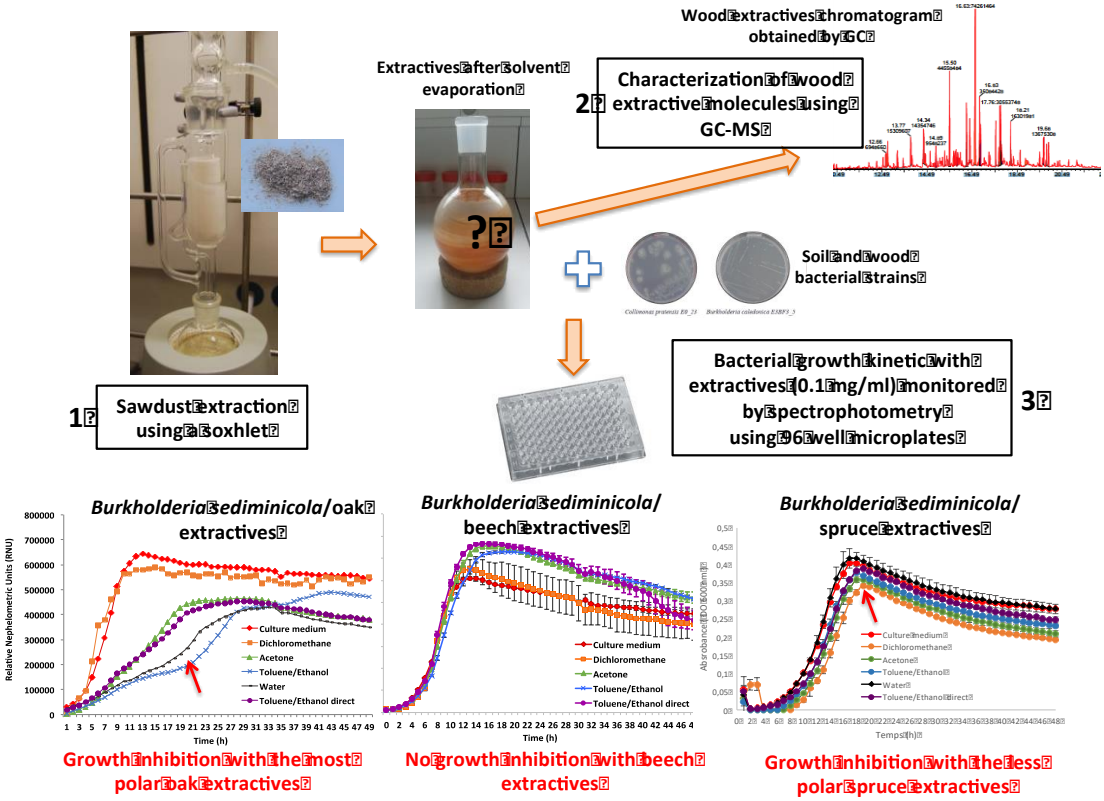
ISABEX



Interactions between Saprotrophic Bacteria and the white-rot *Phanerochaete chrysosporium* in presence of wood Extractives

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Context — At the scale of forest ecosystems, wood degradation contributes to nutrients recycling and represents an important source of fresh organic matter for lingo-cellulolytic microorganisms. However, during wood degradation, low molecular weight compounds, called extractives, are released. These compounds are potentially toxic and may represent a barrier for microbes. If the contribution of fungal communities in term of carbon and nutrients recycling is well established, the possible role of bacterial communities but also their interactions with fungi were largely underestimated. Recent studies have shown the complexity of these interactions that can be beneficial or not to the fungal or bacterial community or to both communities. Among these studies, one carried out at the IAM laboratory highlighted that the association of the white rot fungi *Phanerochaete chrysosporium* RP78 with a bacterial community isolated from its mycosphere improved beech degradation. The functional potential of each bacterium forming the bacterial community was characterised, however, their behaviour in presence of wood extractives still needs to be defined.

Objectives —

- To prepare and characterize beech (*Fagus sylvatica*), oak (*Quercus petraea*) and spruce (*Picea abies*) heartwood extractives.

- To evaluate if the effect of wood extractives on bacterial physiology is linked to the ecological niche of strains (soil *versus* wood, strains isolated from soil and wood come from V. Hervé bacterial collection).
- To evaluate if wood extractives affect bacterial/fungal interactions (BFIs).

Approach — Preparation of extractives was performed with a soxhlet by successive extractions with solvent having increasing polarities. Their characterisation was performed by gas chromatography-mass spectrometry (GC-MS).
Bacterial physiological response in presence of extractives was performed by a growth monitoring of bacterial strains using spectrophotometry and 96 well plates. Bacterial and extractive concentrations were respectively estimated at $1{,}2x10^7$ cells/ml and 0.1 mg/ml. BFIs in presence of wood extractives were evaluated by growth monitoring using spectrophotometry.

Key results —

- Five extractive fractions were obtained per tree-species. GC-MS characterization shows that beech extracts consist mainly of carbohydrates while oak and spruce produce potential antimicrobial molecules. For example, gallic and palmitic acids for oak or abietic acid and dodecanol for spruce were identified.
- Extractive fractions having antimicrobial properties were the most polar for oak ,and at the opposite, the less polar for spruce.
- Bacterial growth inhibition was significantly more important for bacterial strains isolated from soil compared to strains isolated from wood.
- After selection of an experiment to study microorganism interactions, the first results show an inhibitory effect in presence of extractives.

Main conclusions including key points of discussion — Among the tree-species and extractive fractions obtained, oak most polar fractions and spruce less polar fractions had the highest antimicrobial activity toward growth of a selection of bacterial strains. This inhibition could be partly attributed to palmitic and/or gallic acids for oak or to abietic acid and/or dodecanol for spruce. Growth inhibition was significantly more inhibited for bacterial strains isolated from soil than for strains isolated from wood, highlighting the importance of the ecological niche of microorganisms to resist/detoxify wood extractives. Finally, preliminary interaction tests between 2 bacteria show an inhibitory effect in presence of extractives.

Future perspectives — Interaction tests between bacteria and *P. chrysosporium* RP78 will be performed in the next few months to confirm the preliminary results.

The most important inhibition being obtained with the most polar oak extractives, it seems relevant to improve the ecological relevance of these results by testing these extractives on bacterial strains isolated from decaying oak. Therefore, we propose a new incitative LabEX 2017 project (DivBactO) in order to characterized the taxonomic and functional diversity of bacterial strains isolated from heartwood and sapwood of decaying oak and then to evaluate their physiological response in presence of oak extractives obtained with the ISABEX project.

Valorisation —

Mieszkin S., R. Sormani, P. Richet, M. Morel-Rouhier, E. Gelhaye and S. Dumarçay. Effect of wood extractives on wood-inhabiting microorganisms and importance of the ecological niche. *Ecology of soil microorganisms: microbes as important drivers of soil processes*, Prague, République Tchèque, 29 novembre-3 décembre 2015.