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# Soil and crop cover: a potential resource of biocontrol agents against pea root rot caused by *Aphanomyces euteiches*

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# SOIL AND CROP COVER : A POTENTIAL RESOURCE OF BIOCONTROL AGENTS AGAINST PEA ROOT ROT CAUSED BY *APHANOMYCES EUTEICHES*

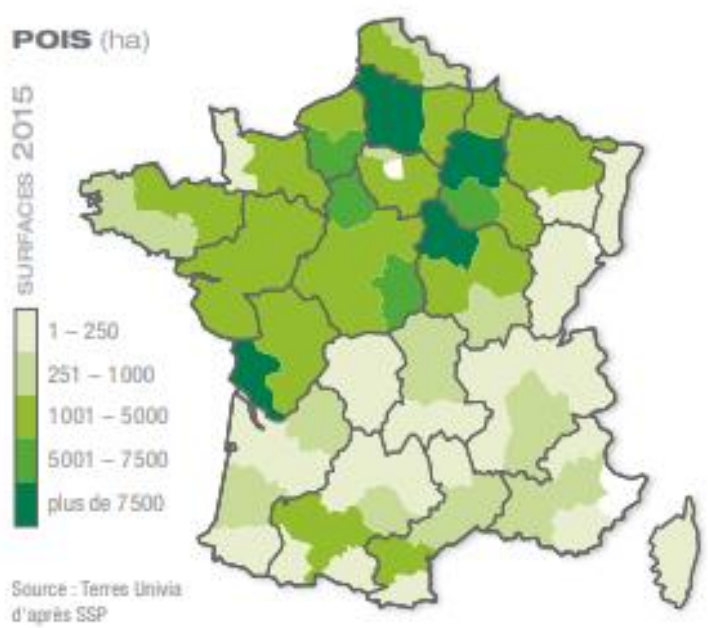
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## Introduction & Context : why research a treatment to avoid Pea Root Rot?

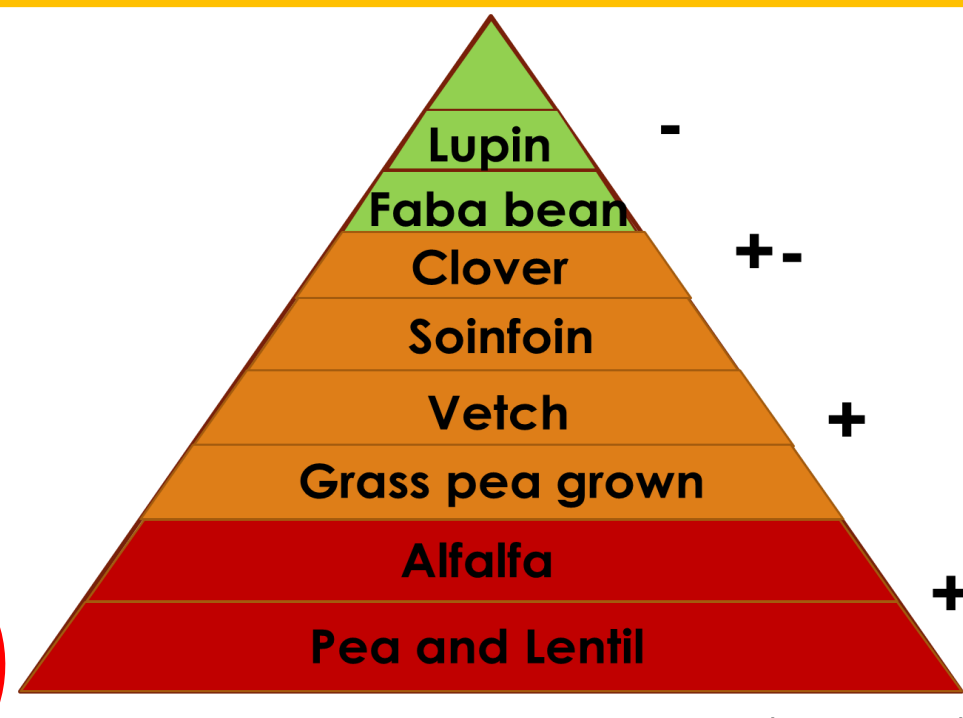


**Fabaceae** : able to fix atmospheric nitrogen  
✓ Numerous nutritional quality: High input of protein  
✓ Environmental preservation: reduction of nitrogen inputs and decrease of N<sub>2</sub>O



Peas : one of the most cultivated crops in France

But !



Pea root rot : field & Plant  
Pea must project\_INRA & Arvalis

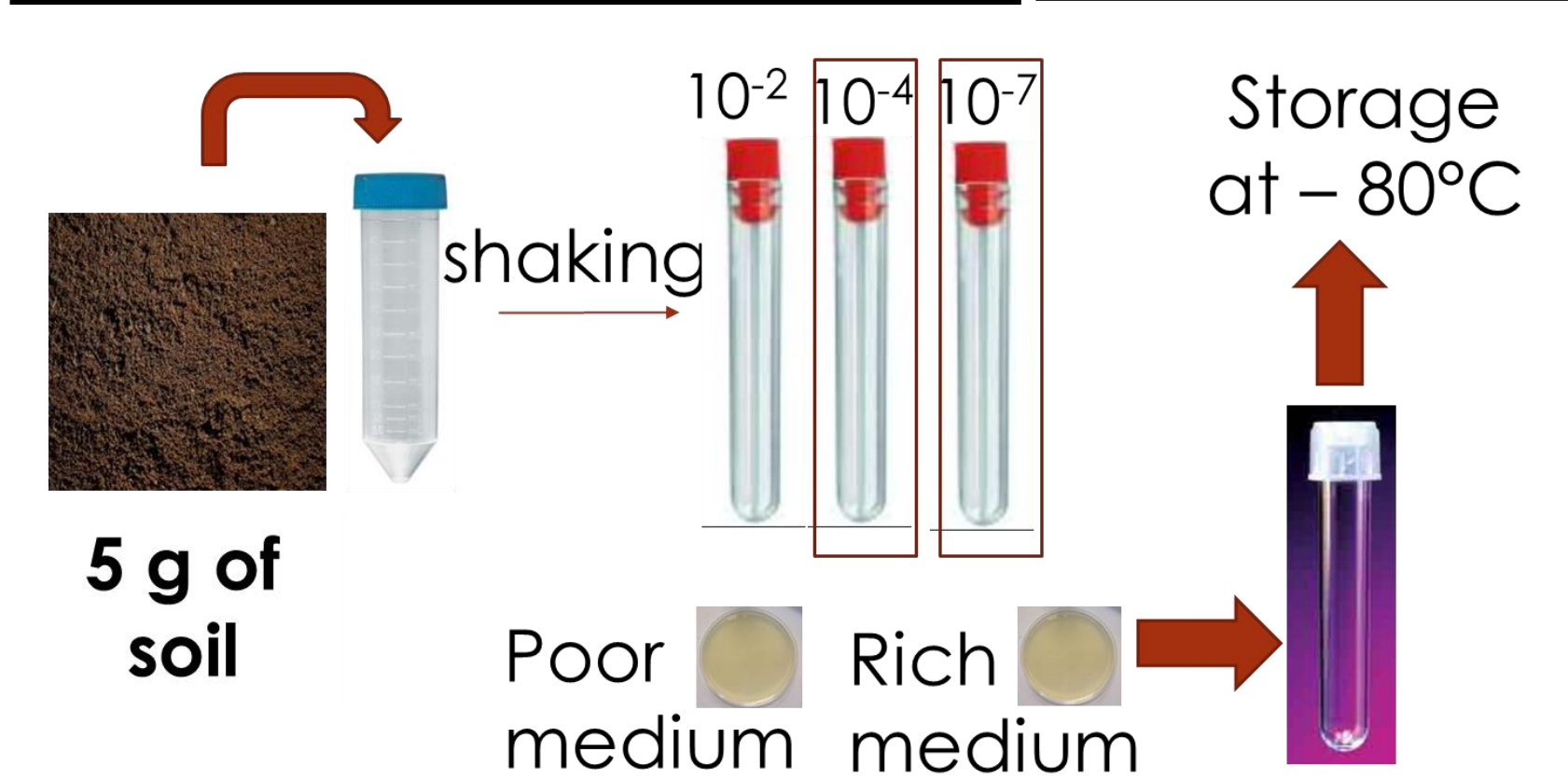
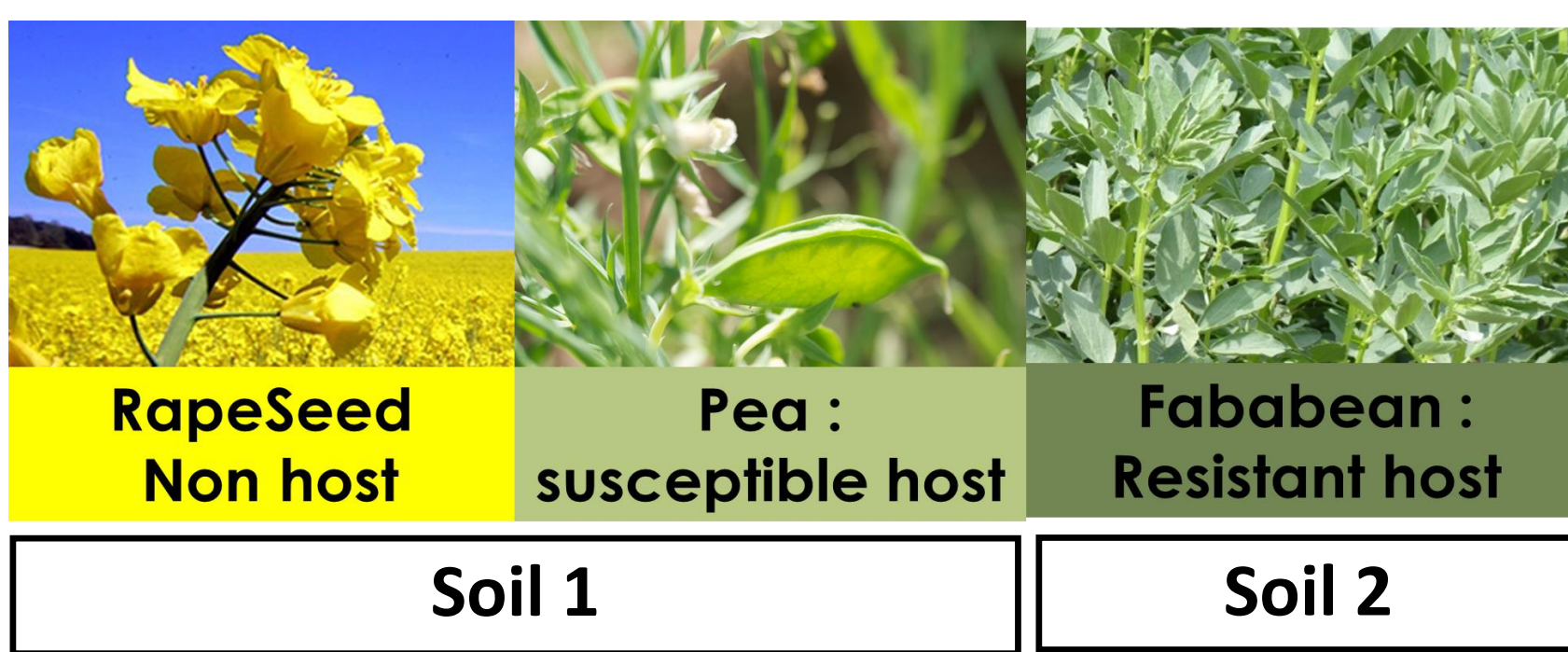
No available efficient treatment  
Potential of total yield loss for farmers  
Requirement to find some solutions !

PESTICIDE FREE

Rhizosphere: Reservoir of potential Biological Control Agents (BCAs)  
A potential alternative against *A. euteiches*

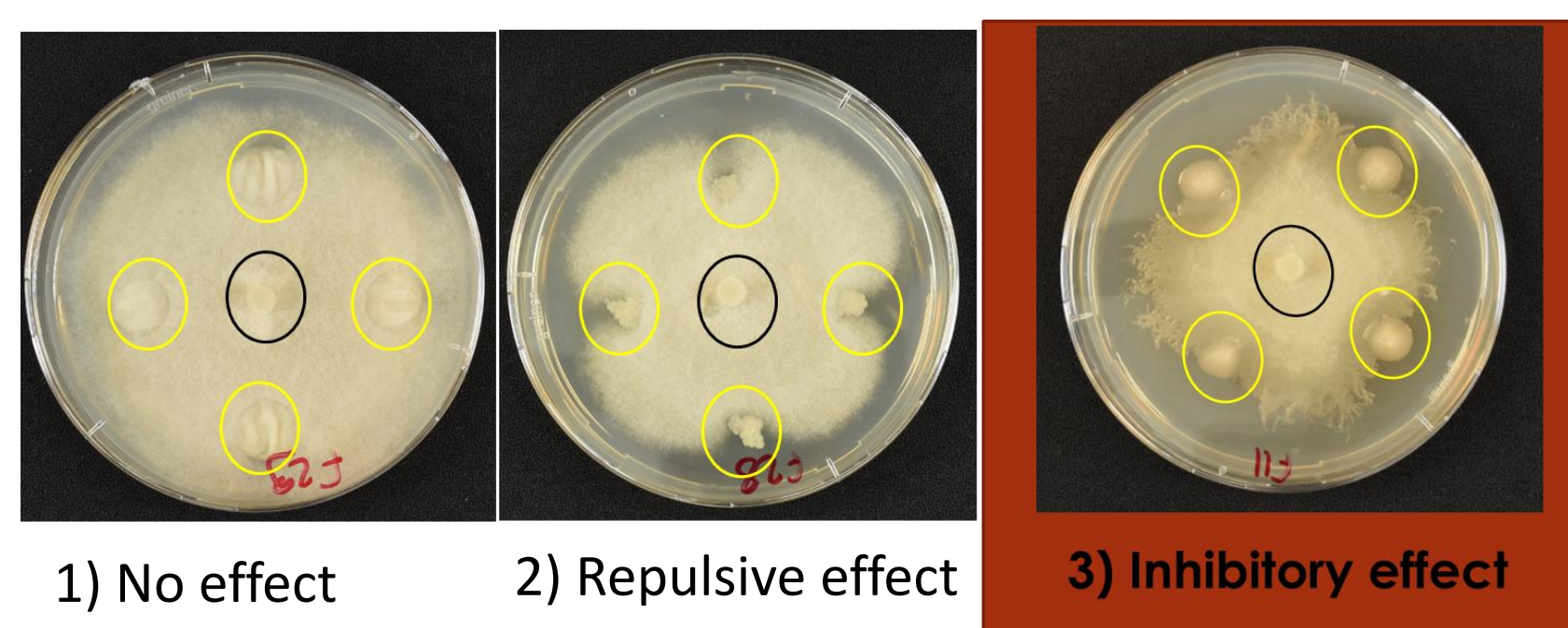
## Material & Method : examination of possible biological treatments

### 1) Extraction of bacteria from soil (depth 0-10 cm) under 3 cover crops



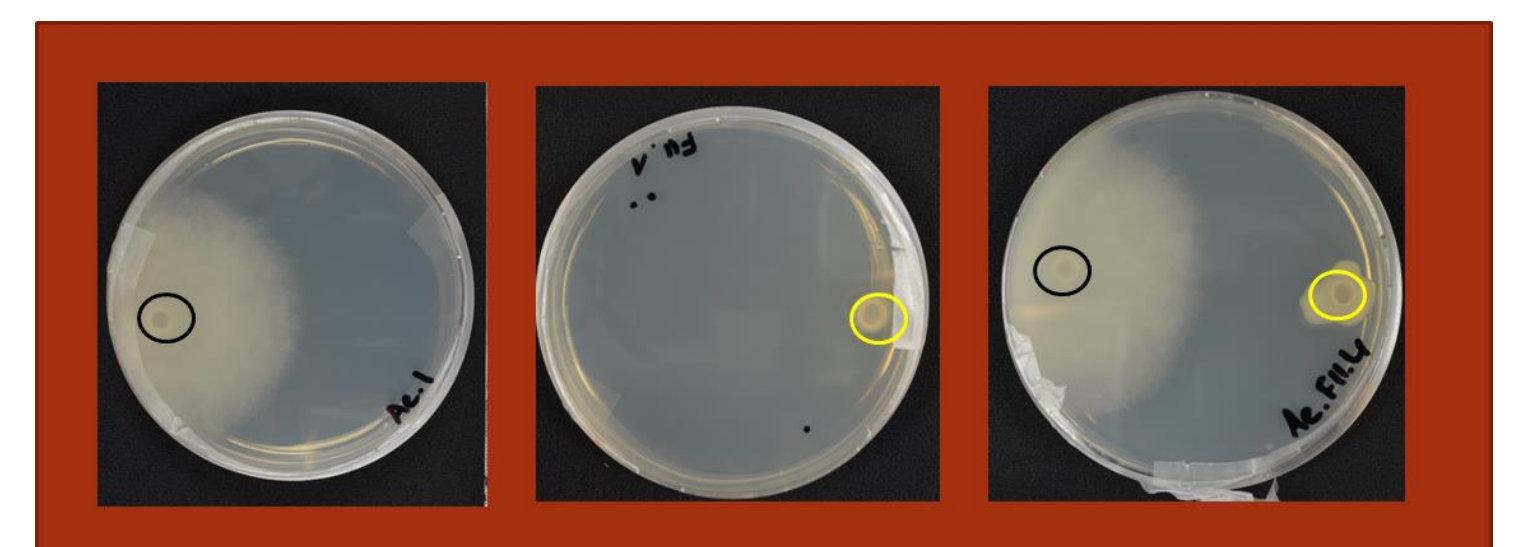
### 2) Screening

On PDA (preferential growth medium for *A. euteiches*) 4 x 1 µL of 48h bacterial culture (yellow ring) were inoculated on agar plate at 2,5 cm around the 5 mm plug of *A. euteiches* (black ring).



### 3) Confrontation tests

On PDA, 5 mm-plug of *A. euteiches* were opposed to 5 mm-plug of bacteria



Mycelium growth measurement from pictures analysed with ImageJ

### 4) Characterization of strains : DNA sequencing (16S), detection of specific enzymatic activities (cellulase [3], protease [4], chitinase [5]).

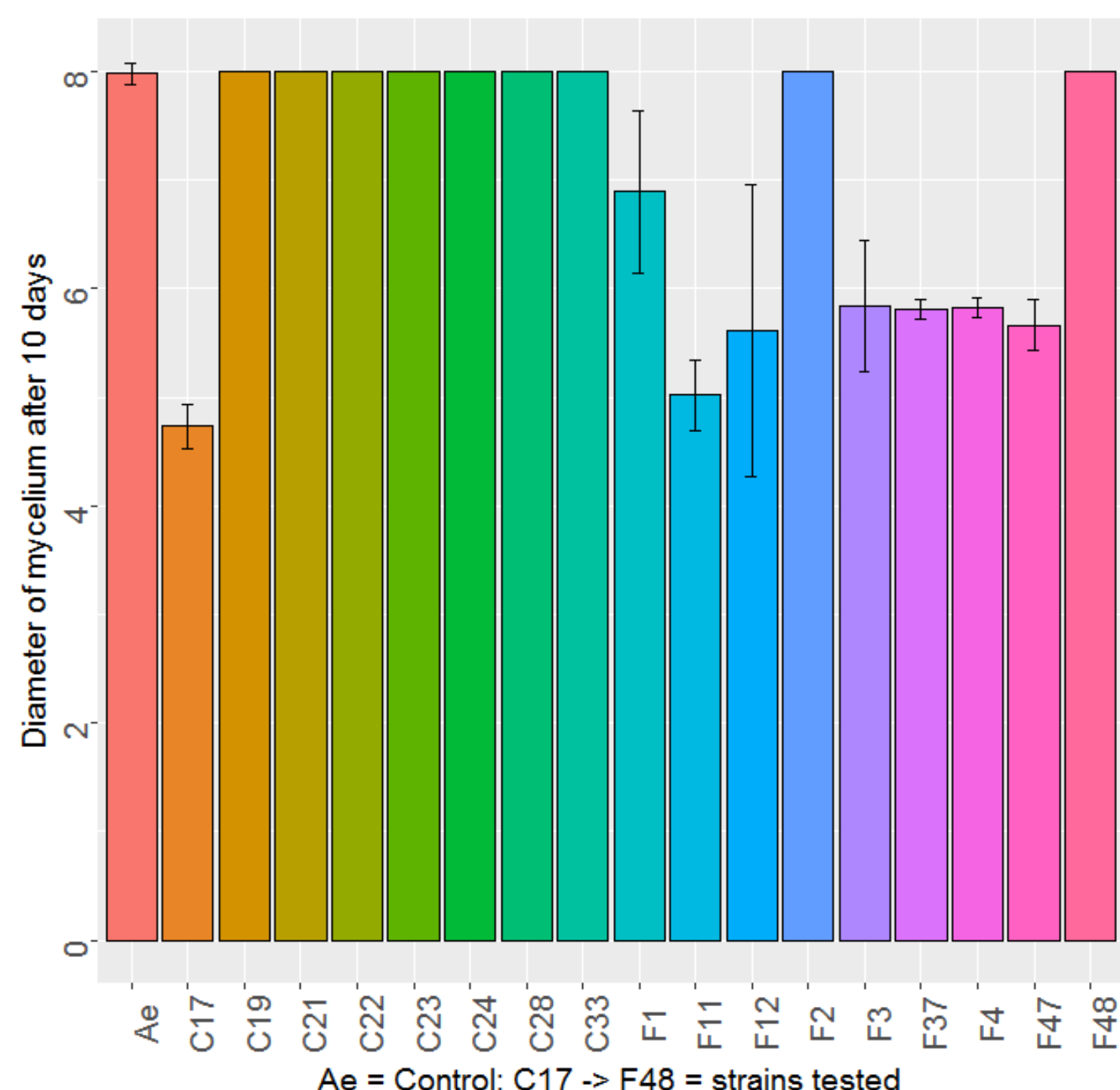
## Results & Analyses : isolation of soil biological control agents (BCA)

### Screening : 17 bacterial strains with inhibitory effect on *A. euteiches*

SOIL	number of strains per cover crop		
	1) No effect	2) Repulsive	3) Inhibitory
Rapeseed	39	2	9
Faba bean	37	6	7
Pea	39	10	0

### Confrontation tests : 8 out of 17 strains reduced *A. euteiches* mycelium

-> Difference in mechanisms of action? (competition/metabolite production)



### Preliminary characterization of 17 strains with inhibitory effect

Cover crop	Id	Molecular identification	Cellulase	Protease	Chitinase
Rapeseed	C17	<i>Pseudomonas sp.</i>	+	++	-
	C19	<i>Bacillus sp.</i>	-	++	-
	C20	<i>Bacillus sp.</i>	-	+/-	-
	C21	<i>Bacillus sp.</i>	-	++	-
	C22	<i>Bacillus sp.</i>	-	++	-
	C23	<i>Bacillus sp.</i>	-	+/-	-
	C24	<i>Bacillus sp.</i>	-	+/-	-
	C28	<i>Bacillus sp.</i>	-	+/-	-
	C33	<i>Bacillus sp.</i>	-	++	-
	Faba bean	F1	<i>Bacillus sp.</i>	-	++
F3		<i>Acinetobacter sp.</i>	-	-	-
F4		<i>Bacillus sp.</i>	-	++	-
F11		<i>Pseudomonas sp.</i>	-	++	+
F12		<i>Bacillus sp.</i>	-	+	-
F37		<i>Pseudomonas sp.</i>	-	-	-
F47		<i>Pseudomonas sp.</i>	+	++	+
F48		/	-	+++	-

For the same soil (soil 1), none of the strains isolated under pea cover showed an inhibitory effect while 9 potential biocontrol agents were found under rapeseed cover.

-> Impact of cover crop on abundance and diversity of BCA against *A. euteiches*? [6,7]

## Conclusion & Perspectives : biological control agents *in vivo*

- 149 strains tested : 17 out of 149 reduced currently *A. euteiches* mycelium growth.
- 17 strains with inhibitory effect : principally *Bacillus* (8/9 rapeseed soil, 3/7 faba bean soil) *Pseudomonas* (1/9 rapeseed soil, 3/7 faba bean soil) and one *Acinetobacter* (faba bean soil). Further characterization studies are in progress (growth, quantitative measurement of enzymatic activities)
- Mechanisms of action against *A. euteiches* : remain to be accurately elucidated.
- Major effects of cover crop on microbial community and so BCAs : metabarcoding in progress...
- Efficiency measurement of each potential BCA against *A. euteiches* : plant tests with artificial contamination (in controlled conditions)