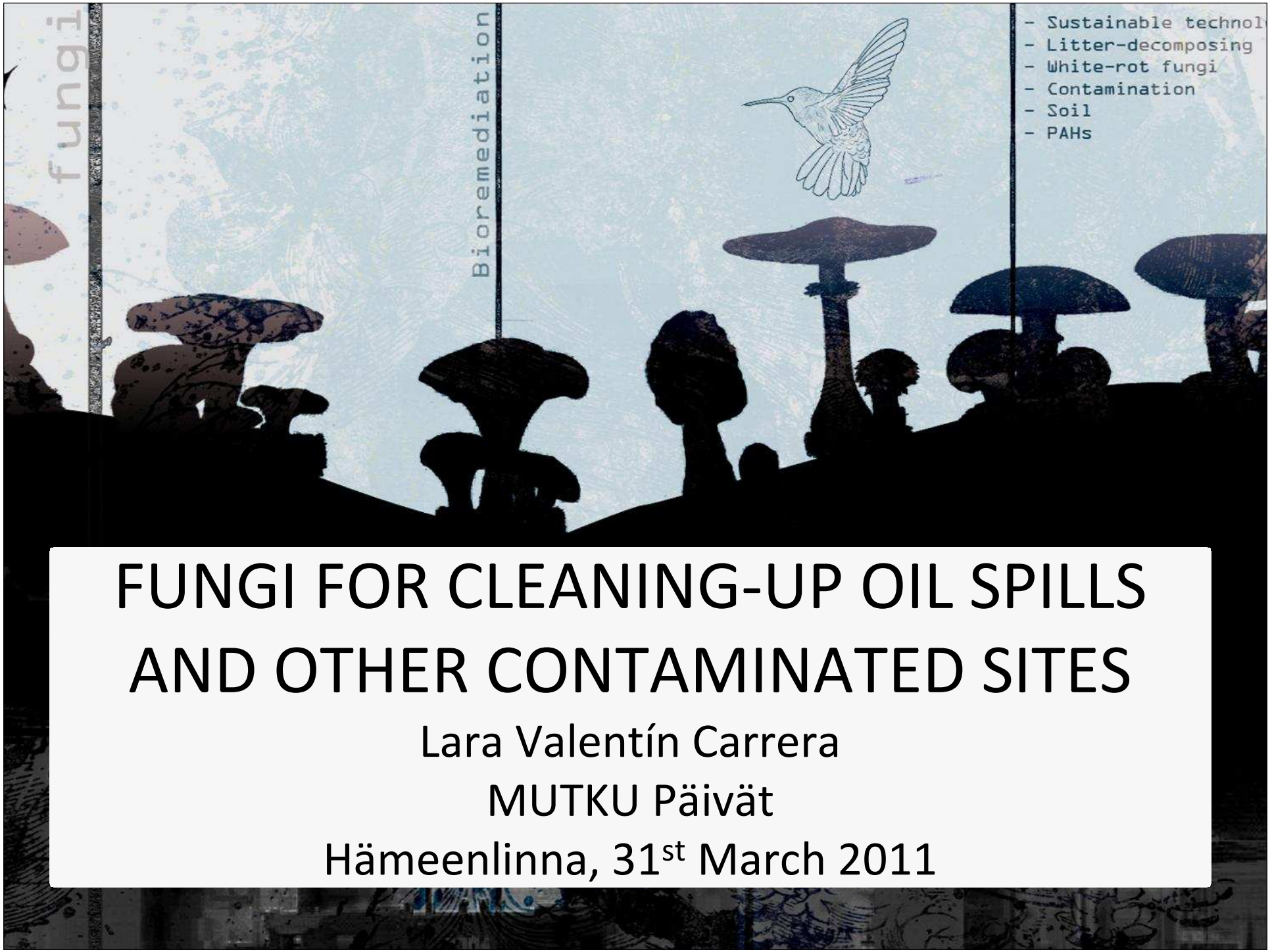


FUNGI FOR CLEANING-UP OIL SPILLS AND OTHER CONTAMINATED SITES

Lara Valentín Carrera

MUTKU Päivät

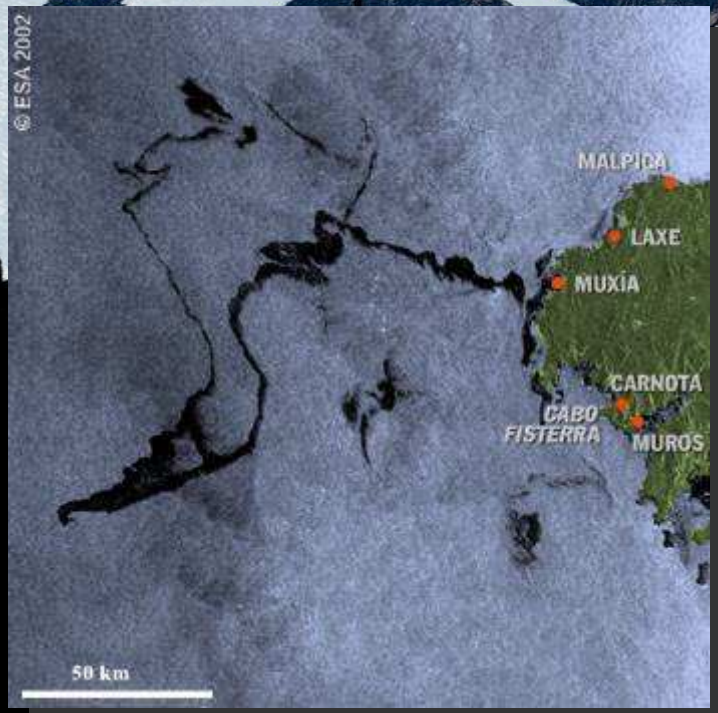
Hämeenlinna, 31st March 2011

CASE 1: OIL SPILLS

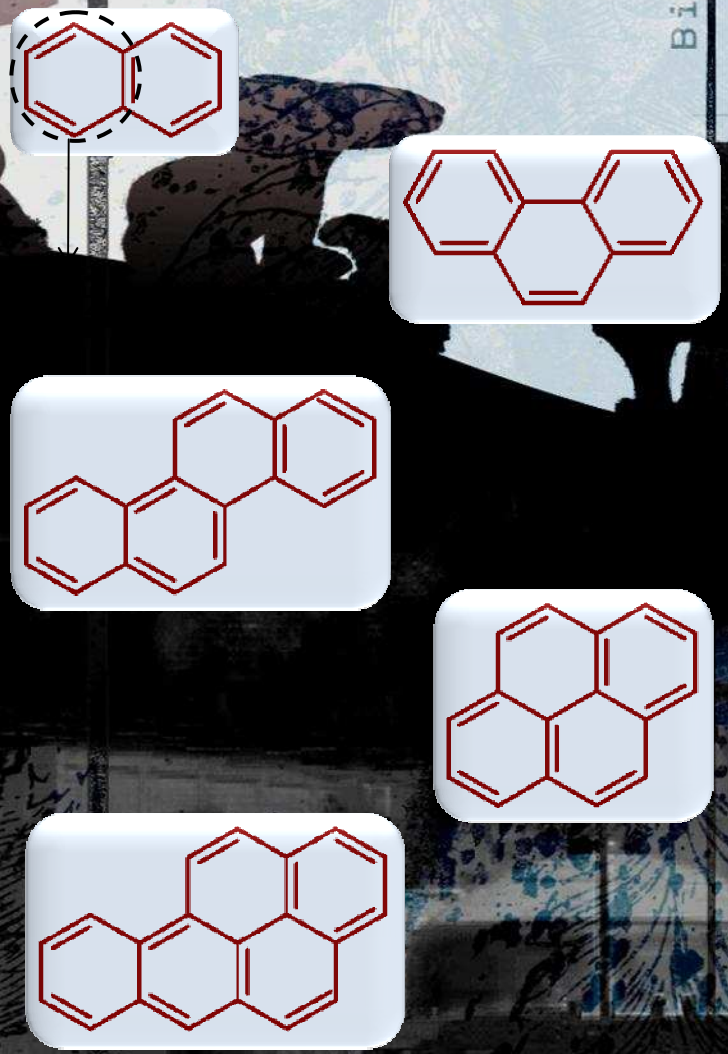
- Sustainable technol
- Litter-decomposing
- White-rot fungi
- Contamination
- Soil

On 19th November 2002, the single-hulled oil tanker Prestige sank on the Cap Finisterre (NW Spain)

- **Spilled quantity on the sea: 64000 tons of heavy fuel oil (N°2 M100).**
- **Affected coastal area : 1900 km of estuaries, marshes, beaches, etc.**
- **Affected countries: Spain, Portugal, France, England.**
- **16000 - 23000 tons still on the ship (Science 22, 2006).**
- **Risk of biocorrosion.**



An important fraction of the Prestige oil consists of Polycyclic Aromatic Hydrocarbons (PAHs)



- **Due to their low solubility, PAHs are mainly deposited into sediments or coastal areas.**
- **Toxic: PAHs are associated with lung and bladder cancers.**
- **More benzene rings:**
  →Higher carcinogenicity risk.
  → Lower solubility.
  → Lower bioavailability.
  →More resistant to bacterial degradation.

CASE 2: CONTAMINATED SAWMILL SOIL

- Sustainable technol
- Litter-decomposing
- White-rot fungi
- Contamination
- Soil

1940 – 1984, Finland: Around 23400 tons of chlorinated wood preservatives (KY-5) were produced.

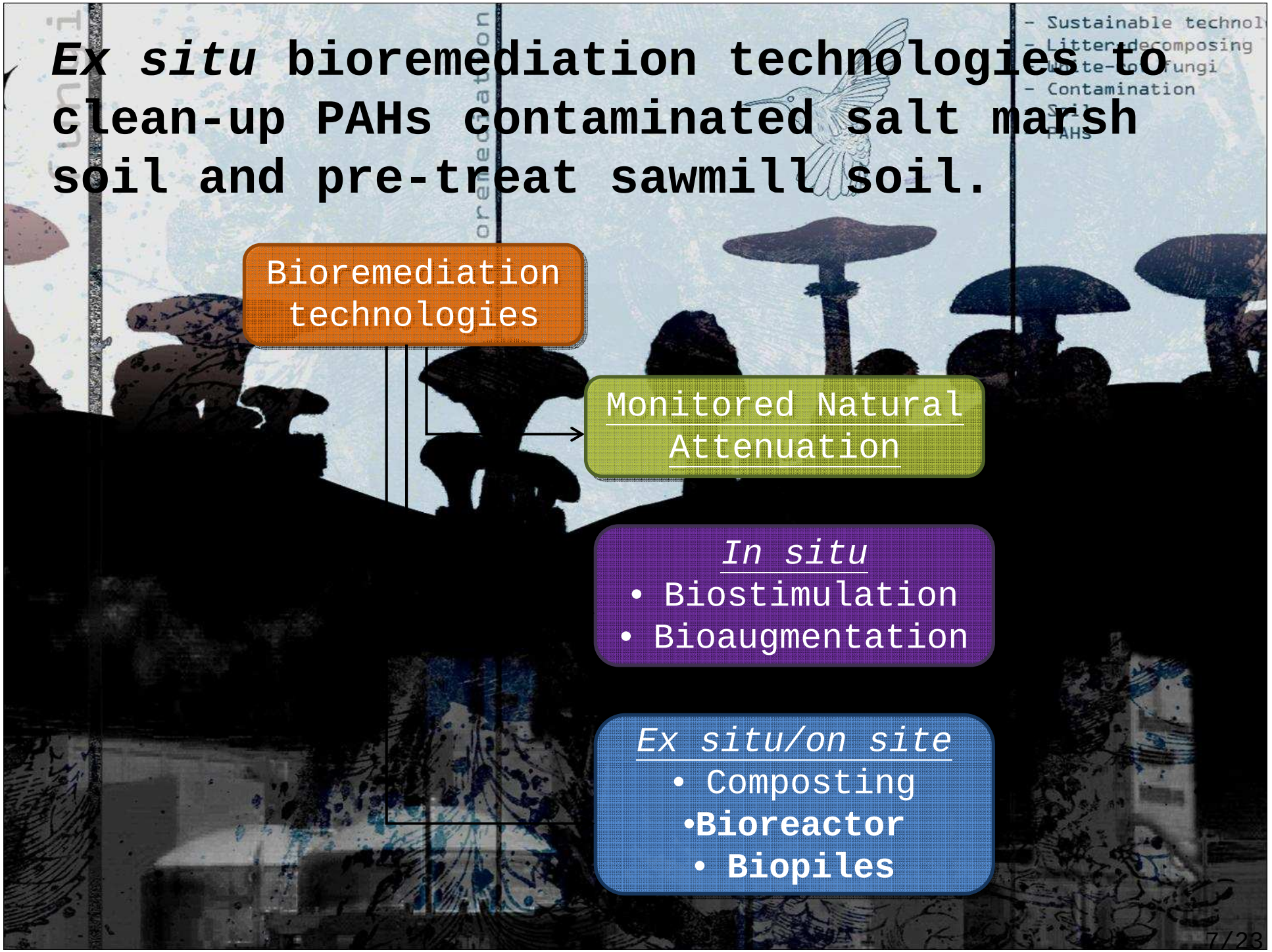
- 550 (former) wood preservation and sawmill sites potentially contaminated → 100 requiring urgent treatment.

- Highly contaminated sites → excavation + combustion(>1300°C).

- High soil organic matter.

- Limitations:
→ decrease combustion process capacity.
→ longer treatment time.
→ more fuel.

Ex situ bioremediation technologies to clean-up PAHs contaminated salt marsh soil and pre-treat sawmill soil.



Fungi with most potential to degrade contaminants are wood-degrading Basidiomycetes

Bjerkandera adusta (WRF)

Gymnopilus penetrans (LDF)

Irpex lacteus (WRF)

- **White-rot fungi (WRF)** live in standing or fallen dead wood (hardwoods; beech, birch). Degrade lignin, hemicellulose and cellulose from wood cells.

- **Litter-decomposing fungi (LDF)** live in the upper layer of the soil. Decompose dead leaves, needles, branches, roots causing white-rot to soil-litter.

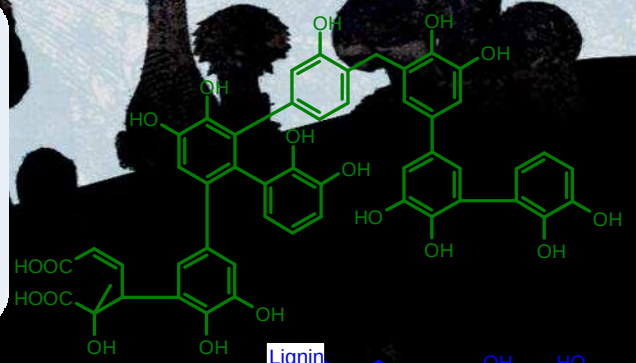
- **Fungi whose habitat is wood in direct contact with the soil (e.g. *Hypholoma* spp.).**

Why these fungi can be used for bioremediation applications?

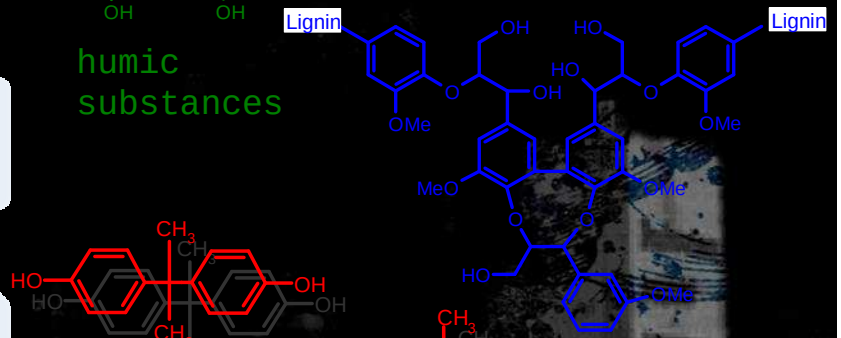
- Sustainable technol
- litter-decomposing
- White-rot fungi
- Contamination
- Soil
- PAHs

- **Production of mycelium spreading upon the soil.**

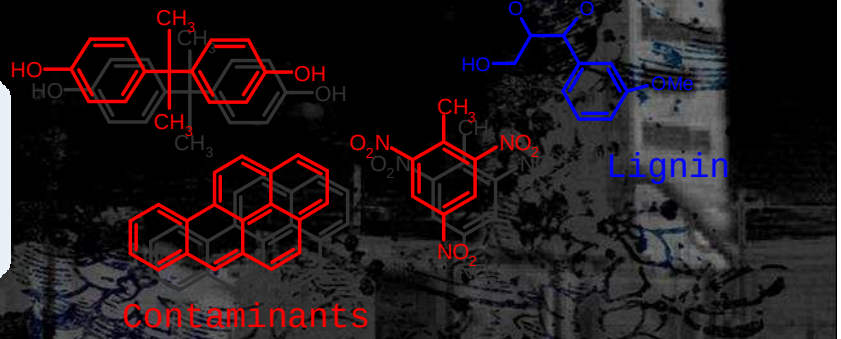
- **Non-specific and extracellular lignin-modifying enzymes**
  - → **Lignin Peroxidase**
  - → **Manganese Peroxidase**
  - → **Laccase**



- **Tolerate organic contaminants and heavy metals.**

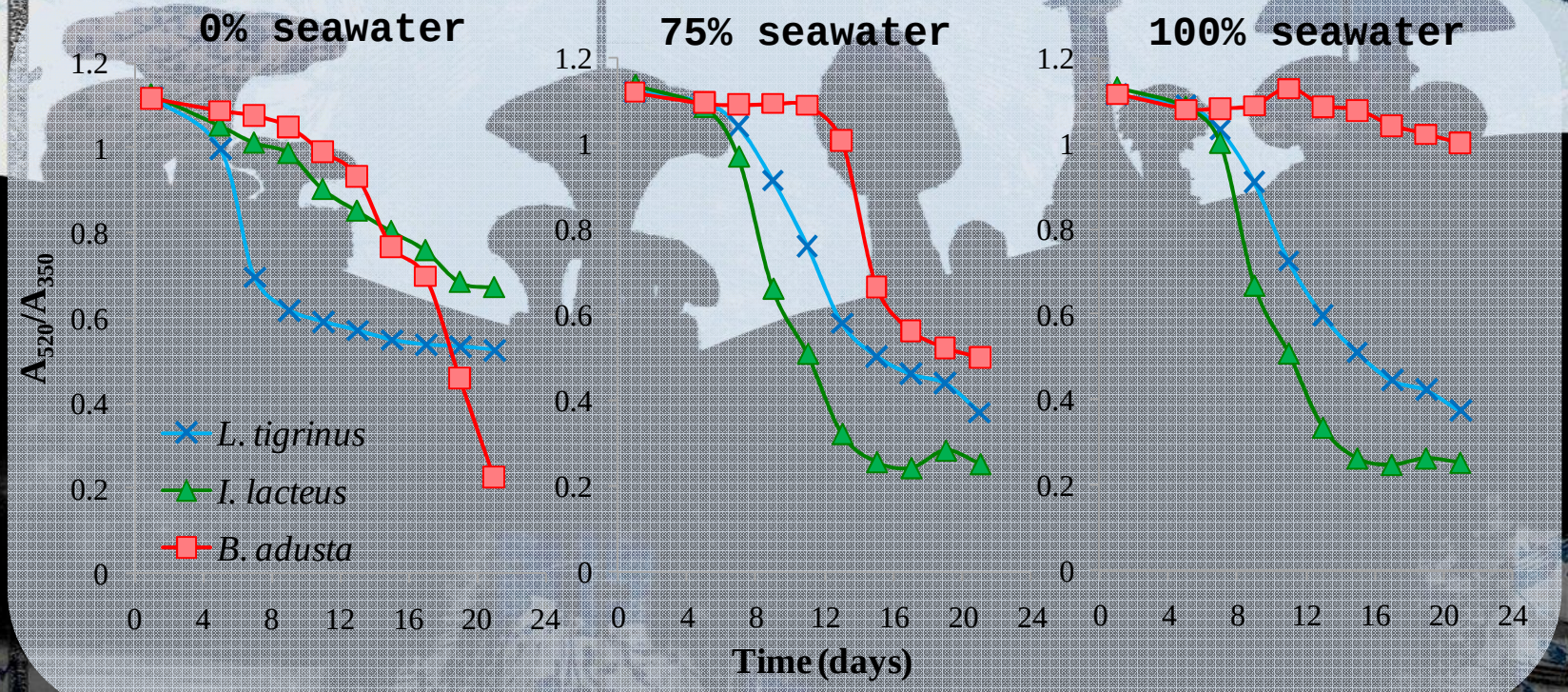


- **Lignocellulosic material as substrate for fungi (e.g. wood chips, bark, straw, etc)**



Case 1: FUNGAL DEGRADATION OF PAHs FROM SALT MARSH SOIL IN A SLURRY BIOREACTOR

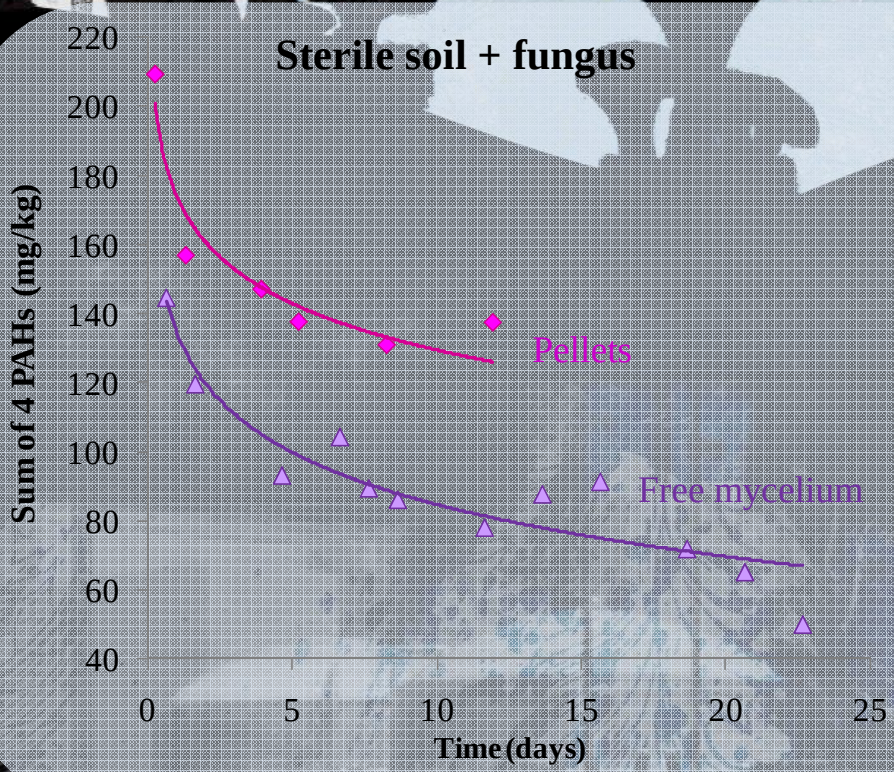
After the screening of PAH degradation in small slurry reactors with 9 fungi, the effect of seawater on the enzymatic system of 3 fungi was evaluated

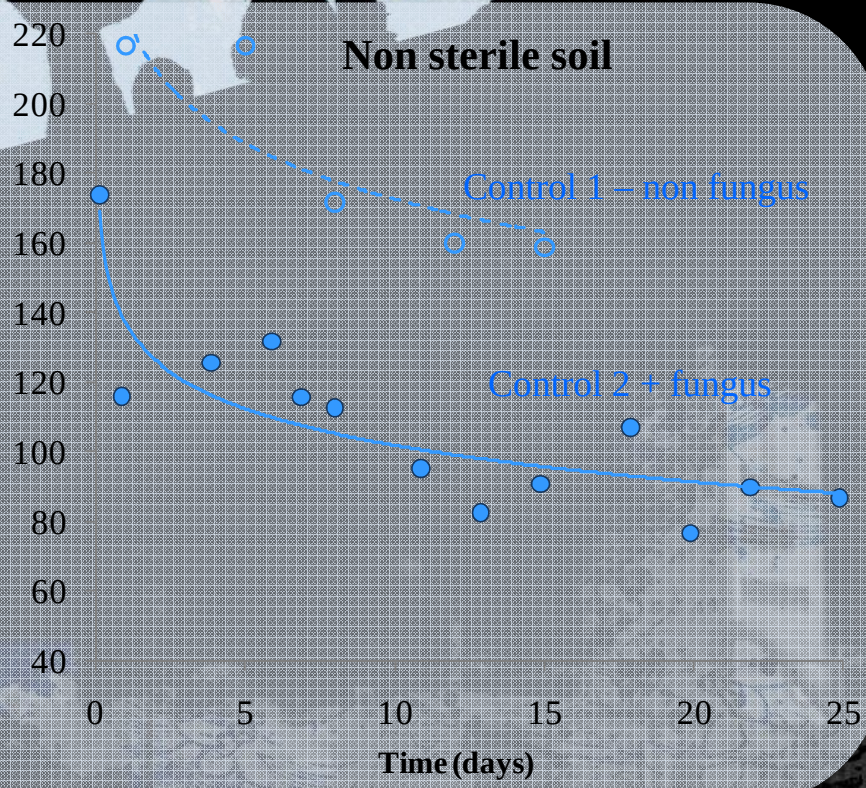


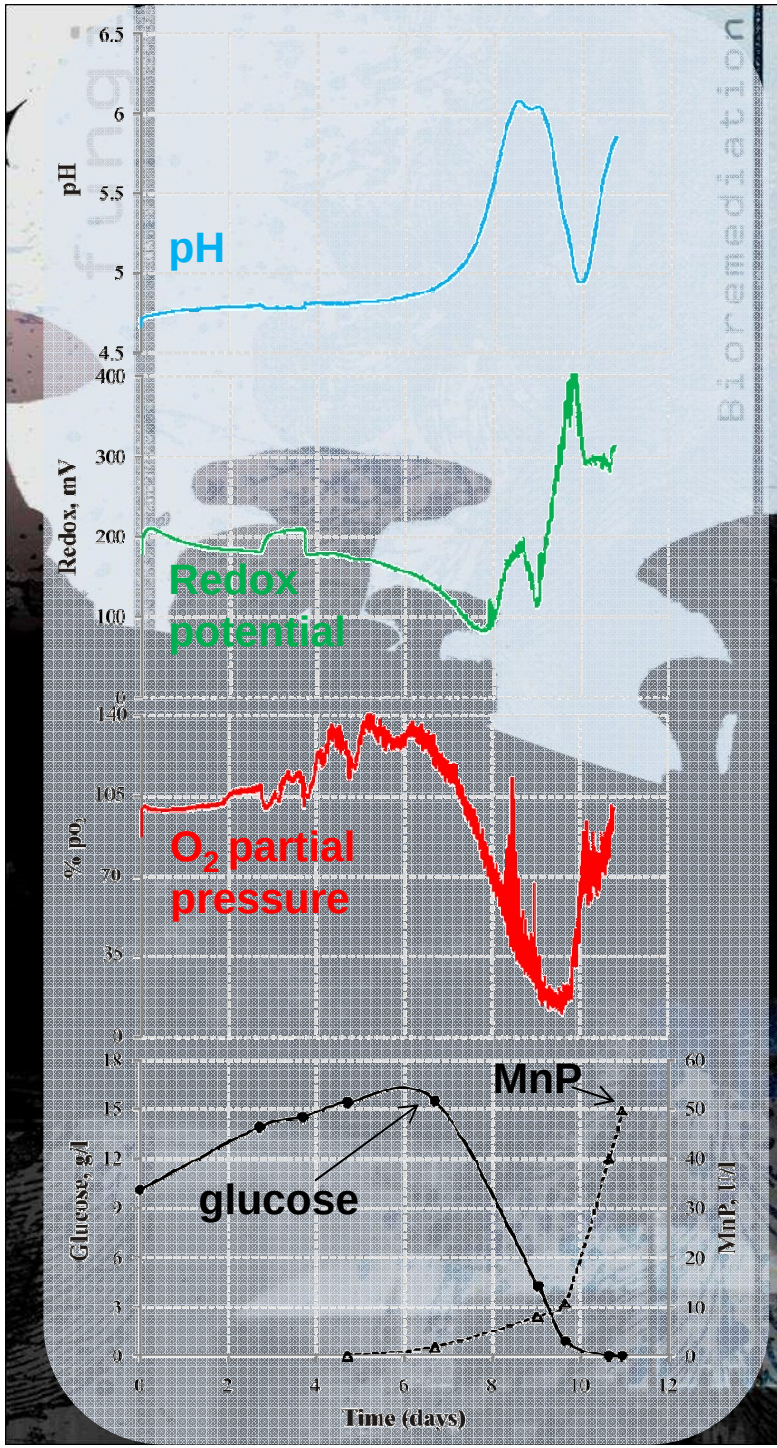
A_{520}/A_{350} is the absorbance rate of [illegible] Poly[illegible]

PAH degradation in 5 L bioreactor operated with *Bjerkandera adusta*

PAHs = dibenzothiophene, fluoranthene, pyrene and chrysene







- Sustainable technol
- Litter-decomposing
- White-rot fungi
- Contamination
- Soil
- PAHs

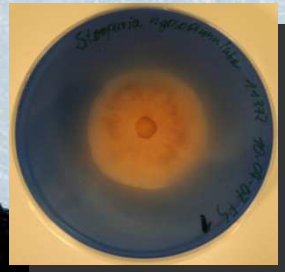
Similar fermentation profile of *Bjerkandera* *adu* in a ma oil s rry an con

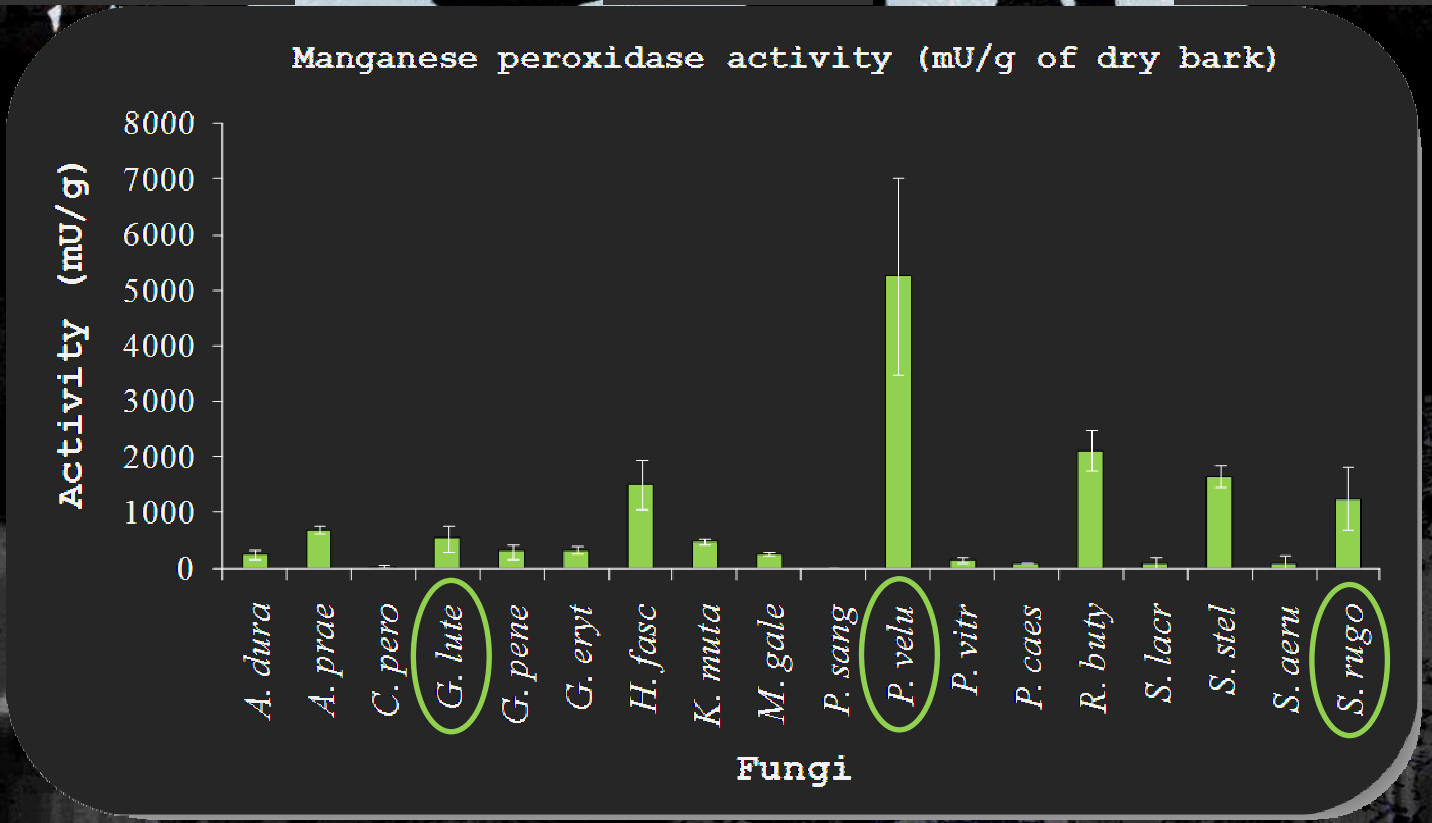
Conclusions case 1

- Several white-rot fungi (WRF) are halotolerant (tolerate salt) and are able to degrade PAHs under slurry conditions: *Bjerkandera adusta*, *Irpex lacteus* and *Lentinus tigrinus*.
- The process was successfully scaled-up (5 L) using B. *adusta* as free mycelium.
- Fungus and soil endogenous microbes cooperate in the degradation of PAHs.

CASE 2: FUNGAL SOLID PHASE PRE-TREATMENT OF CONTAMINATED SOIL TO DECREASE ORGANIC MATTER CONTENT

Screening of 146 wood-degrading fungi in contaminated sawmill soil resulted in the selection of 18 fungi

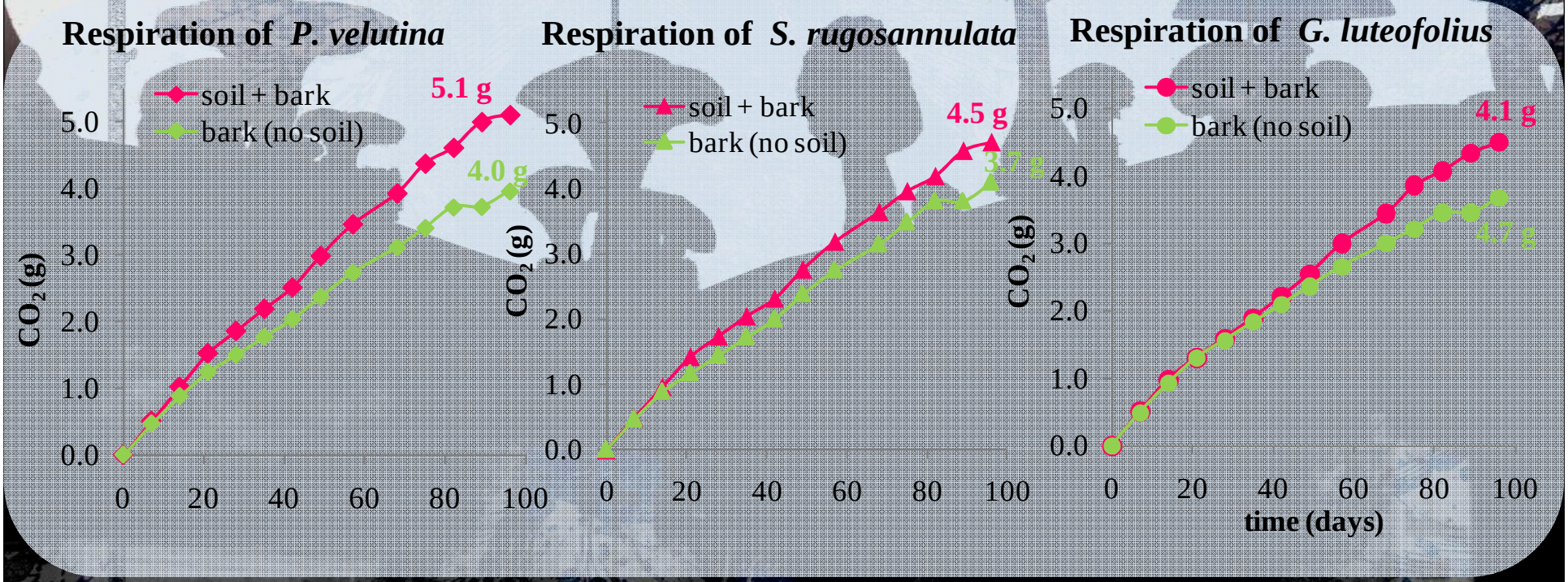




Degradation of organic matter from contaminated sawmill soil by fungi

2.1 mg/kg I-TEQ PCDD/Fs; 500 ml bottles; bark:soil = 21:100 (w/w)

Initial soil organic matter (OM) was 84%



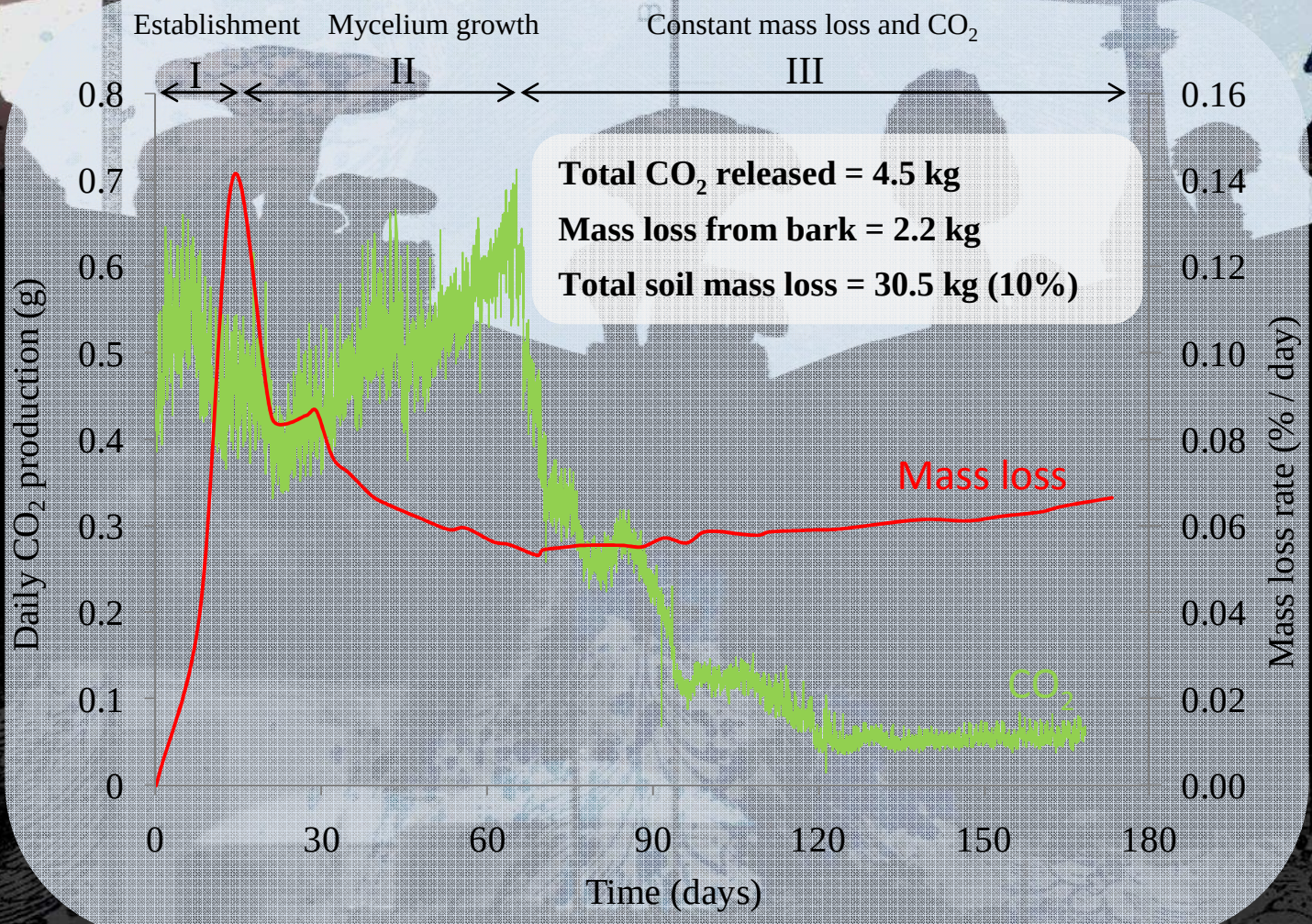
C loss = 3.4%
OM (6 months) = 79%

C loss = 2.4%
OM (6 months) = 80%

C loss = 1.8%
OM (6 months) = 81%

Scale-up of the fungal pre-treatment process

Biopile 0.56 m³; 308 kg of contaminated sawmill soil
82% OM; 0.07 mg/kg I-TEQ PCDD/Fs
Fungal-bark inocula in 6 mesh tubes (1.5 kg bark in each tube)
Stropharia rugosoannulata




Set-up biopile


Fungal-bark tube


Mycelium in wood (6 months)

Conclusions case 2

- Sustainable technol
- Litter-decomposing
- White-rot fungi
- Contamination
- Soil
- PAHs

- **Litter-decomposing fungi (LDF) are the most outstanding colonizers of contaminated soil.**
- **Manganese peroxidase and endo-1,4-ß-glucanase are the main enzymes produced by fungi in bark and soil.**
- **White-rot fungi and LDF are able to degrade soil organic matter during a pretreatment process using pine bark as substrate.**
- **Scots pine bark promotes fungal growth and production of extracellular enzymes (MnP).**

POTENTIAL FULL-SCALE APPLICATION (*in situ* OR *ex situ*)

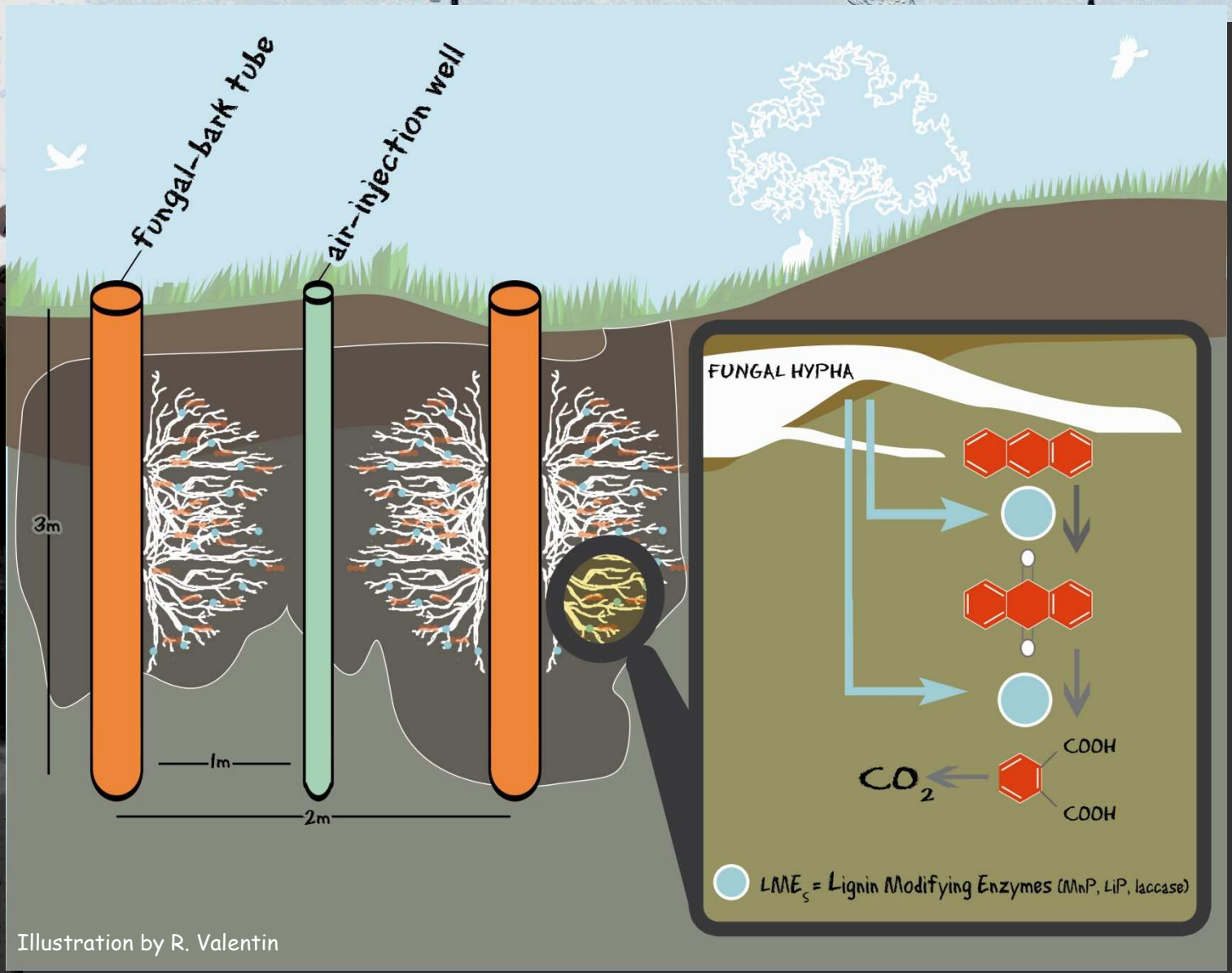
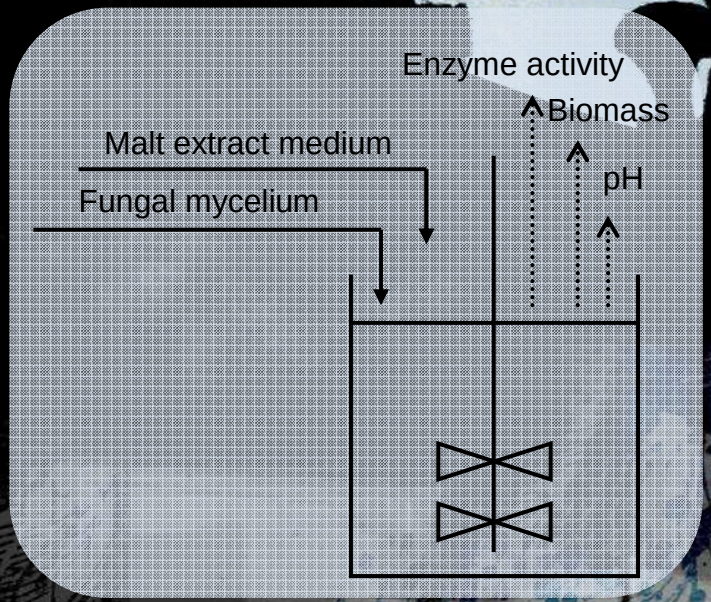


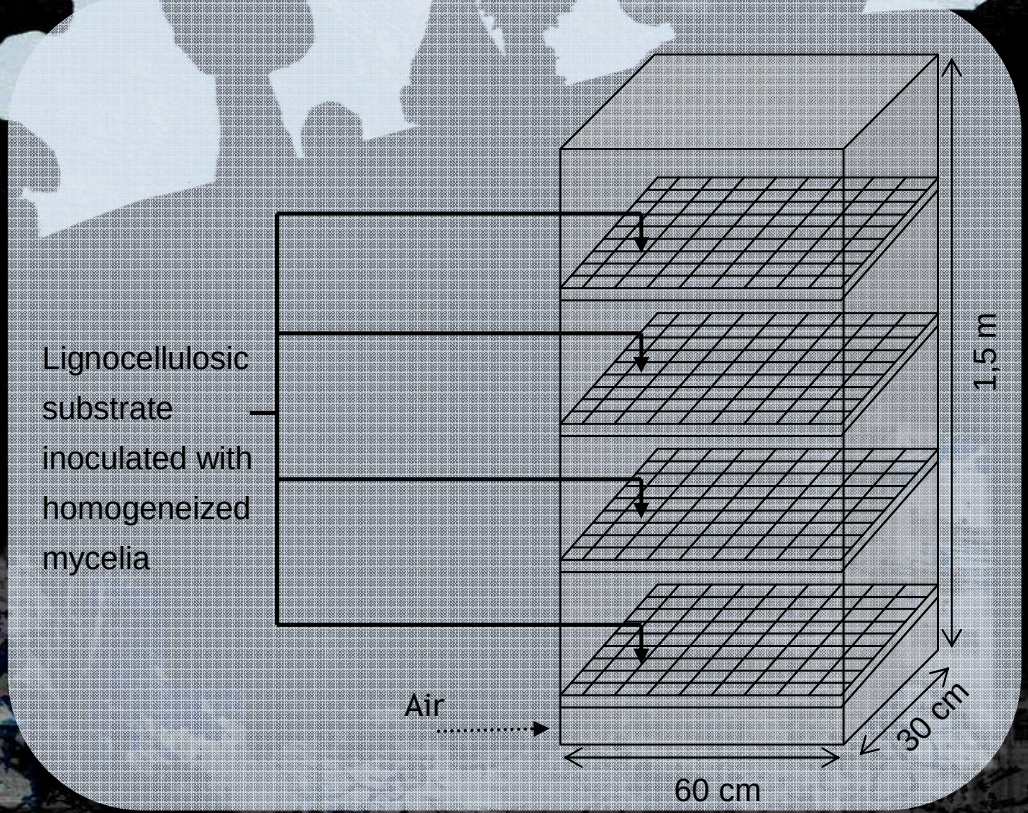
Illustration by R. Valentin

POTENTIAL FULL-SCALE APPLICATION
Production of fungal inocula

1st step: Production of liquid inocula in continuous-stirred tank reactor (CSTR).

2nd step: Production of fungal inocula in lignocellulosic substrate (bark) in aerated and ste[illegible]sterilized st[illegible]chambers.

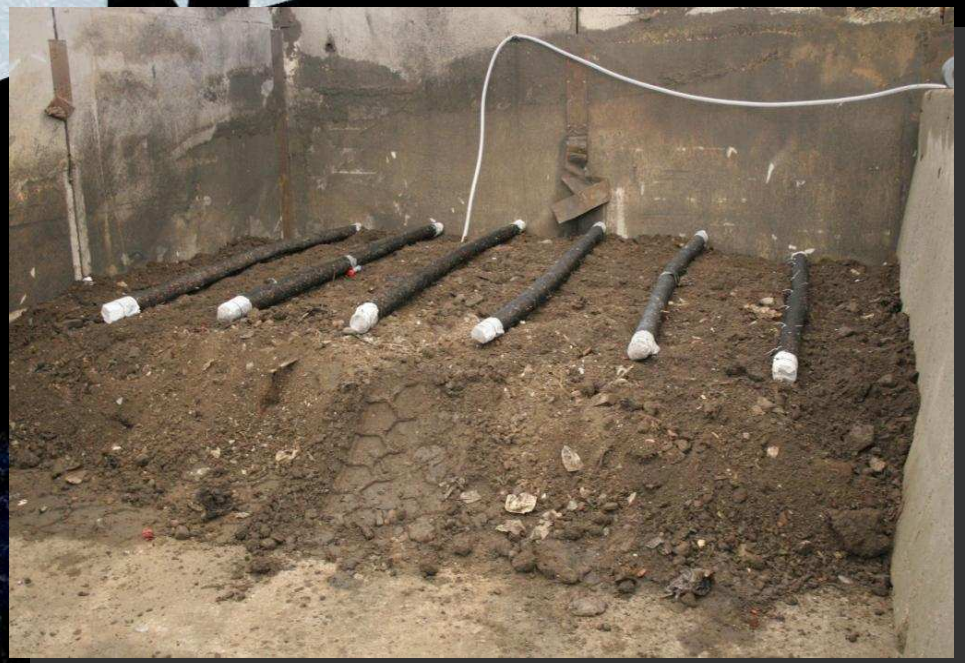




POTENTIAL FULL-SCALE APPLICATION
Introduction of fungal inocula into soil

Perforated tubes filled with fungus

growing on selected lignocellulosic substrate



FUNGI FOR CLEANING-UP OIL SPILLS AND OTHER CONTAMINATED SITES

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