Minerals in wood: which role in the fungal decay process?

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**Context** — Saproxylic fungi and bacteria can use wood as nutrient source. It induces significant structure and composition alterations of the material. In the context of improved wood utilization in building (preservation), or biorefinery (chemical valorization of biomass) fields, understanding of mechanisms supporting wood deconstruction is of high priority. Relative abundance of mineral elements such as iron, copper and manganese in various soil types and the tree physiological activity lead to their variable accumulation in wood. Several studies showed that minerals seem to play important roles in the wood decay mechanisms setup by microorganisms. However, knowledge about their actual contribution remains scarce.

**Objectives** — Determine the impact of wood mineral contents on its durability when subjected to an attack by bacteria and fungi. Bring clues to better understand the bacteria/fungi interactions in the wood degradation process.

**Approach** — A multidisciplinary approach was carried out to investigate the degradation of beech wood samples impregnated with iron (FeSO₄ and FeCl₃), manganese (MnCl₂) or copper (CuSO₄) salts. Wood specimens were challenged with two white rot fungi, *Trametes versicolor* and *Phanerochaetes chrysosporium* alone or in combination with a consortium of 9 bacterial strains isolated from wood colonized by *P. chrysosporium*. Different imaging and wood material characterization methods combined with biochemical measurements were used to investigate the impact of mineral on the wood decay caused by the bacteria/fungi system.

**Key results** —

- Setup of the FISH method on wood colonised by the fungi and bacteria.
- Bacterial growth rely on nature and amount of the mineral tested.
- GFP or DsRED expressing strains of *Burkholderia spp.* et *Diella spp* isolated from wood were generated.
Wood impregnated with Mn and Fe at low concentrations (ICP quantification) promoted fungal decay.

The iron salts that were tested dramatically influenced β-glucosidase and laccase activity in *T. versicolor*.

Several strains of *Burkholderia spp.* and *Diella spp.* were observed in interaction with *P. chrysosporium* hyphae but never with *T. versicolor*.

Different observed bacterial locations suggested specific functional interactions between bacteria and fungi. Namely bacteria were observed near to oxalate crystals, gliding on the *P. chrysosporium* hyphae or eroding wood cell wall.

**Main conclusions including key points of discussion** — Tested minerals affected fungal growth and decaying activity. The mineral bioavailability was an important factor in their activity threshold determination on bacteria and fungi. Bacterial communities exhibit heterogeneous distribution in wood complicating their visualization and the FISH method validation. Interestingly, the bacterial consortia inhibited *P. chrysosporium* growth on petri dishes but not during wood sample colonization. In wood distinct spatial distribution of the different bacterial morphologies was observed.

**Future perspectives** — Real time monitoring of wood colonization by bacterial strains expressing either GFP or DsRED together with *P. chrysosporium* will be performed by confocal microscopy. The precise repartition of minerals and bacteria will be determined by SEM. This will be achieved by correlative microscopy technology that was developed in the INABACT labex project. The optimization of the FISH technique should provide tool for elucidating spatial bacterial distribution upon wood decay. The infrared spectrometry analyses under process will bring clues on the wood cell wall chemical alterations caused by the different microorganisms. Finally, the use of the different transformed bacterial strains in combination with laser microdissection technique would allow elucidating preferential bacterial/fungus interactions and would be used as a tool for the *in situ* characterization of these interactions.