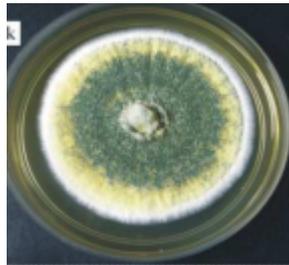


# One health under the context of environmental transition: a case study with project ALCOV

N. Dupuy, T. Orsière, A.M. Farne

# Biodiversity valorisation to help environmental transition



**Filamentous fungi have high enzymatic activities and a large spectrum of metabolites with multiple potentials**

**Good candidates for biotechnological applications**

Introduction

# Solid State Fermentation (SSF) a way to produce active metabolites

## Generalities and advantages of SSF

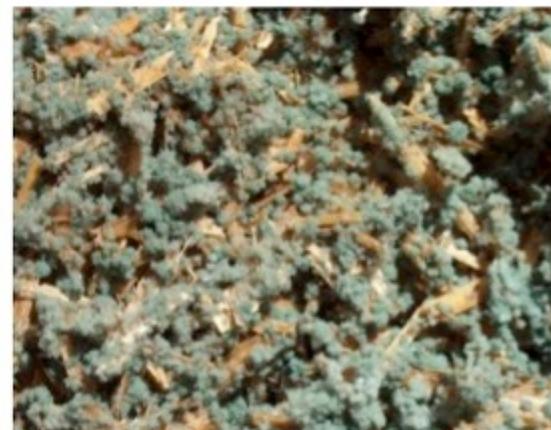
Solid state fermentation (SSF), is a biotechnological process involving the use of solid material under controlled conditions in the absence of free running water.

FMS is an ancient process, initially used in human food.

### Typical SSF

*Penicillium roquefortii* for cheese production

*Aspergillus oryzae* to initiate the koji



Growth of *Trichoderma* sps. on wheat straw in Solid State Fermentation

# Solid State Fermentation (SSF) a way to produce active metabolites

## Generalities and advantages of SSF

### Benefits

- Promotion of agro-industrial co-products as a growing medium.
- Improved quantitative and qualitative production.
- Absence of a liquid phase and substrate low humidity allow reduced volumes of fermentation reactors.

### Disadvantage

- Monitoring of fermentation more difficult parameter control (pH, water content, temperature=).
- Difficulty in increasing the production scale.

Metabolic heat  
CO<sub>2</sub>

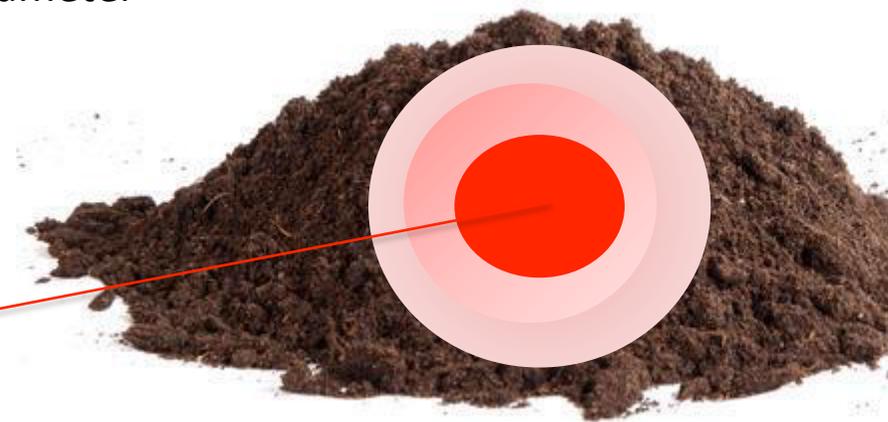
valorization of agricultural byprod



advantages

Economical

Environment



**Solid State  
Fermentation  
(SSF)  
a way to  
produce  
active  
metabolites**

## Agroindustrial wastes as substrate /support on SSF

Agro-industrial wastes are generally considered as the best substrates for SSF and enzyme production, because it provides the necessary nutrients for microbial growth.

→ The most used substrates are plant wastes:



**Sugarcane bagasse**



**Wheat flour**



**Oil palm wastes**



**Coconut fiber**



**Beet pulp**



**Coffee wastes**



**Wheat bran**



**Apple pomace**

## Biological control agents: an improved alternative

### Definition biopesticides:

- ➔ Formulations based on biological control agents (BCAs) such as bacteria, fungi, viruses, and their combinations.
- ➔ It is “the use of living organisms to prevent or reduce pest damage”
- ➔ It is an exploitation by man and for his benefit of a natural relationship between two living beings.
- ➔ The target is undesirable organism, crop pest, weed, livestock parasite...
- ➔ The control agent (or auxiliary) is an organism most often a parasite (or parasitoid), a predator or pathogen of the former, which kills it more or less quickly by feeding on it or at least limiting its development. It can be a competitor (autocid struggle).



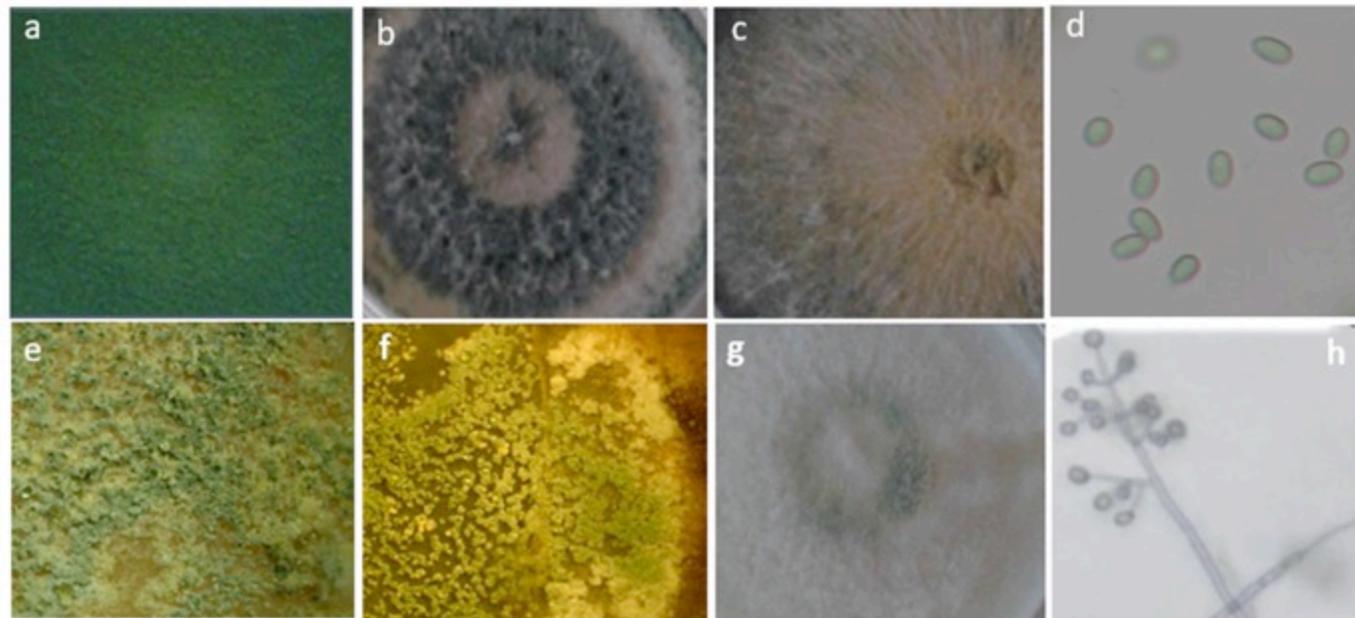
Lady beetle (Beetle) among its prey consisting of aphids (Homoptera). © Photo C. Slagmulder/INRA

## Microorganisms used in biopesticides production

### *Trichoderma* strains

*Trichoderma* genus are an important fungus as a biopesticide, these fungi could be a good model for biocontrol application.

They are well known producers of secondary metabolites (SMs) with a direct activity against phytopathogens and compounds that substantially affect the metabolism of the plant, they contributing as high as 50% of BCA's fungi.



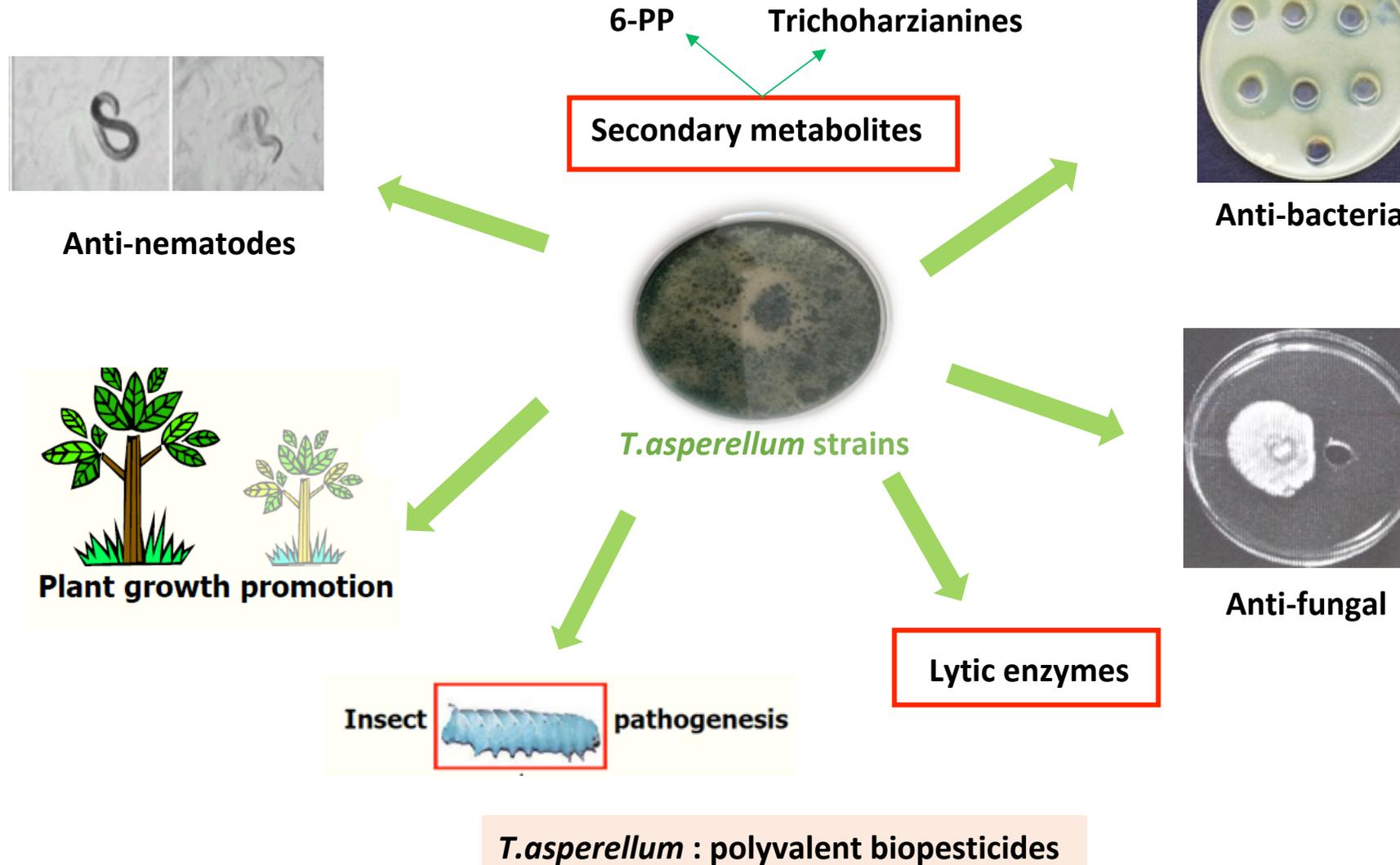
a) *T. viride*, b) *T. asperellum* DWG3, c) *T. harzianum*, d) conidia of *T. asperellum*, e) *T. longibrachiatum*, f) *T. longibrachiatum* during confrontation with *Fusarium* strain, g) *T. asperellum* Tv 104, h) *T. harzianum* germination.

Nowadays, the most important species in this field are *T. asperellum*, *T. harzianum*, *T. virens*, *T. viride* and *T. atroviride*.

amentous  
fungi

## Trichoderma strains

Which are potentially useful biocontrol activities?



amentous  
fungi

Biopesticides are part of the fight against plague organisms and are based on the use of agents or factors related to life.

Characteristics of biopesticides :

Very specific action

Biodegradable biological material

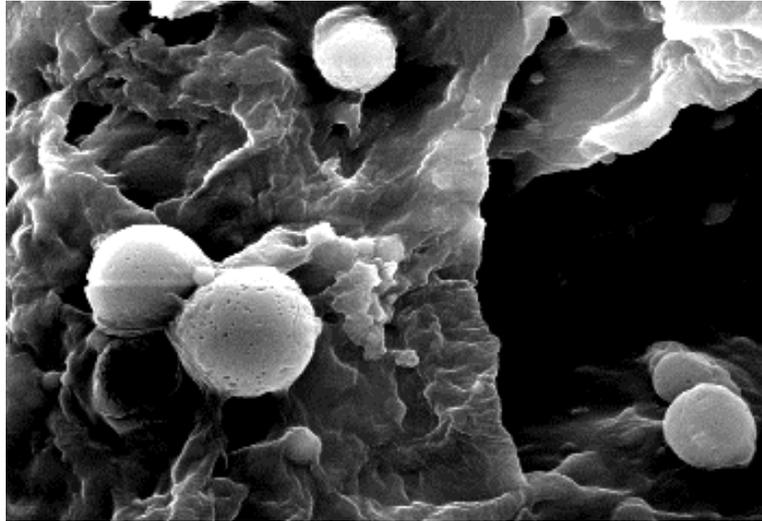
Non-toxic to pollinating insects



Biopesticides: Microorganisms (bacteria, filamentous fungi, etc.) insects

amentous  
fungi

# Optimisation of F conditions



Support



Wine shoots

Starch



Potatoes flour

Amino acids



Jatropha cake



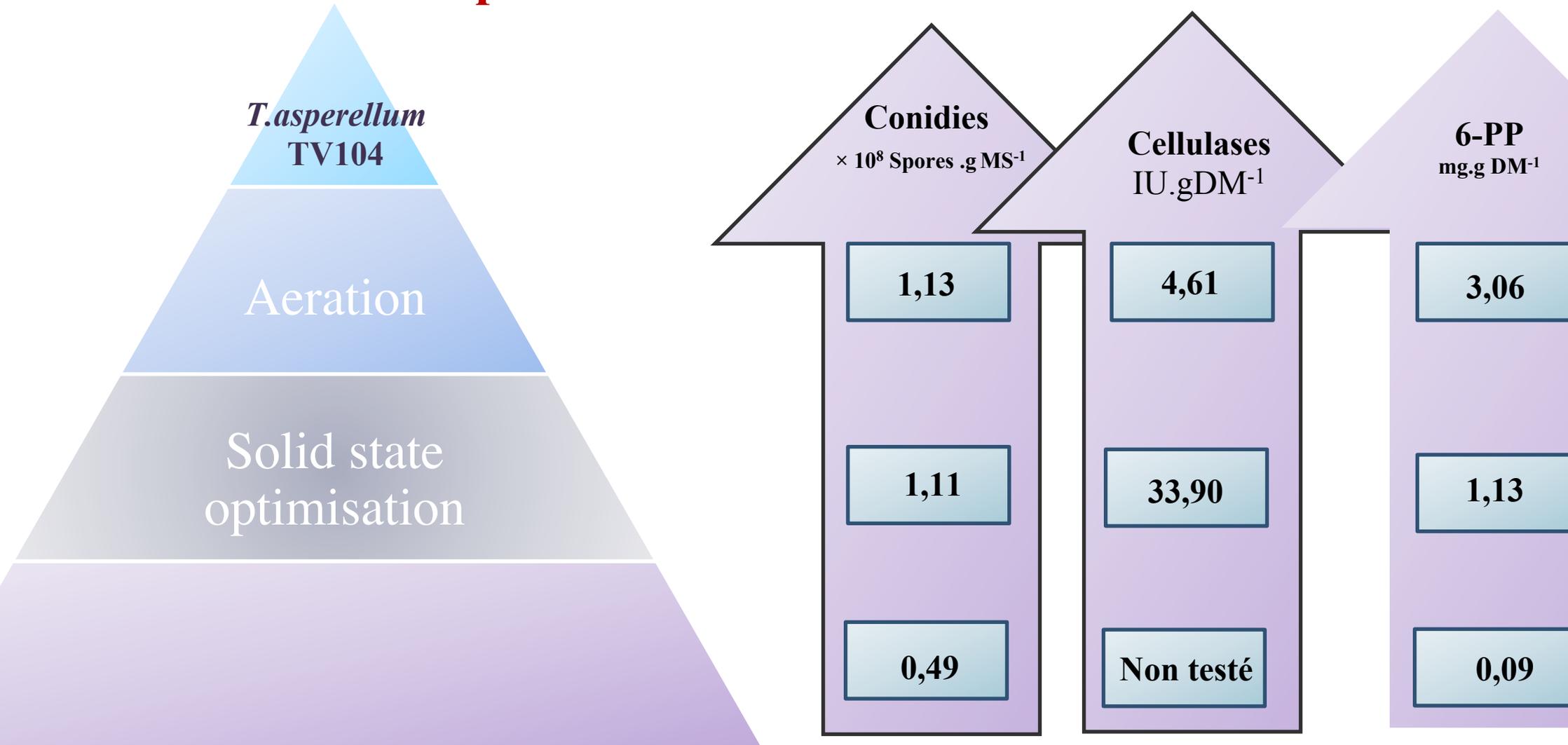
Olive cake

Oil and enzymatic precursor



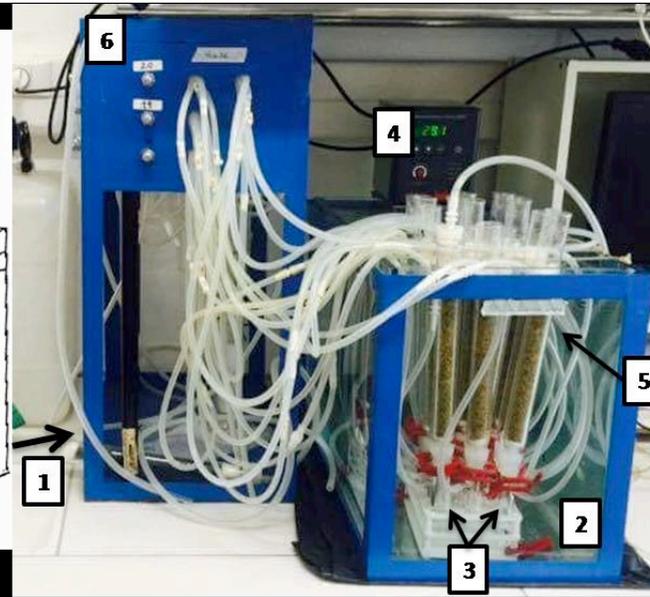
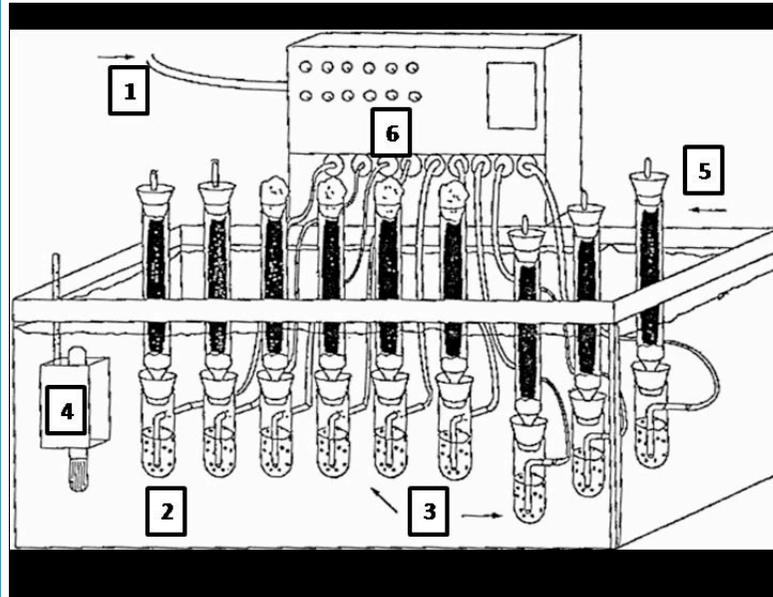
Olive oil

# Optimisation results



Solid State  
Fermentation  
Extrapolation  
large scale-  
up

## 1) Raimbault columns : conidia production



- 1- Pressurized air
- 2- Water bath incubator
- 3- Humidified air
- 4- Thermoregulator
- 5- Packed bed column
- 6- Air flow regulation system



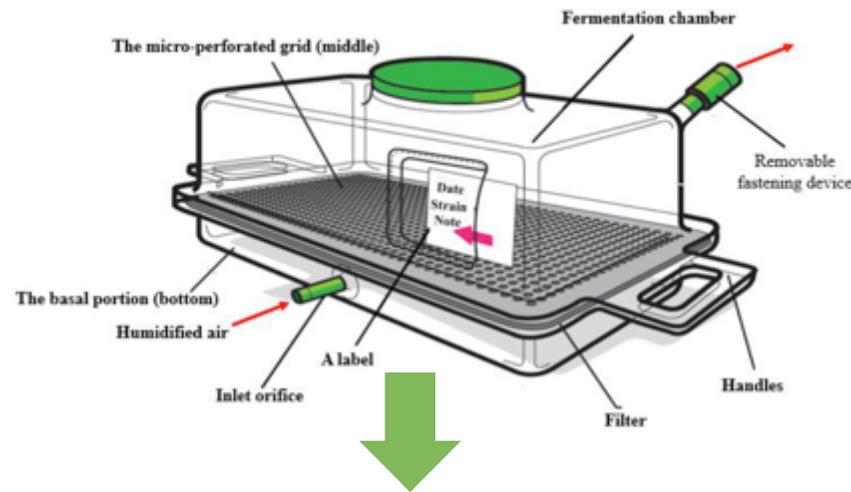
*Beauveria bassiana*



*Trichoderma asperellum*



# One use static bioreactor



Bioreactor filled with waste residues and fungal conidia



Fermented substrate :Biopesticide product

Solid State  
fermentation  
Extrapolation  
of large scale-  
up



6-Pentyl-alpha-pyrone "6-PP" (antifungal compound)

Lytic enzymes ( Cellulases, chitinase, lipases...)

Spores (virulent form)

Maïga Y, Carboué Q, Hamrouni R, Tranier MS, Ben Menadi Y, Roussos S, Development and Evaluation of a Disposable Solid-State Cultured Packed-Bed Bioreactor for the Production of Conidia from *Trichoderma asperellum* Grown Under Water Stress. Waste Biomass Valorization <https://doi-org.lama.univ-amu.fr/10.1007/s12649-020-01210-2>



# Project ALCOVE:

A research and teaching project for a change of paradigm in winegrowing

*Project manager: AM FARNET DA SILVA*

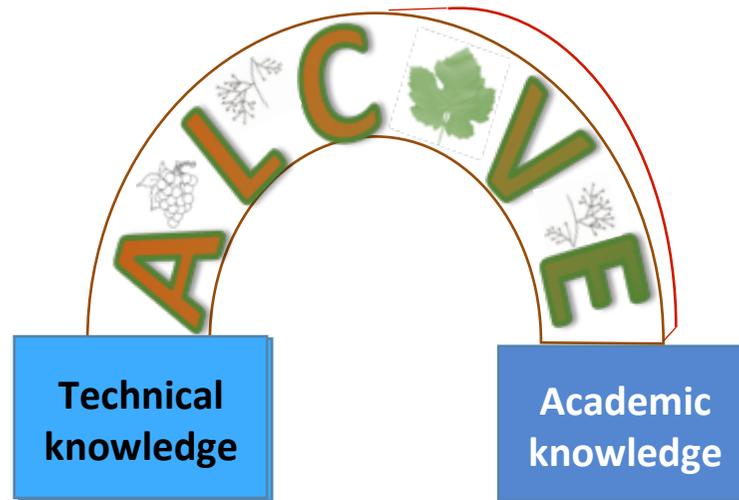


12 EC/C, 6 Tech and  
IE

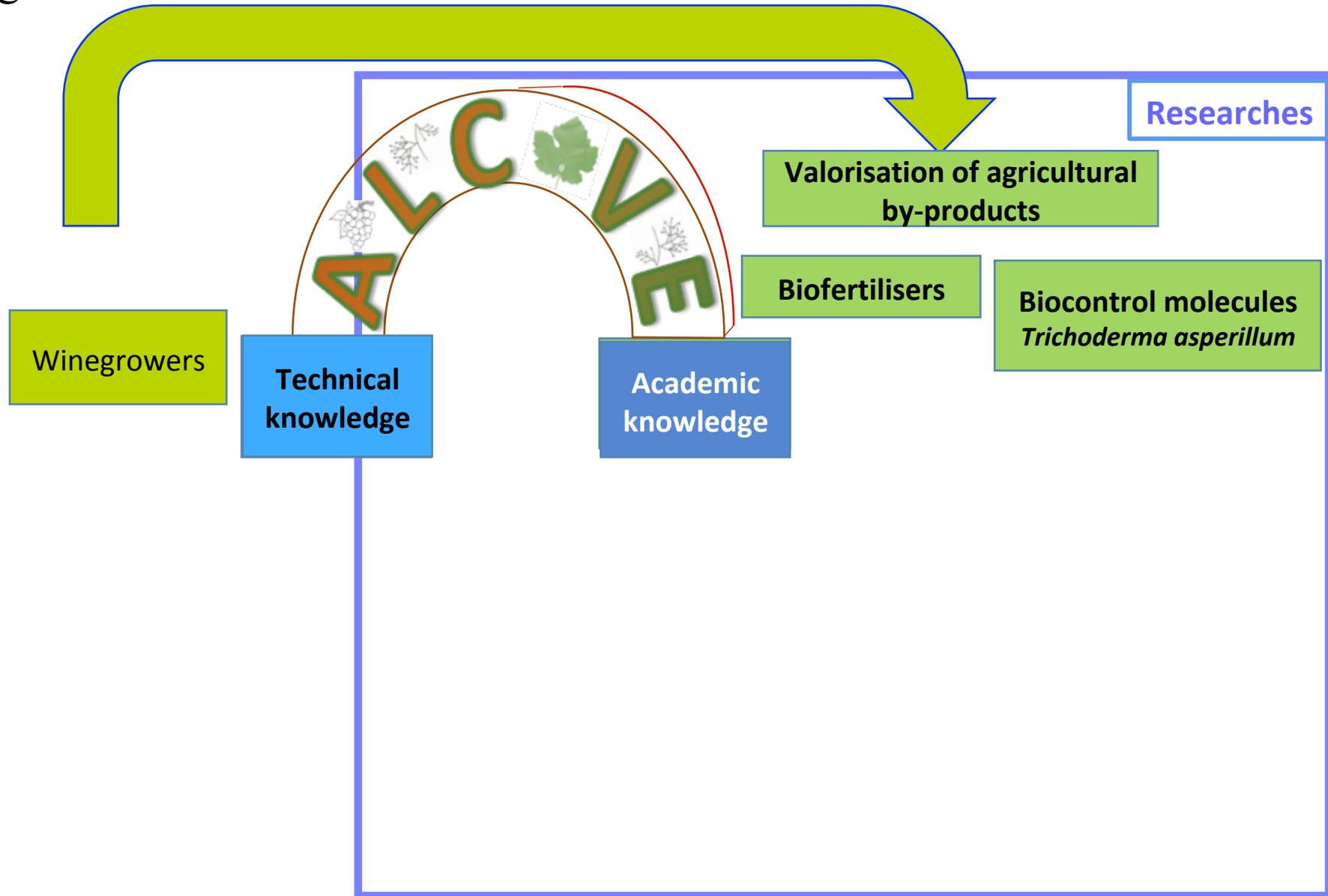
1 Post Doc, 1 PhD



# AgricuLture bioContrôle biOfertilisant ViticolEs

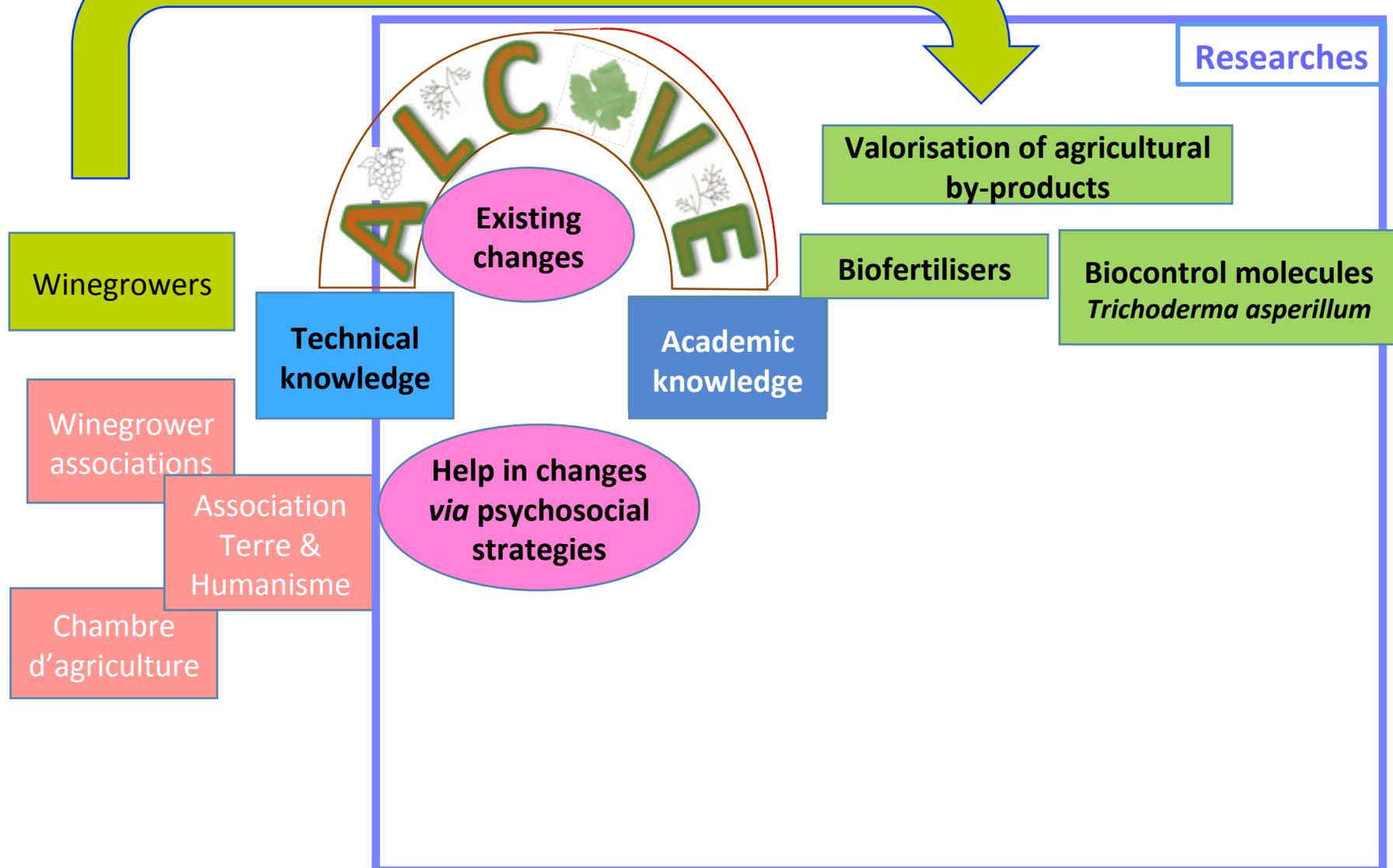


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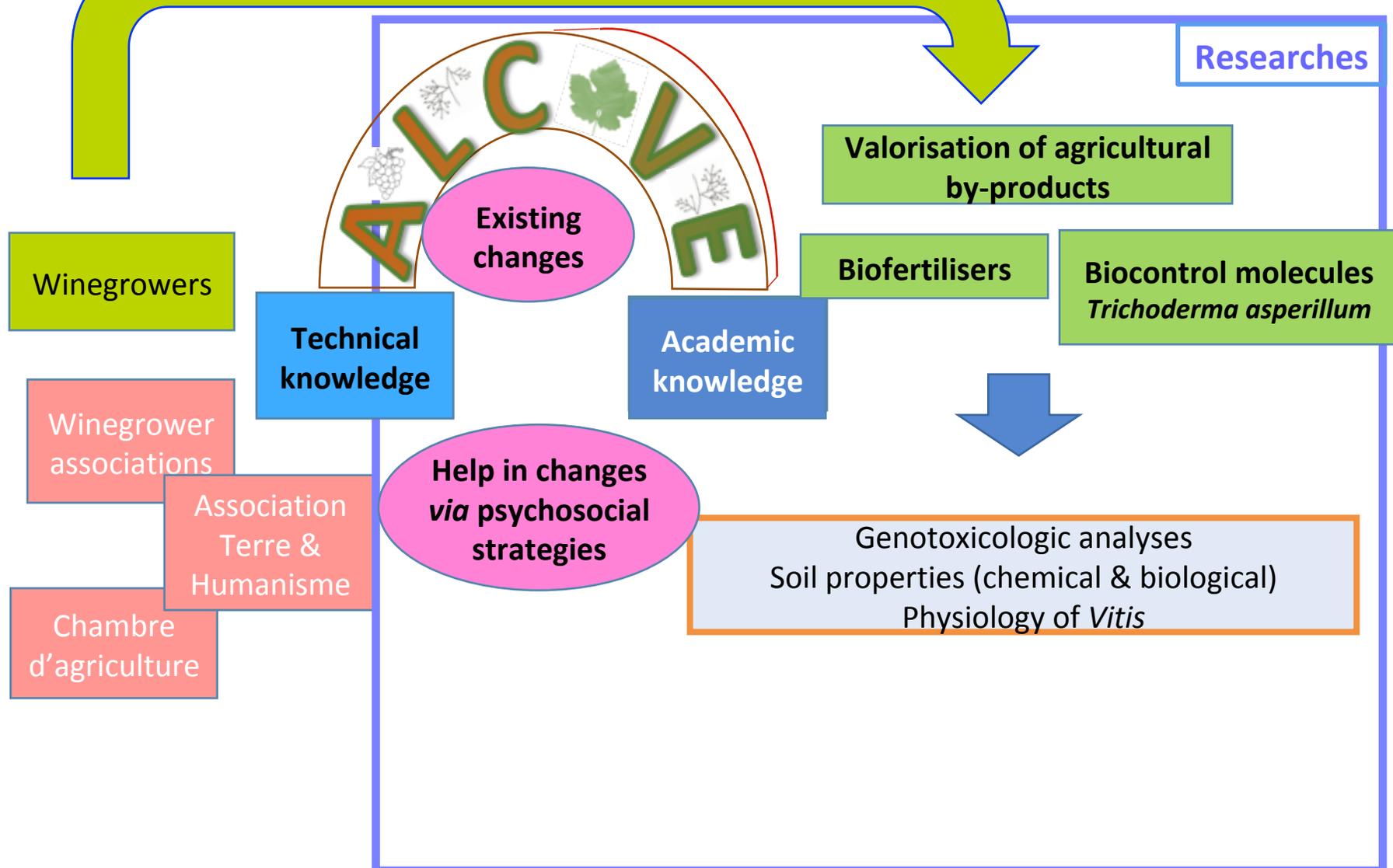
# AgricuLture bioContrôle biOfertilisant ViticolEs

Society



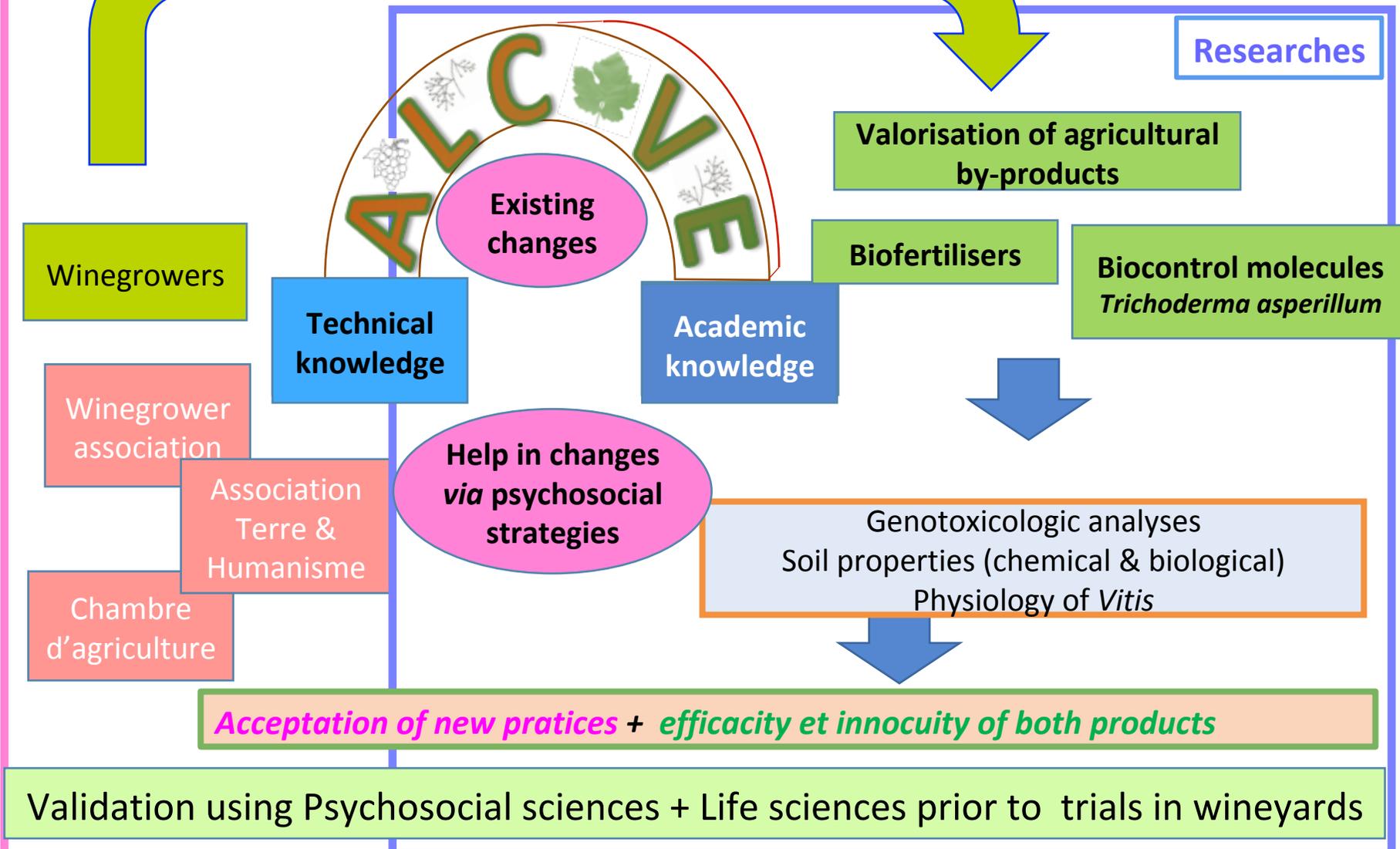
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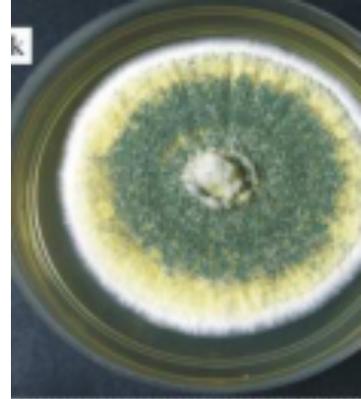
# ALCOVE ORGANIZATION

Task 1 : To produce **biocompounds with high antifungal activities** against phytopathogens of *Vitis* (N. Dupuy)

Task 2 To produce a **bioamendment** from fermented forest litter (P. Christen)



Valorisation of agricultural by products

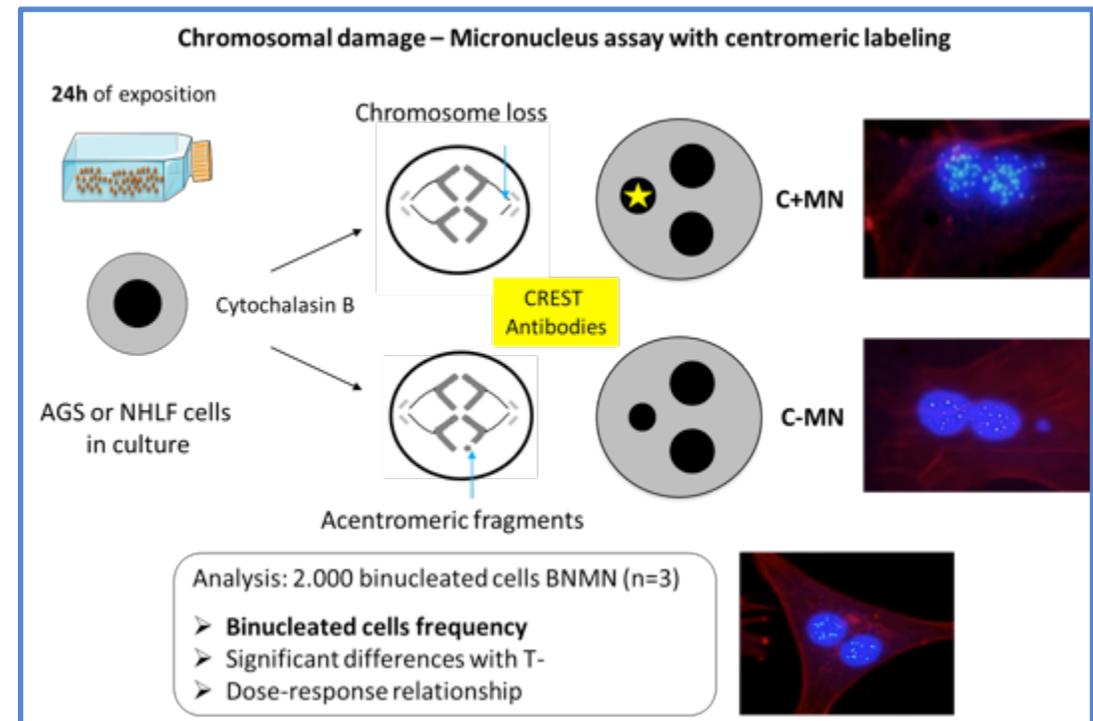


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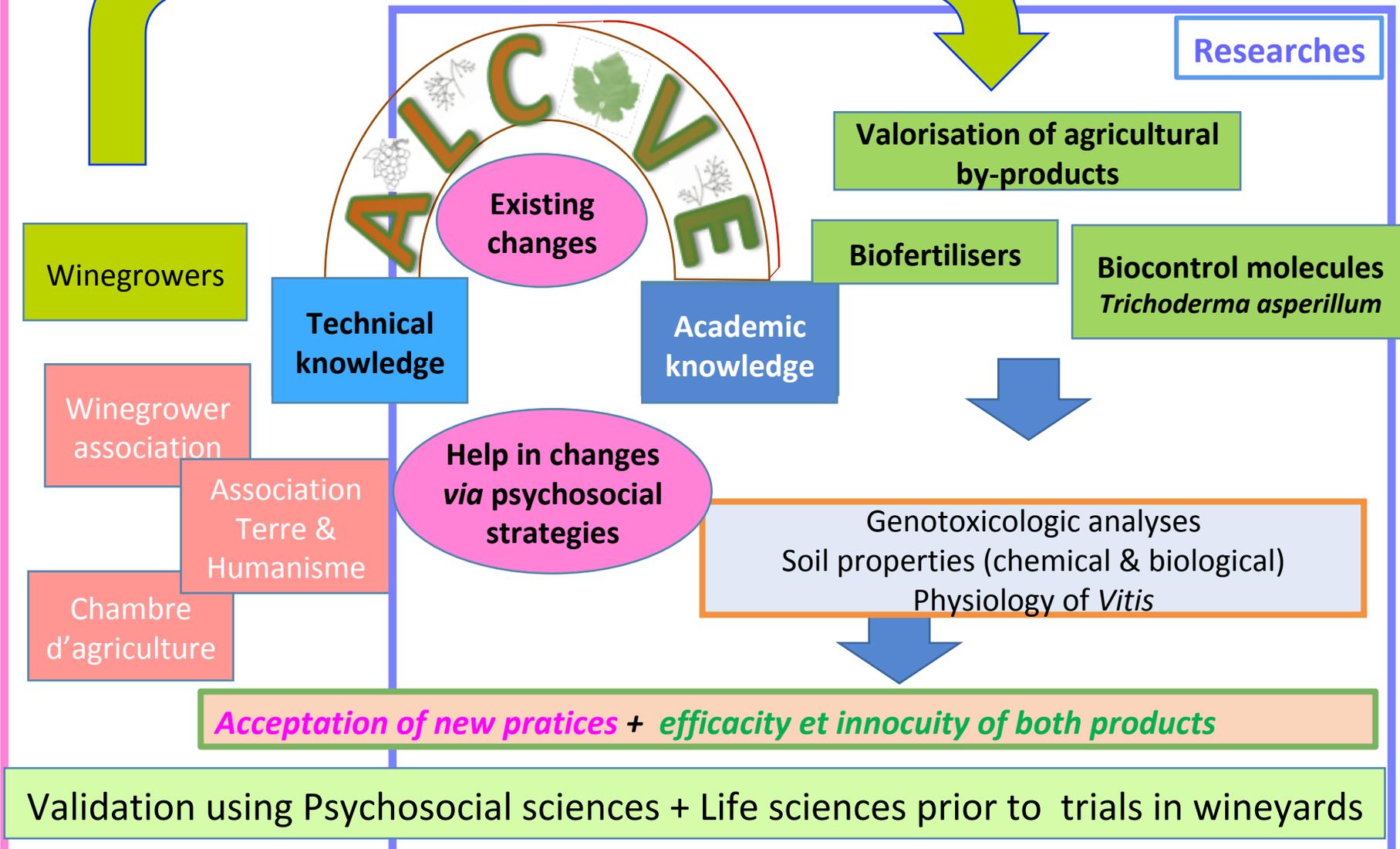
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Task 6 : **Management of the interdisciplinarity** of the project (AM Farnet Da Silva & I. Laffont Schwob)

# AgricuLture bioContrôle biOfertilisant ViticolEs

Society



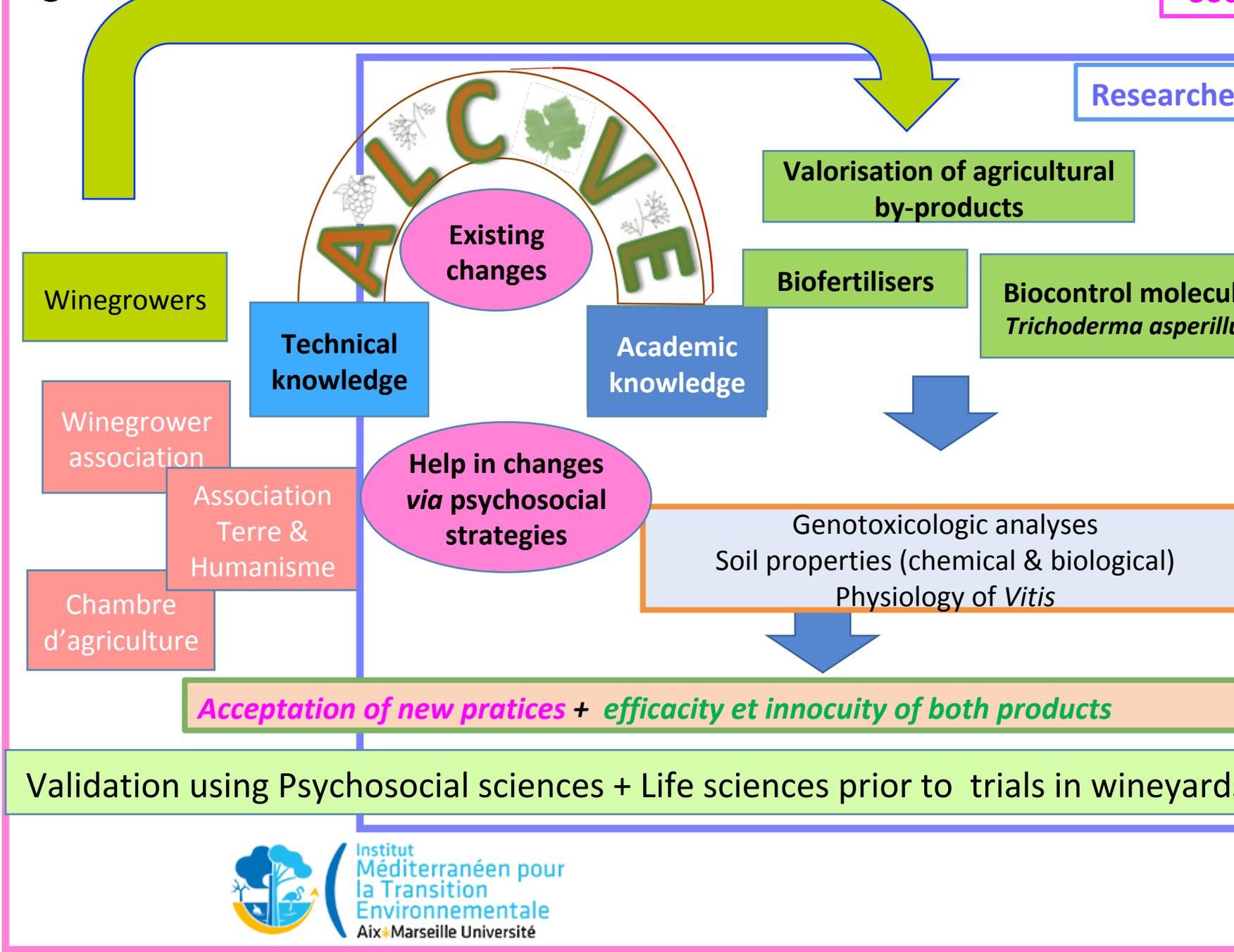
One Health  
in the  
Environmental  
Transition  
Context

Experiences using  
above and other  
projects

Thierry Orsière  
IMBE

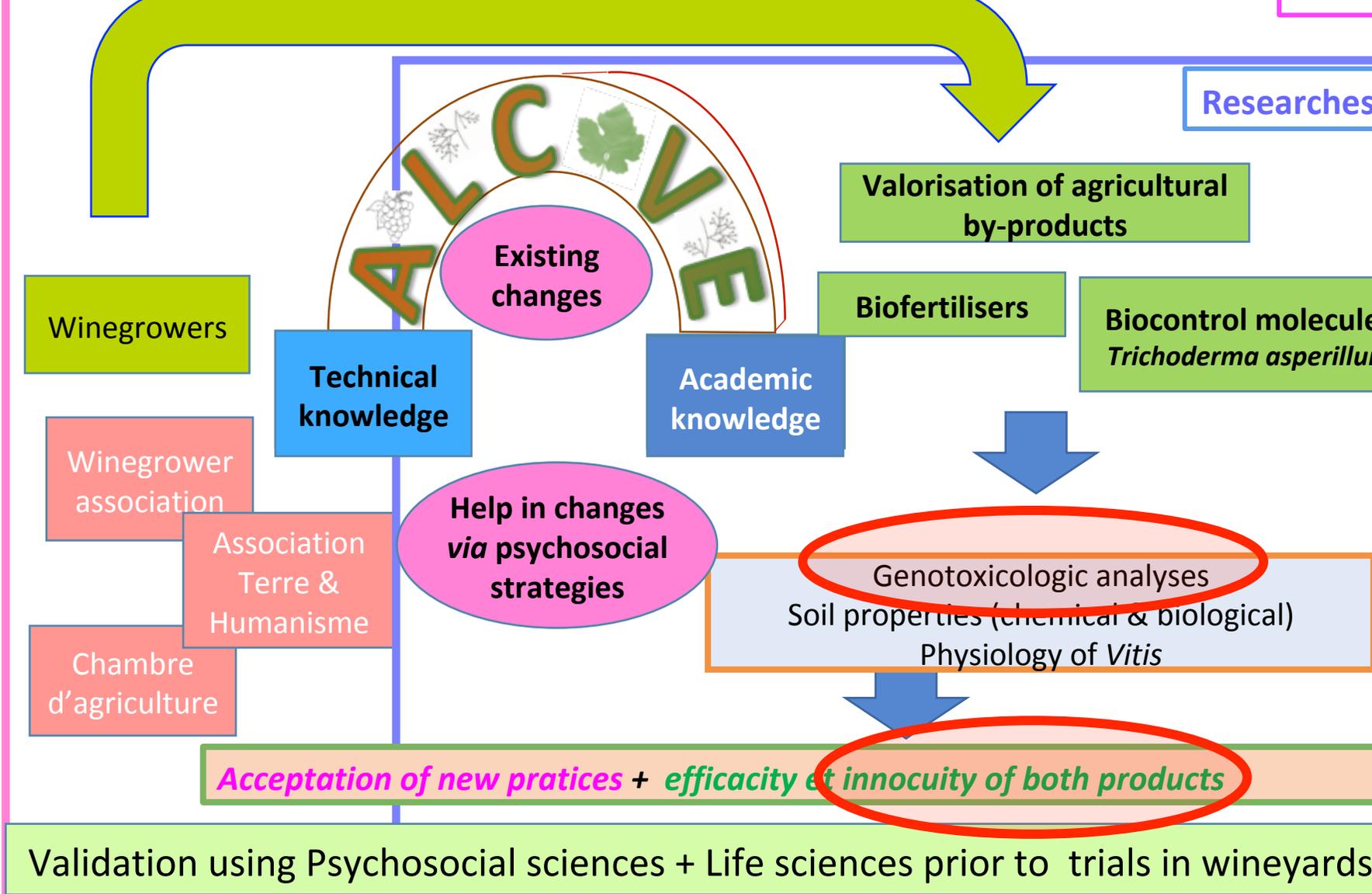


# Agriculture bioContrôle bioFertilisant Viticoles

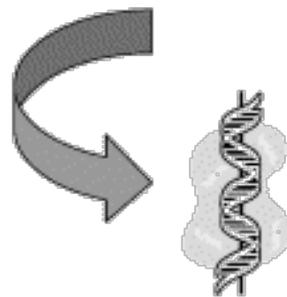


One-Health project should address the human safety and new strategies

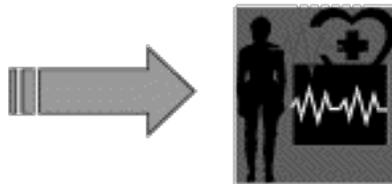
# Agriculture bioContrôle biOfertilisant ViticolEs



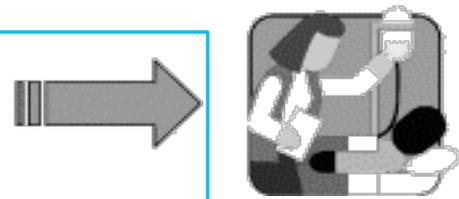
Why are we so concerned by neurotoxic effects?



EARLY BIOLOGICAL EVENTS



LATE BIOLOGICAL EVENTS



CLINICAL DISEASE

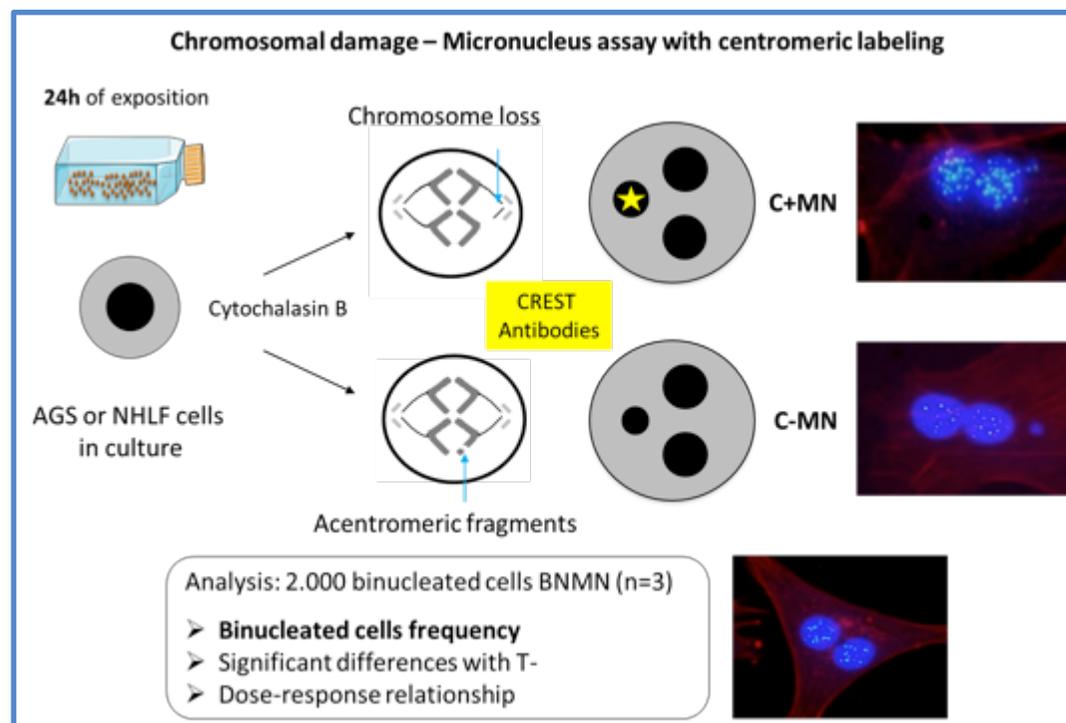
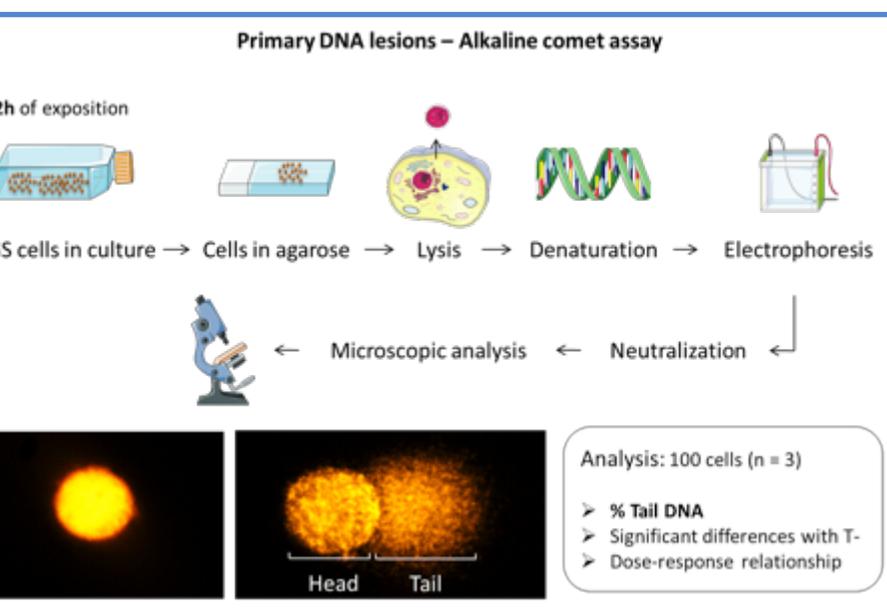
- Farmers could be occupationally exposed to biopesticides
- Consumers could be exposed through alcohol consumption

- No clinical signs during exposure
- No acceptable threshold

- The severity of the health effects do not decrease with the dose; only the effect occurrence probability decreases with the dose (deterministic versus stochastic effects)

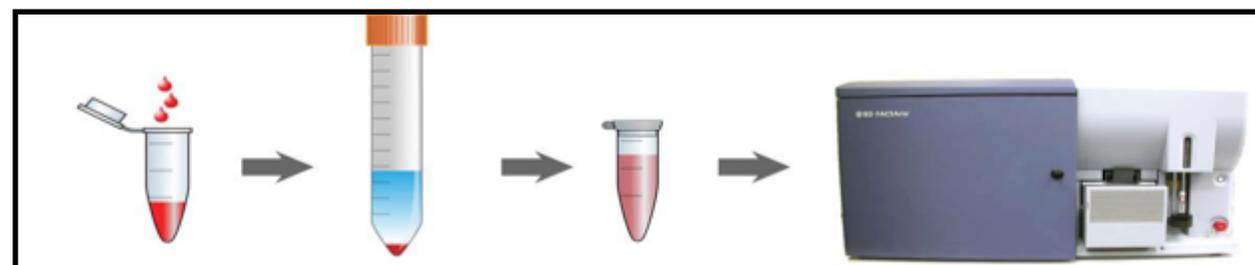
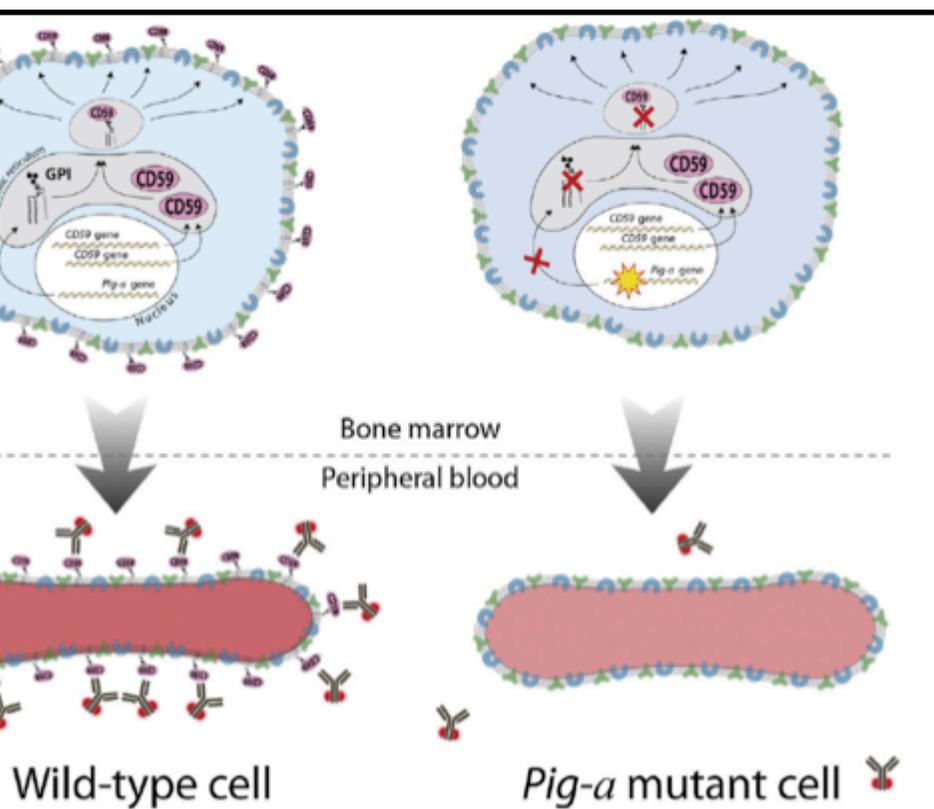
### 3 : To test the innocuity of both BCCs and bioamendment (JL Boudenne, Th. Orsière) 2

Genotoxicity assessment of biocontrol products fractions will be performed by the use of the comet assay, the cytokinesis block micronucleus assay, and if possible by the PIG-a Assay on human cell lines (BEAS-2B and spermatogonia GC-6spg)



### 3 : To test the innocuity of both BCCs and bioamendment (JL Boudenne, Th. Orsière) 3

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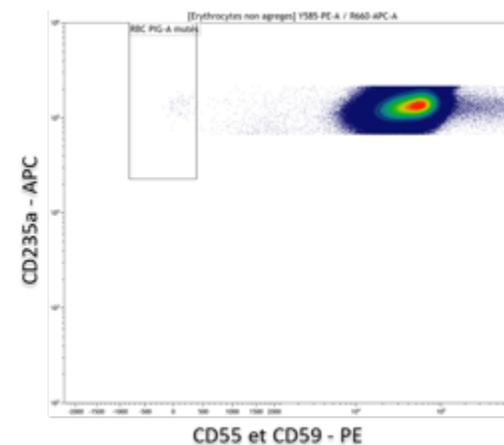


Cell exposure  
(24h)

Cell collection)

Immunostaining for two  
GPI-anchored proteins  
and cell viability

Flux cytometry analysis  
of  $> 5 \times 10^6$  cells



Example of data obtained  
on human erythrocytes

# TRANSAT: TRANverSal Actions for Tritium

Human Health in  
the context of  
Environmental  
Transition

Coordination with  
the TRANSAT  
Project (H2020)

<http://transat-h2020.eu/>

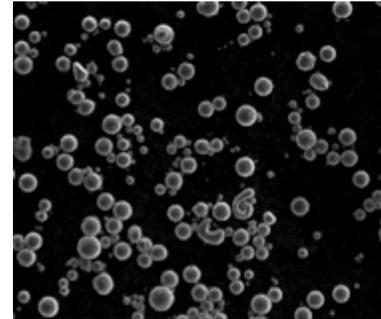
Terrestrial environment  
**plants**



Aquatic environment  
**pond**



**TRITIATED STEEL AND  
CEMENT PARTICLES**



Aquatic environment  
**sea**



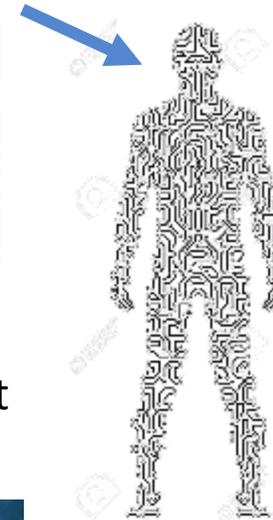
In vitro models  
**Human Lung**



**Rodent model**



WP3 TRANSAT 2021-10-11

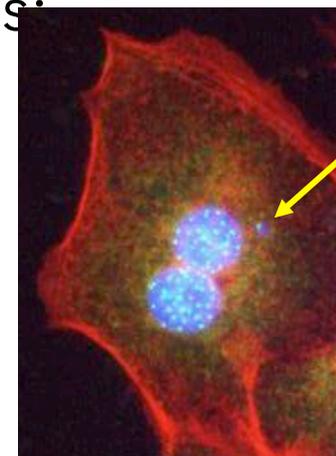


# TRANSAT: TRANverSal Actions for Tritium

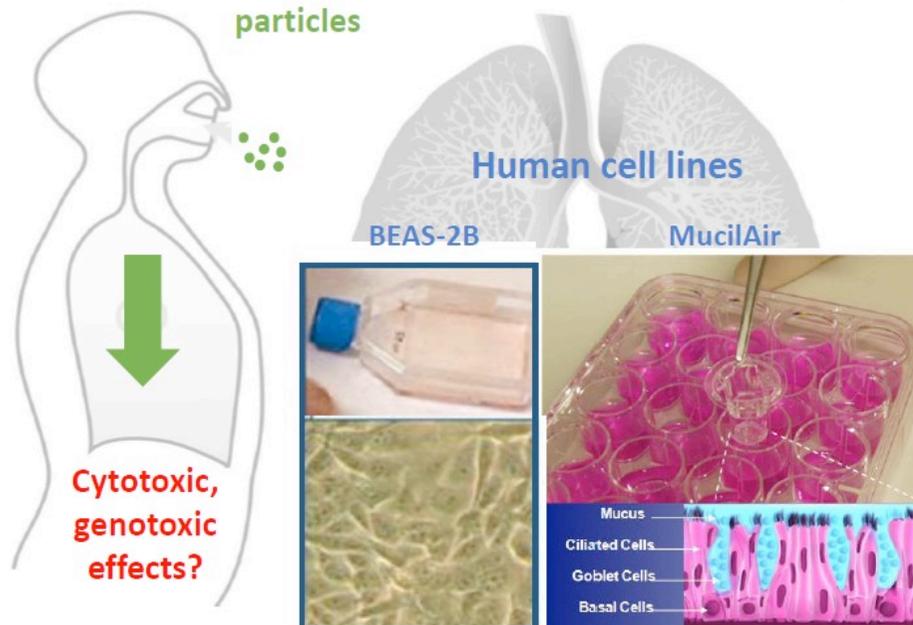
Tritium is of concern in fusion and fission facilities:

- Fusion facilities could be built if ITER is successful;
- The old fission facilities will have to be dismantled.

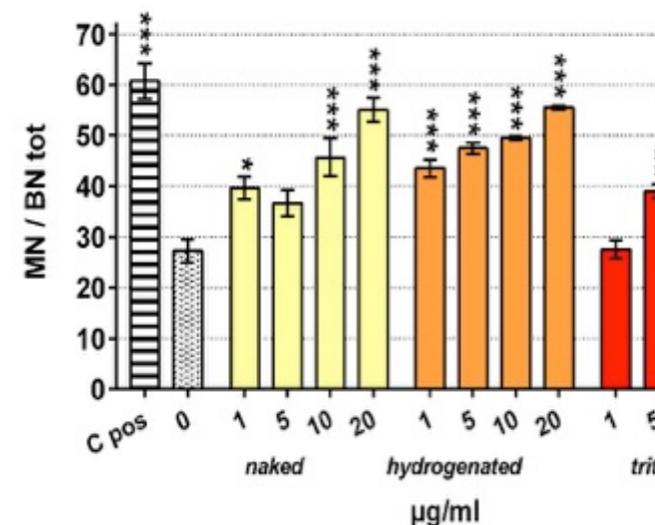
Both operations will produce tritiated dust



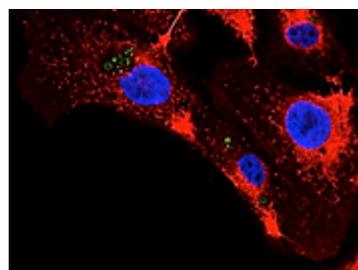
Tritiated stainless steel and cement particles



Plasma W-Ps



24h exposure C pos: 0.1 µg/ml MMC Centromeric staining: C  
One-way ANOVA with Sidak post-hoc test: \* p<0.05, \*\*p<0.01, \*\*\* p<0.001



TRANSAT  
Regarding  
Human Health

[//transat-h2020.eu/](http://transat-h2020.eu/)

Human health in context of environmental transition

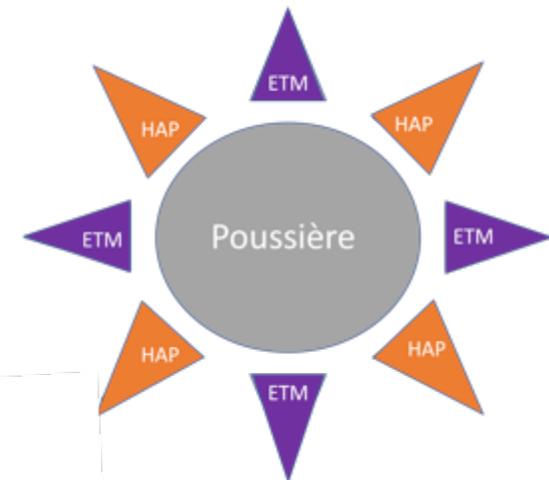
Concentration with a project studying vehicles that are (yet?) of concern

## Health impact of soil dust in preschools



Ingestion involontaire: 20-100 mg/jour

Effets sanitaires: inflammation, stress oxydant, génotoxicité ?



on de la  
évolution  
omme-Milieu à  
vers ces  
strations

Approche pluri- à trans-disciplinaire (idéalement) d'une situation

Préoccupation conjointe des impacts Santé Humaine et Santé Environnementale

Connexion bilatérale entre les chercheurs et la société civile

Etude d'impact aussi amont que possible des nouvelles technologies ou d'agents environnementaux non préoccupants à ce jour

Merci pour votre attention