

**Nanocellulose-based materials for the  
plant protection against *Xylella*  
*fastidiosa***

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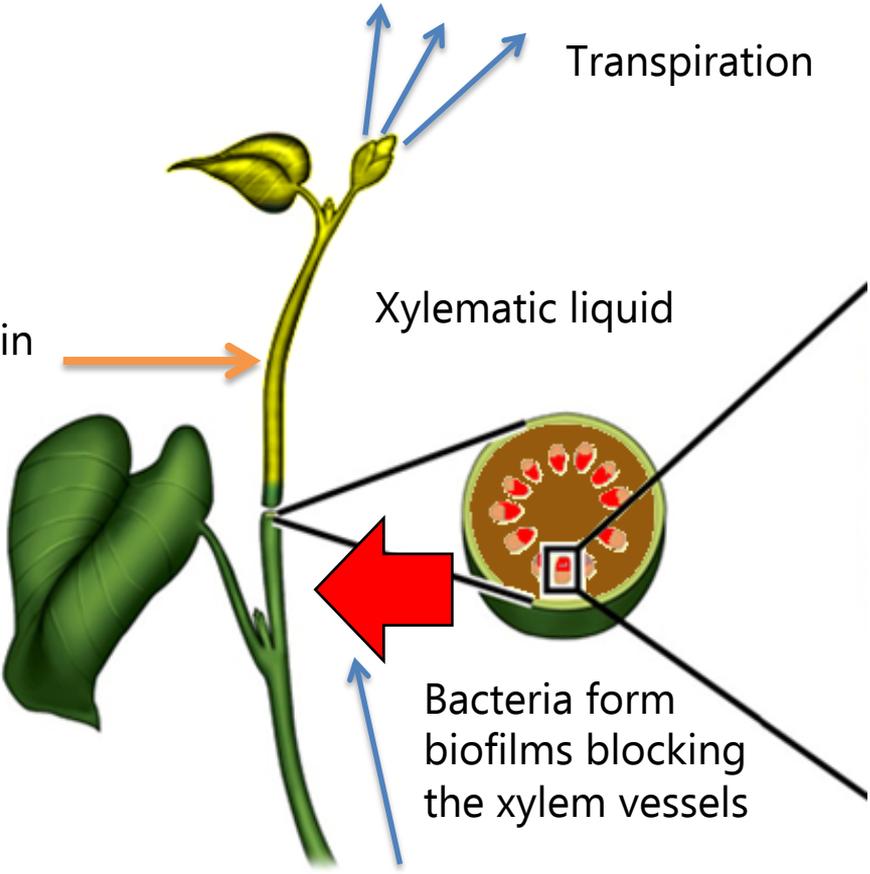
*Dipartimento di Biologia e Biotechnologie Charles Darwin*

# Olive quick decline syndrome

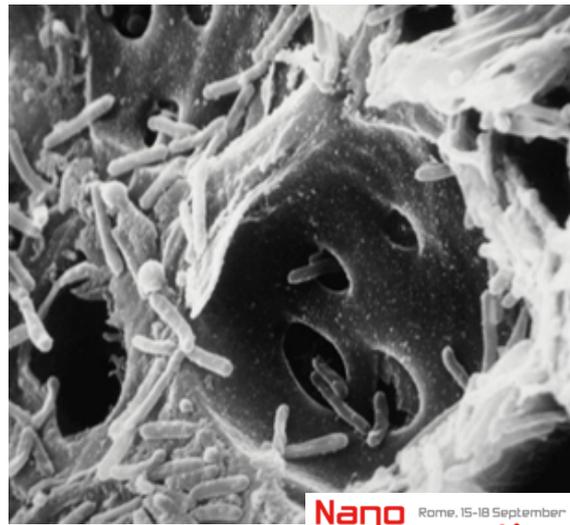
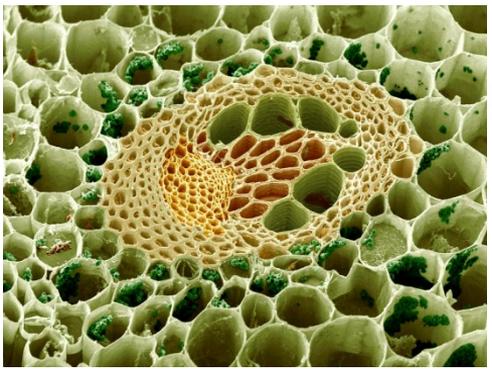


# Olive quick decline syndrome

Scorching is caused by low-level moisture that occurs as xylem vessels in leaves become blocked



Pathogen lives in the xylem vessels of host plant.



# Green Chemistry

## Progetto NEMESI

Nanotechnology for the protection of sustainable plants

- ✓ Design and validation of nanocellulose as nanocarrier
- ✓ Comparison of nanocellulose with other biocompatible nanomaterials
- ✓ Enhancement of waste material

## Progetto DEMETRA

Design and experimentation of innovative technologies for the early diagnosis and treatment of CoDiRO

- 4 universities
- 1 CNR institute
- 1 Regional research body
- 5 Companies

## Progetto FATA

Innovative phytotherapeutic treatments based on chitosan



UNIONE EUROPEA



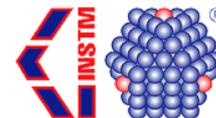
Ministero dello Sviluppo Economico



Regione Puglia  
Dipartimento Sviluppo Economico, Innovazione, Istruzione, Formazione e Lavoro



Il futuro alla portata di tutti



EUROBIOFERT



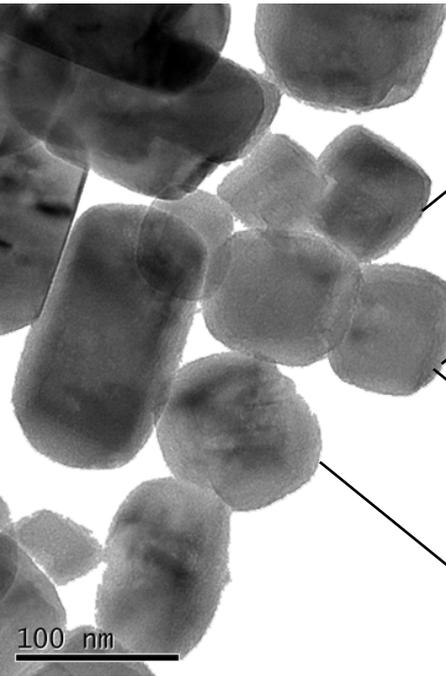
UNIVERSITÀ DEL SALENTO



Nano 2020 Innovation Conference & Exhibition  
Rome, 15-18 September

# Nanocarriers design and synthesis

Factors to be considered in designing a nanocarrier



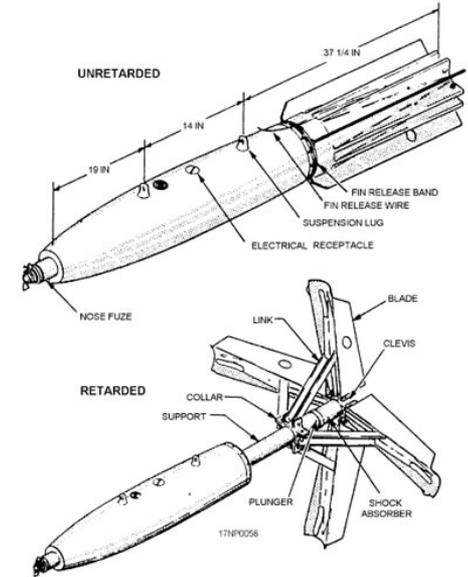
2.1 Small dimensions

2.2 Plant compatibility

2.3 Selectivity towards the pathogen

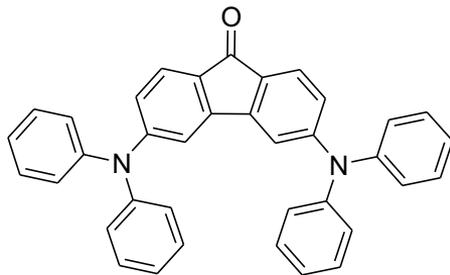
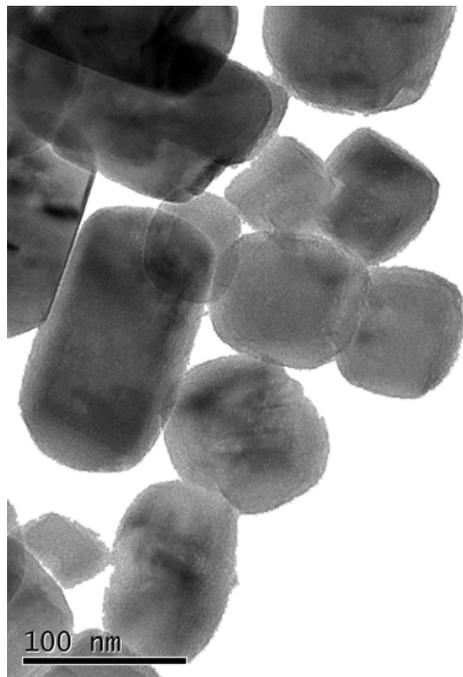
2.4 Multiple cargo capability

2.5 Slow release

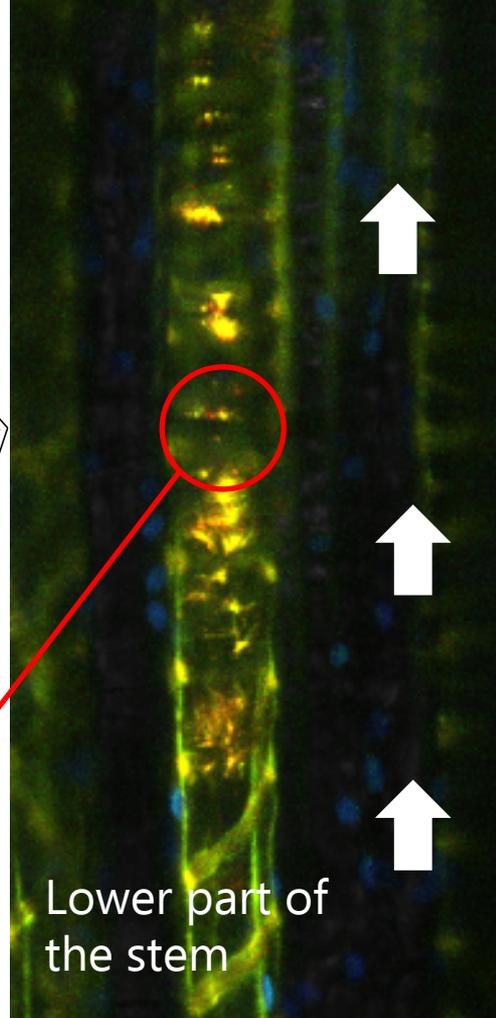


# Nanocarriers design and synthesis

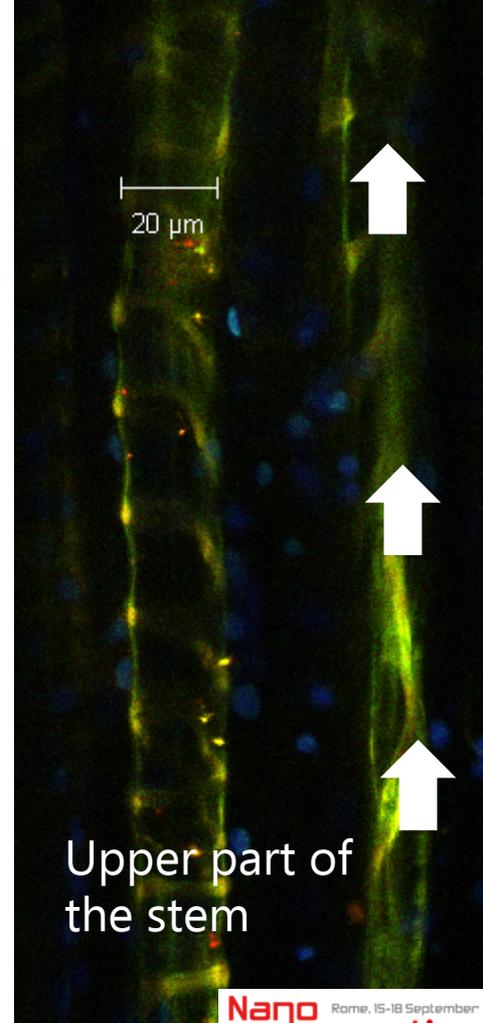
Plant compatibility - Endotherapy



fluorescent  
Nano-CaCO<sub>3</sub>



Lower part of  
the stem

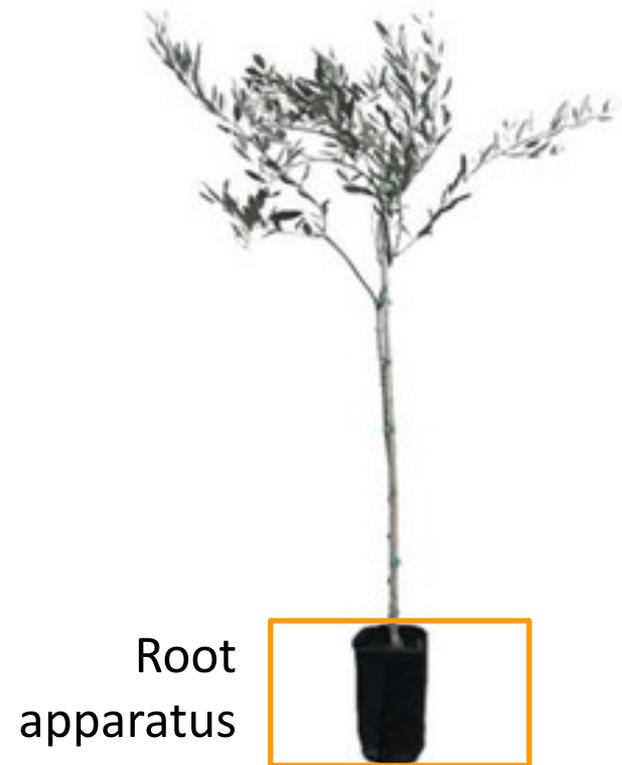
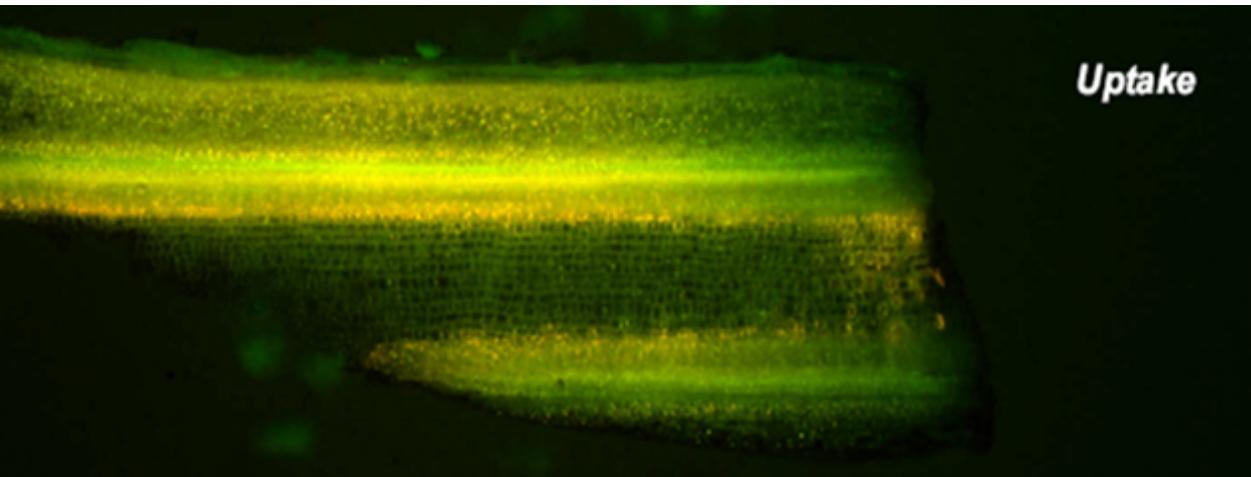
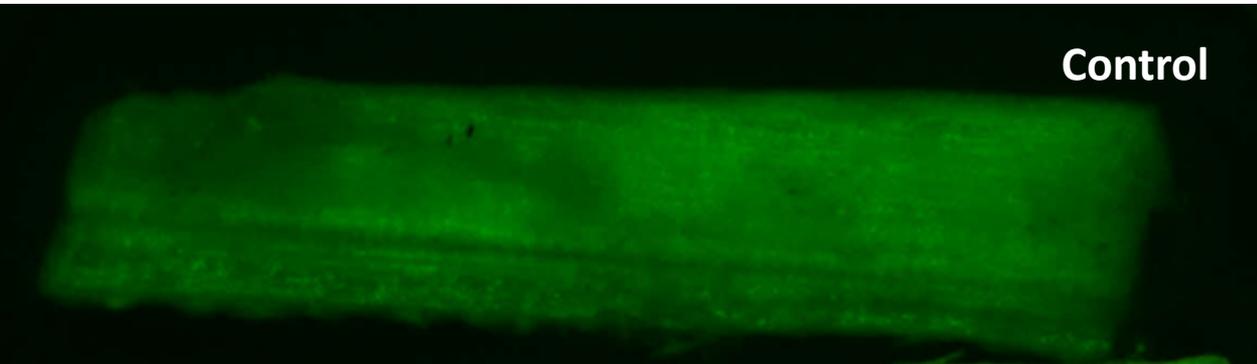


Upper part of  
the stem

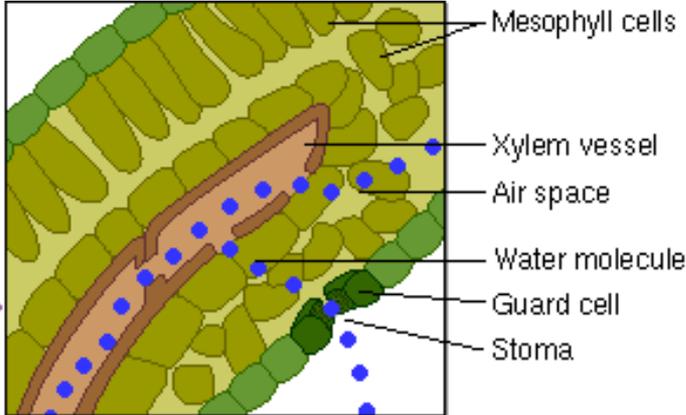
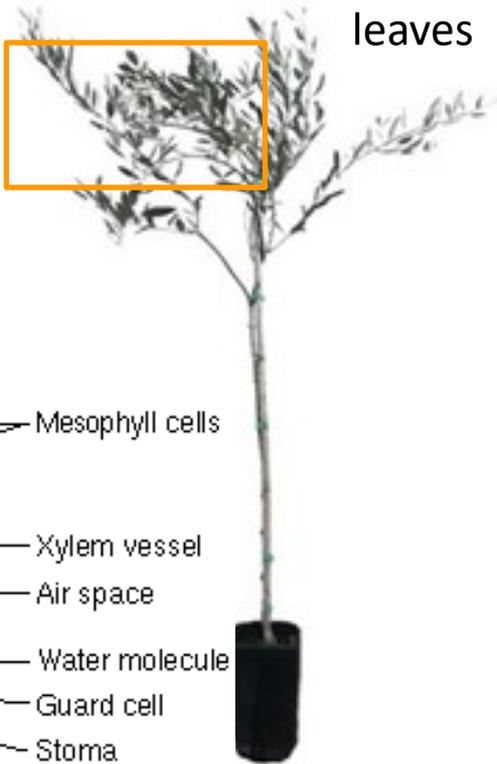
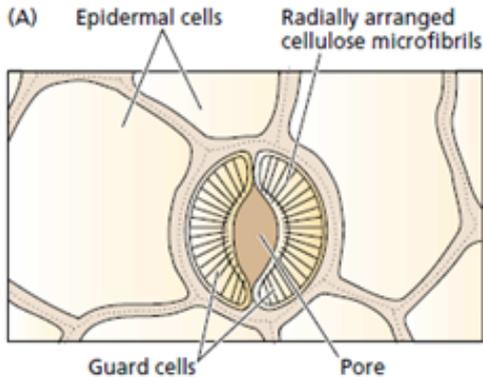
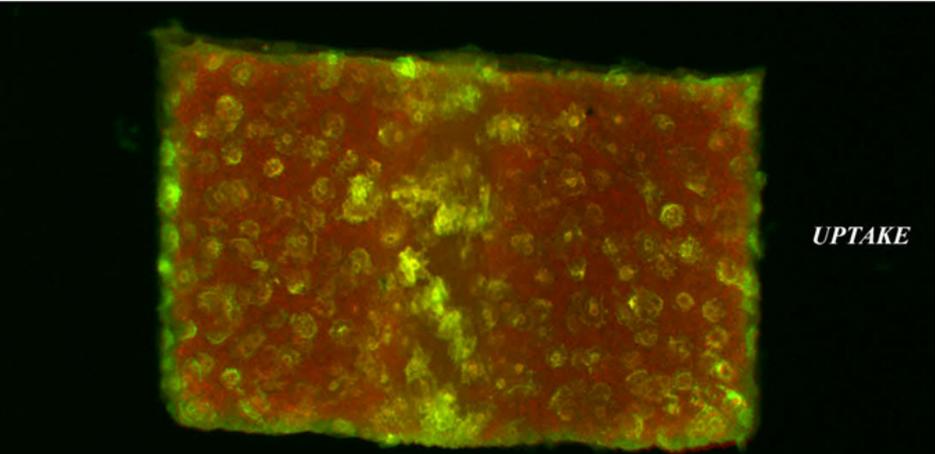
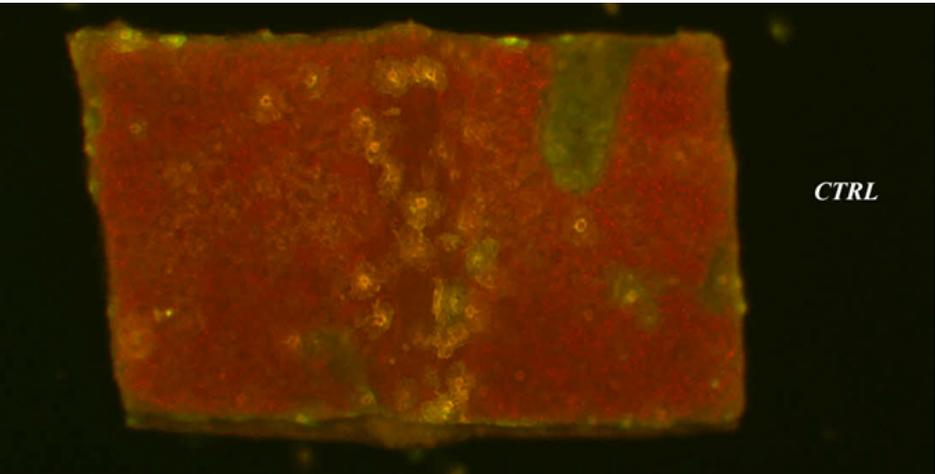
Experimental tests:  
Injection of fluorescent  
nanoCaCO<sub>3</sub> inside the plant

# Nanocarriers design and synthesis

Plant compatibility - **in Soil Experiments - Fertigation**

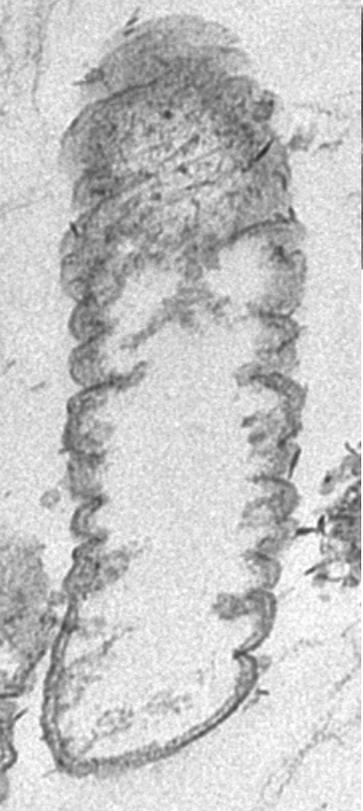
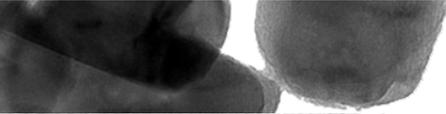


# Nanocarriers design and synthesis

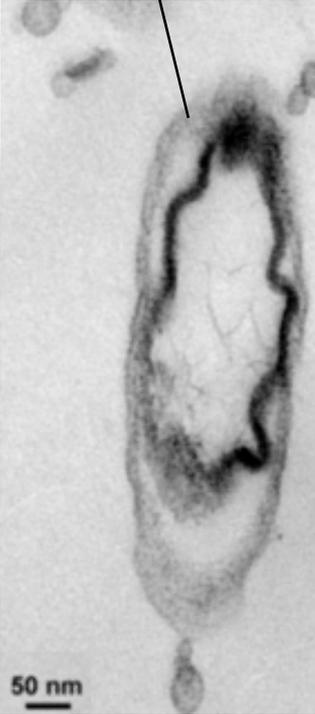


# Nanocarriers design and synthesis

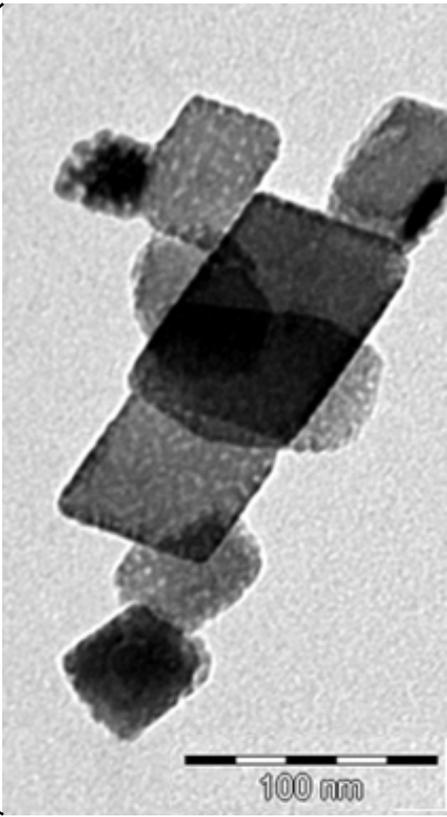
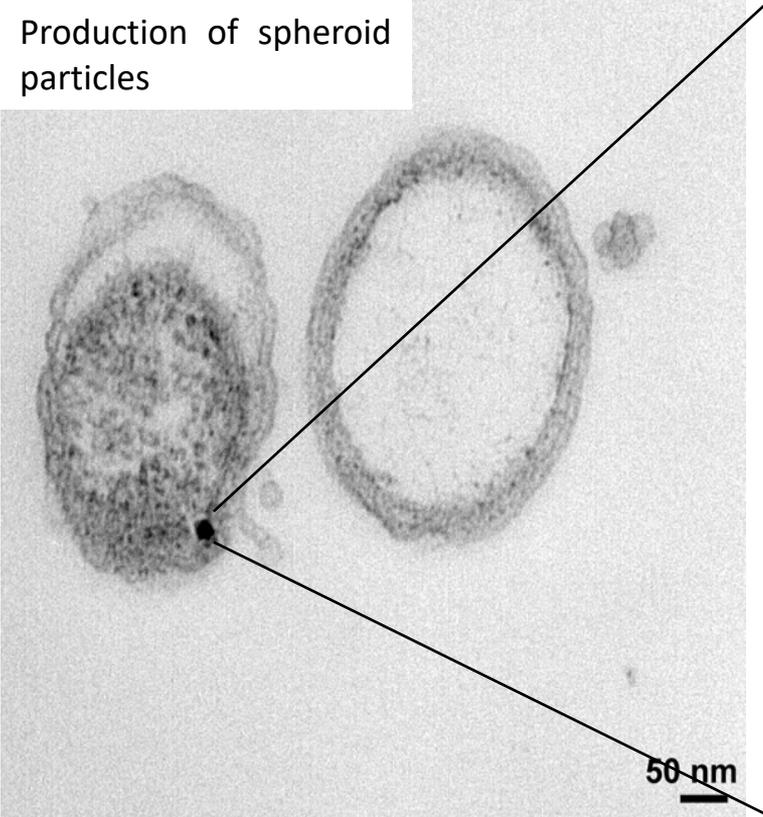
## 2.3 Selectivity towards the pathogen



Detachment of cell membranes



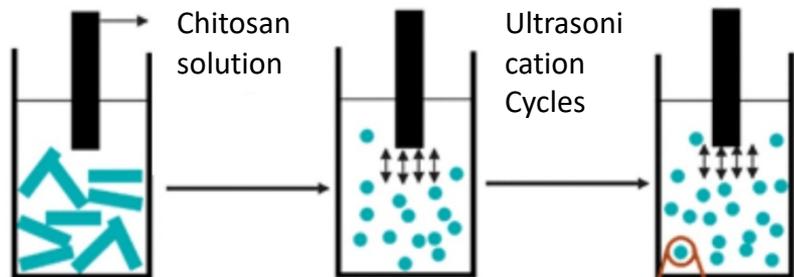
Production of spheroid particles



# Preparation of II-generation nano-structured phyto-drugs against *Xf*

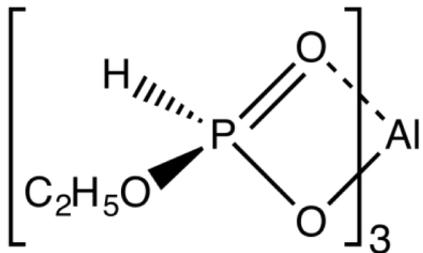
## Trattamenti Fitoterapici innovativi a base di vettori di chitosano-FATA

(Innovative phytotherapeutic treatments based on chitosan carriers)



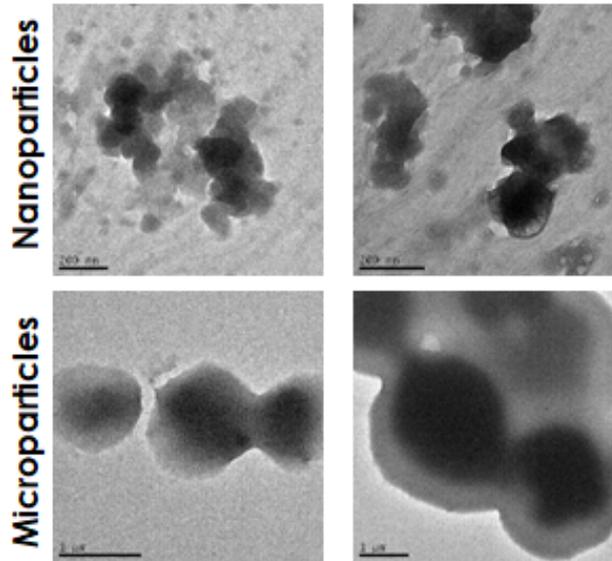
Fosetyl-Al

### Ultrasonication



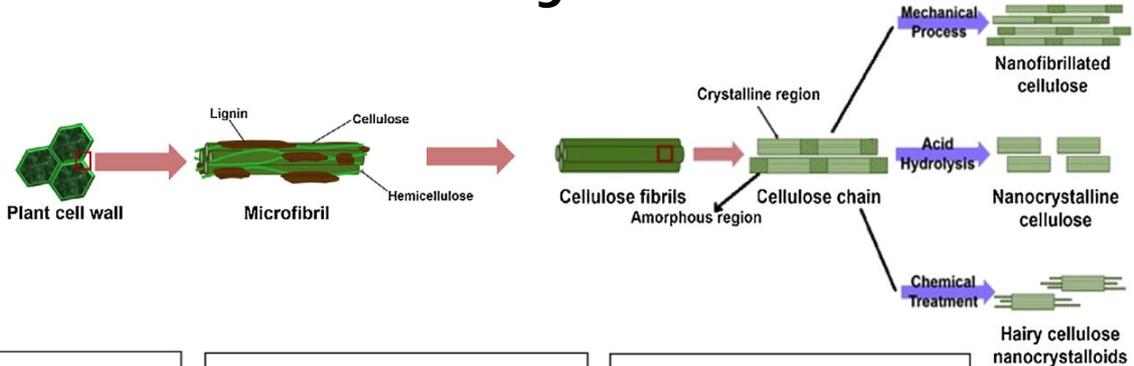
Chitosan-coated Fosetyl-Al Nanoparticles

TEM Images

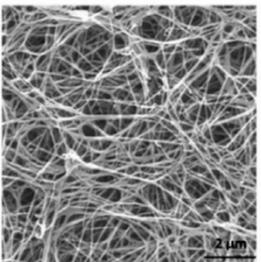


Production of nano-carriers which combine the antibacterial potential of chitosan with those of Fosetyl-Al.

# coming soon: nanocellulose-based agrichemicals



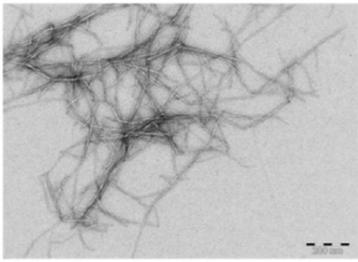
BNC fleece produced by *Gluconacetobacter* strain



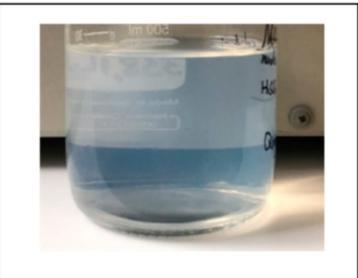
SEM



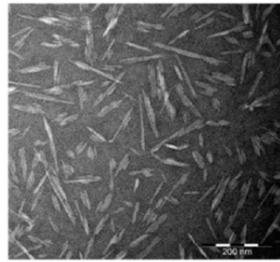
CNF hydrogel produced from softwood ~0.7 wt%



TEM



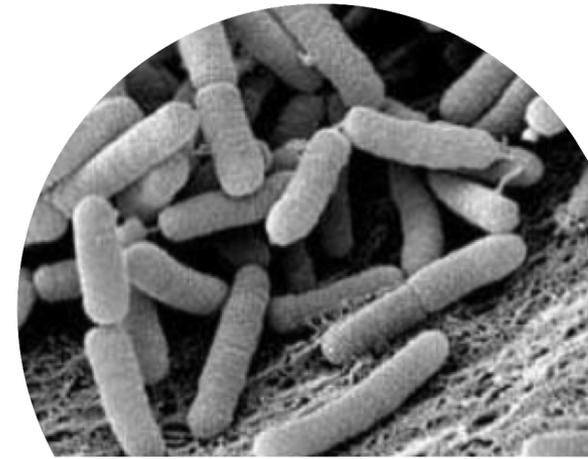
CNC suspension produced from softwood ~0.9 wt%



TEM

- ✓ Excellent mechanical properties
- ✓ High surface area
- ✓ Rich hydroxyl groups for modification
- ✓ 100% environmental friendliness.

coming soon



***Ultrastructural analysis of cultured bacteria and related biofilms treated with encapsulated phyto-drug***

**Electron  
microscopy**

- What is the effect of encapsulated phyto-drugs on bacteria vitality?
- How can nano/microparticles enter into bacteria?
- What is the fate of particles once inside the bacteria?



## Ultrastructural and ELISA analysis on plant samples infected with *Xylella fastidiosa* and treated with phyto-drugs encapsulated in nano- or microcellulose

The ultrastructural analysis by electron microscopy is the **ONLY** tool that allows an in-depth assessment of the alterations in the Xylem vessels whose occlusion causes the death of the plant.

### ELECTRON MICROSCOPY (TEM and SEM):

- ✓ It will support the development of the nanocarriers
- ✓ it will allow researchers to analyze the presence, the localization of bacteria in the plant
- ✓ It will allow researchers to observe the presence of biofilms that block water flow (ESEM)

The effectiveness of the treatment on the plant will be monitored by protein analysis by **ELISA** tests (to be developed) which will allow us to detect changes in the levels of protein markers identified by proteomics, in order to produce a rapid verification test on the outcome of the treatment.



# Nanotoxicological characterization of nano-agrichemicals

The evaluation will be carried out in vitro by monitoring the effects of nano-agrichemicals encapsulated with pesticides on the gastrointestinal (GI) barrier.

Soil

Aquifer

Food Chain

## GI BARRIER 3D MODEL

Caco-2 (epithelial cells of colon)

HT29-MTX (mucus-secreting intestinal cells)

Raji B (lymphoblast-like cell)

Scanning and transmission electron microscopy

- Evaluation of NAGs encapsulation
- Evaluation of the morphological effects on GI by in vitro 3D model





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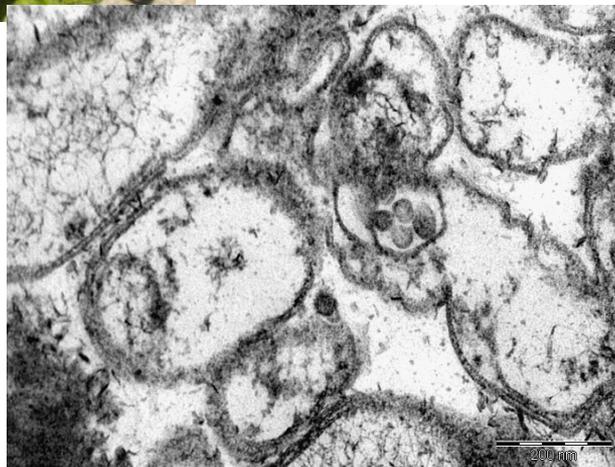
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Elisabetta Carata

Stefano Tacconi

Giorgio Mariano Balestra e team

Luciana Dini



# Thank you for your attention

