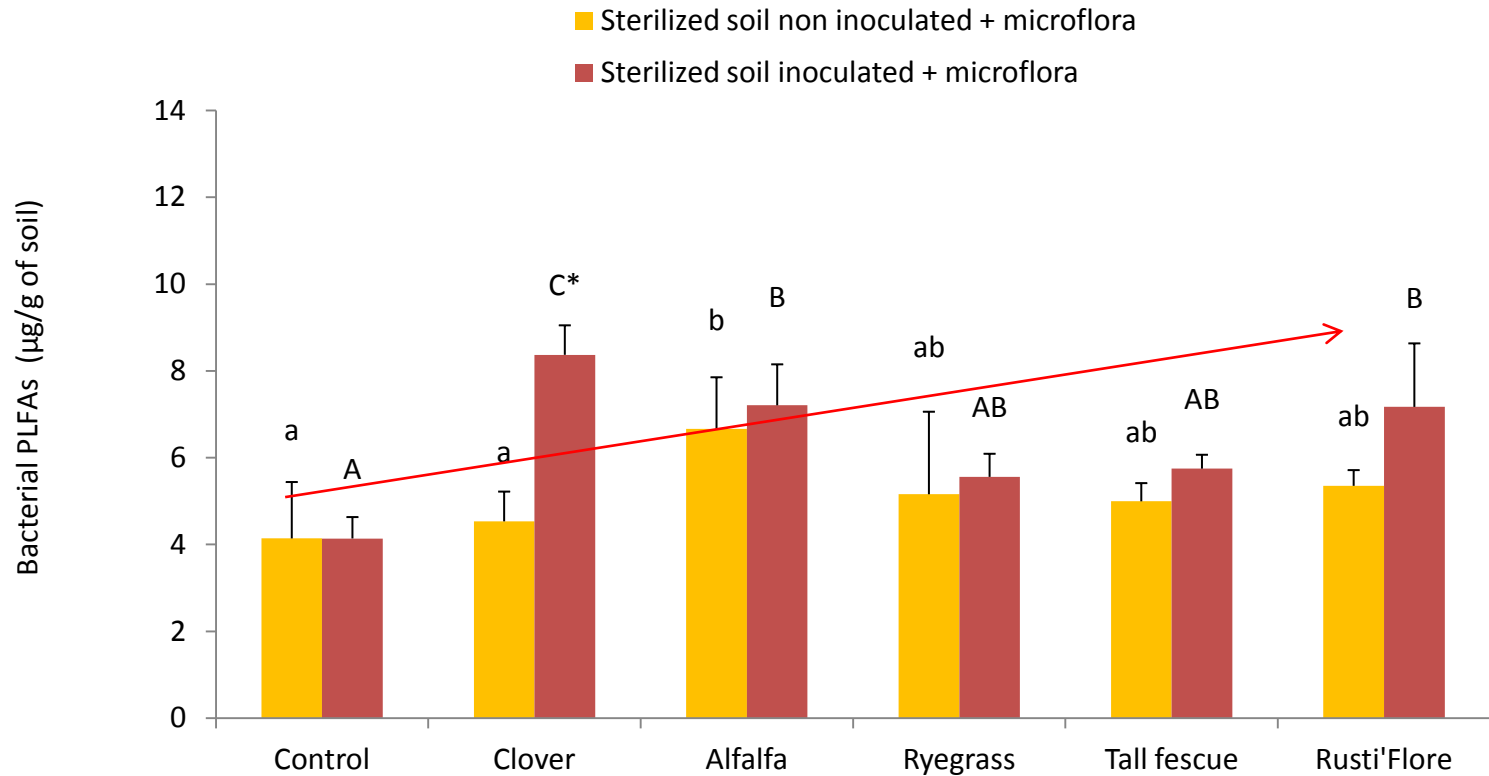
- Stimulation of microbial activity after 20 weeks on vegetated soil

FUNGAL BIOMASS IN THE SOIL



- Stimulation of Fungi biomass after 20 weeks on vegetated soil

BACTERIAL BIOMASS IN THE SOIL



- Stimulation of bacterial biomass after 20 weeks on vegetated soil especially in inoculated Clover

CONCLUSIONS

- The ability of AMF spores to germinate at high concentrations of dioxins/furans.
- The ability of exogenous inoculum to colonize plant roots and improve their growth (Clover) despite the presence of dioxins/furans.
- Dissipation of dioxins/furans after 20 weeks in vegetated soil.
- Stimulation of the microflora (bacteria and fungi) and microbial activity after 20 weeks on vegetated and inoculated soils.

PERSPECTIVES

1

■ Optimizing dioxin degradation conditions :

- Biosurfactant (Rhamnolipid) : to increase the bioavailability of dioxins
- Degrading bacterium capacity : *Sphingomonas wittichii* rw1
- Other mycorrhizal inoculum : The native inoculum (most adapted to pollution)

2

■ Analyzing the microbial communities with metagenomic approaches (microbiome changes before and after phytoremediation)

3

■ Performance evaluation and monitoring of a potential risks through toxicity and ecotoxicity tests



Université Lille Nord de France
Pôle de Recherche
et d'Enseignement Supérieur



*Thank you for your
attention*

Hacene.Meglouli@univ-littoral.fr
(033) 6 68-03-69-25

