Susceptibility of Bamboo to Fungi

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Content

- **Fungal Isolation and Identification**
- **Blue-stain Test**
- **Degradation Test**
- **Micromorphological Studies of Degradation**

Moulded bamboo after ship transport from Asia

Methods of identification fungi:

Classical identification methods

- colony morphology
- vegetative spore formation
- fruiting body formation

Modern identification method

- DNA sequencing analysis

Disadvantage of classical identification methods:

- Morphological characters between two species are too similar to distinguish these from each other.
- Morphological characters within a species are very variable so that two individuals are not necessarily recognized to belong to the same species.

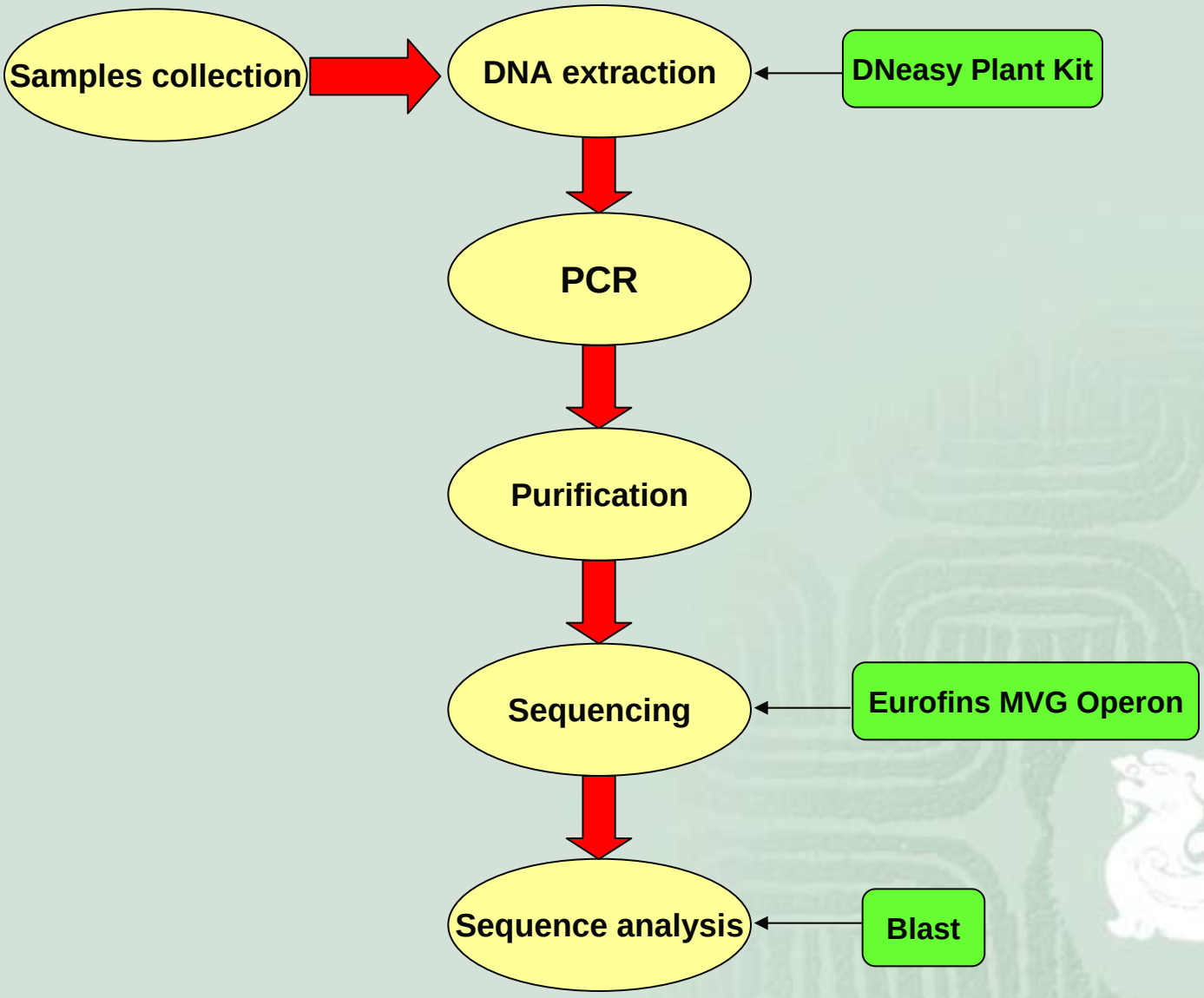







Pleurotus ostreatus ***Schizophyllum commune*** ***Trametes versicolor***

Molecular Methods of fungal identification



Samples collection

Country	Number of isolates	Number of identified isolates
Ethiopia	5	1
China	25	18
Costa Rica	9	1
Germany	6	5
Indonesia	5	1
Philippines	15	2
Thailand	43	19
Vietnam	42	29
	150	76

Botryosphaeria subglobosa
(Vietnam)

Epicoccum nigrum
(Thailand)

Penicillium commune
(China)

Identified isolates from bamboo by DNA sequencing (1)

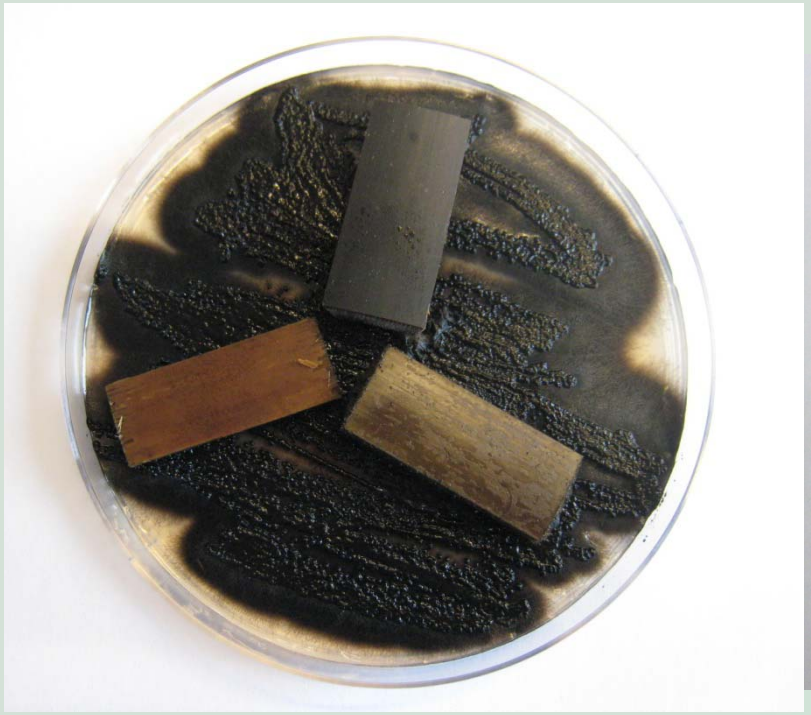
Country	**Deuteromycetes/Ascomycetes** (number of strains)	**Basidiomycetes** (number of strains)
Ethiopia		*Schizophyllum commune* (1)
China	*Alternaria alternata* (1) *Alternaria tenuissima* (1) *Arthrinium phaeospermum* (1) *Cladosporium cladosporioides* (2) *Dothiorella gregaria* (1) *Fusarium asiaticum* (1) *Fusarium culmorum* (1) *Fusarium zeae* (1) *Nigrospora oryzae* (4) *Penicillium commune* (1) *Penicillium chrysogenum* (1) *Penicillium tricolor* (1) *Penicillium variabile* (1) *Phoma macrostoma* (1)	
Costa Rica		*Schizophyllum commune* (1)
Germany	*Trichoderma koningiopsis* (2) *Trichoderma viride* (3)	
Indonesia		*Cyathus stercoreus* (1)

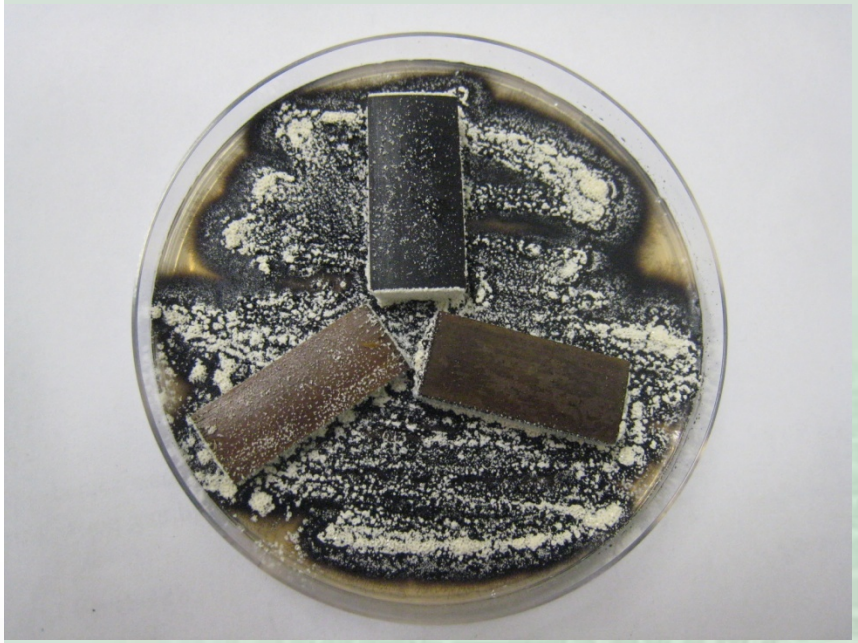
Identified isolates from bamboo by DNA sequencing (2)

Philippines	*Penicillium citrinum* (1) *Penicillium sumatraense* (1)	
Thailand	*Aspergillus nomius* (1) *Aspergillus repens* (1) *Botryosphaeria subglobosa* (1) *Cladosporium cladosporioides* (2) *Epicoccum nigrum* (2) *Penicillium brevicompactum* (1) *Penicillium citrinum* (2) *Penicillium pinophilum* (1) *Trichoderma atroviride* (1) *Trichoderma koningiopsis* (1)	*Schizophyllum commune* (6)
Vietnam	*Apiospora montagnei* (2) *Arthrinium phaeospermum* (1) *Arthrinium sacchari* (3) *Aspergillus flavus* (5) *Aspergillus niger* (2) *Botryosphaeria subglobosa* (5) *Epicoccum nigrum* (4) *Penicillium bialowiezense* (1) *Penicillium biourgeianum* (1) *Penicillium brevicompactum* (2) *Penicillium expansum* (1) *Penicillium islandicum* (1) *Pestalotiopsis microspora* (1)	
	67	9

Blue-stain Test

- inoculate bamboo with blue stain
- embed bamboo in PEG
- cut to 10 μm sections with microtome
- visualize by light microscopy.

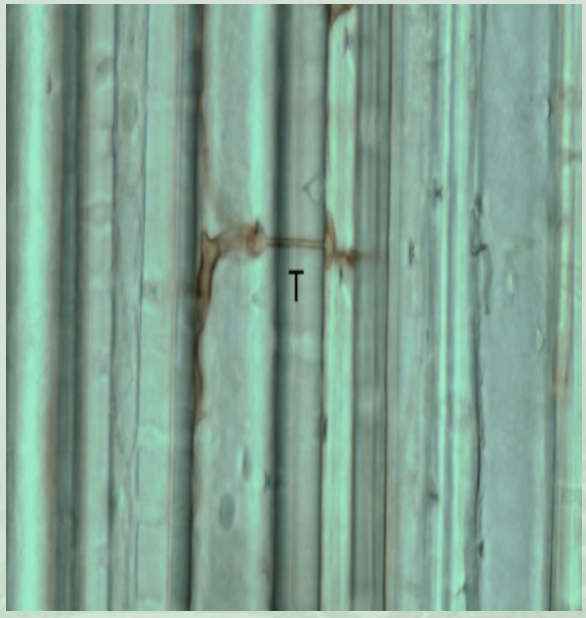




Bamboo samples inoculated with *Aureobasidium pullulans* after 1 month (left) and 5 months (right)

thick, brown hyphae of *Cladosporium cladosporioides* in a vessel of *Phyllostachys pubescens* (100x)

brown chlamydospores of *Botryosphaeria subglobosa* in *Gigantochloa atroviolacea* (100x)



transpressorium (T) of *Alternaria alternata* in *Bambusa maculata* (600x)

Degradation Tests

- **Degradation in preserving jars**
- **Degradation in metal tubs ("Fungus cellar-test")**

Degradation test in preserving jars

- inoculate fungi in preserving jars
- cut bamboo samples into small pieces and autoclaved
- place bamboo samples in preserving jars two weeks after inoculation
- evaluate mass loss according to EN113

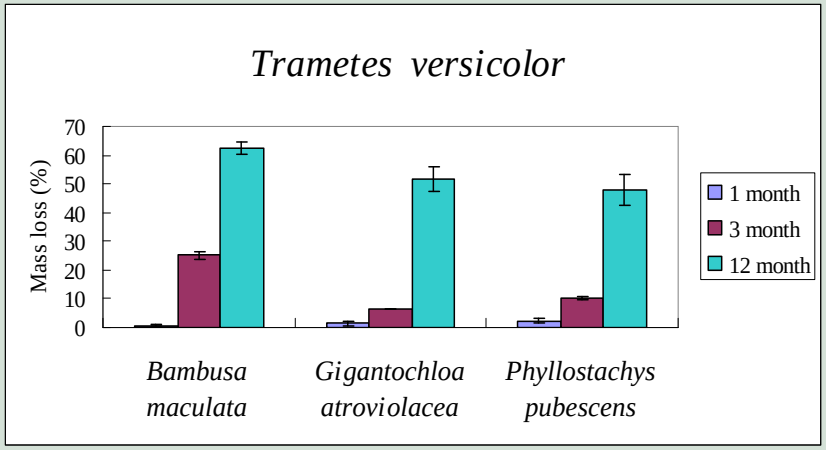
  after 1, 3, 12 months of incubation

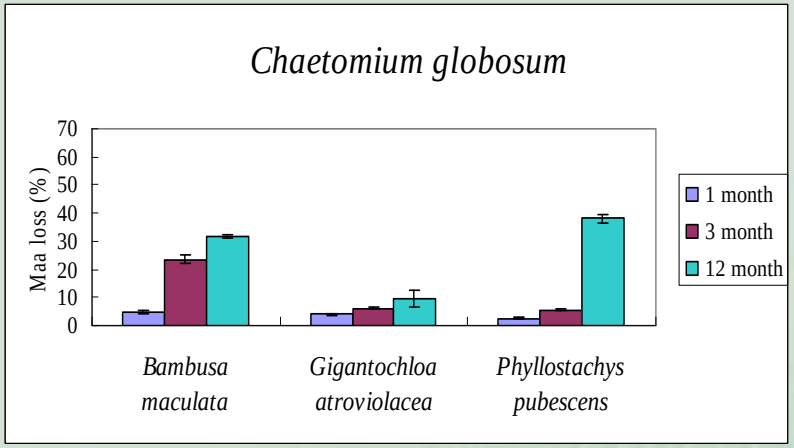
Degradation test in preserving jars

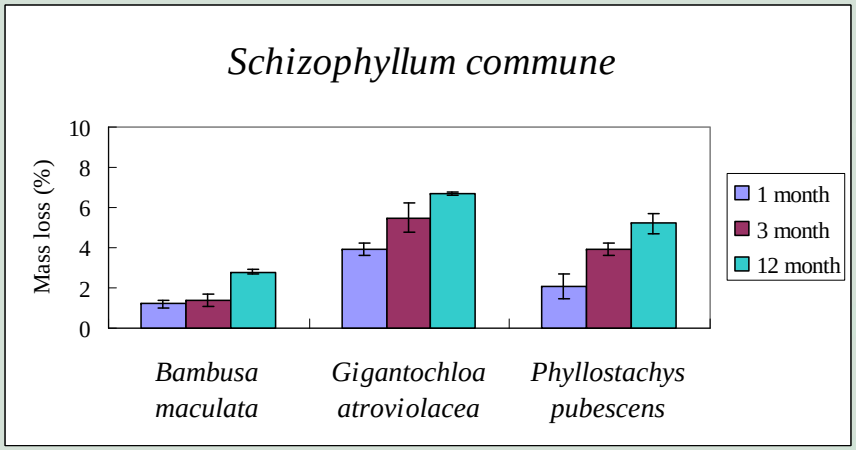
Samples in preserving jars

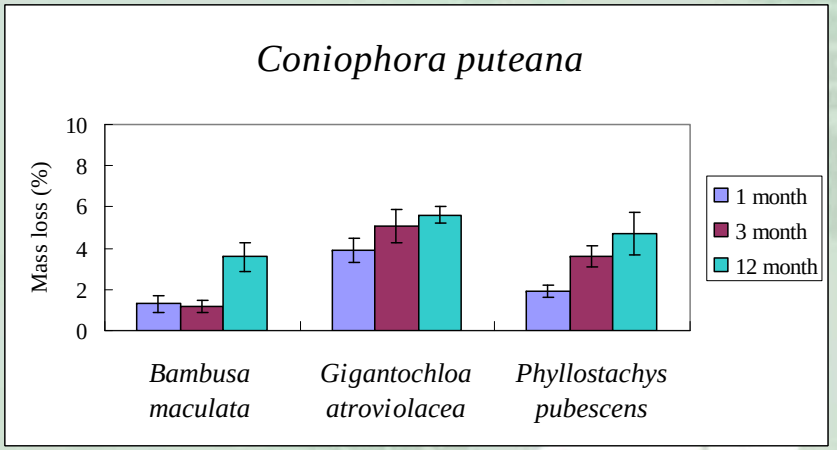
Samples with mycelium of *Trametes versicolor* after 1 week incubation

Mass loss (%) of bamboo in preserving jars





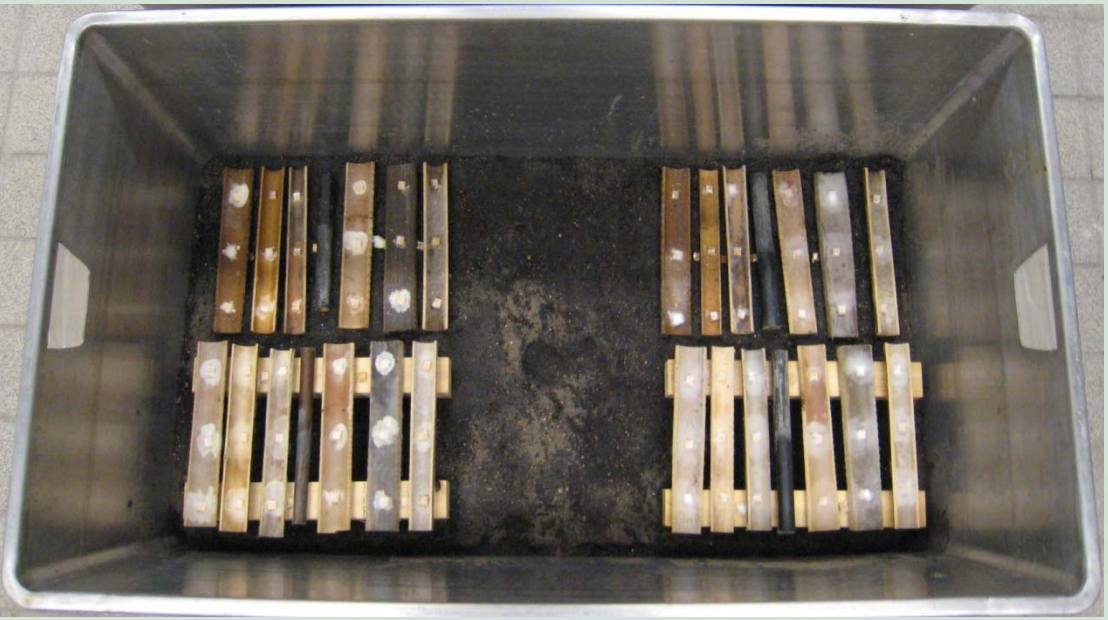




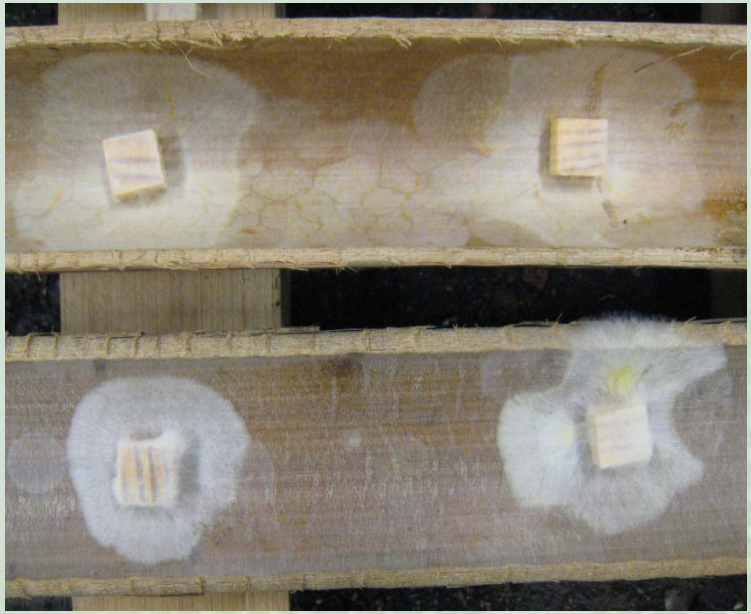
Degradation test in metal tubs

- fill with 30 litres of compost soil in metal tubs
- autoclave bamboo samples
- place samples either on autoclaved wood supports or directly on the soil
- inoculate samples with fungi
- cover the tubs with glass
- moisten the soil weekly by spraying tap water
- evaluate mass loss and moisure content after 1 year

Degradation test in metal tubs (“Fungus cellar-test”)



Samples in the “Fungus cellar”



Samples with mycelium of *Schizophyllum commune* after 1 week incubation

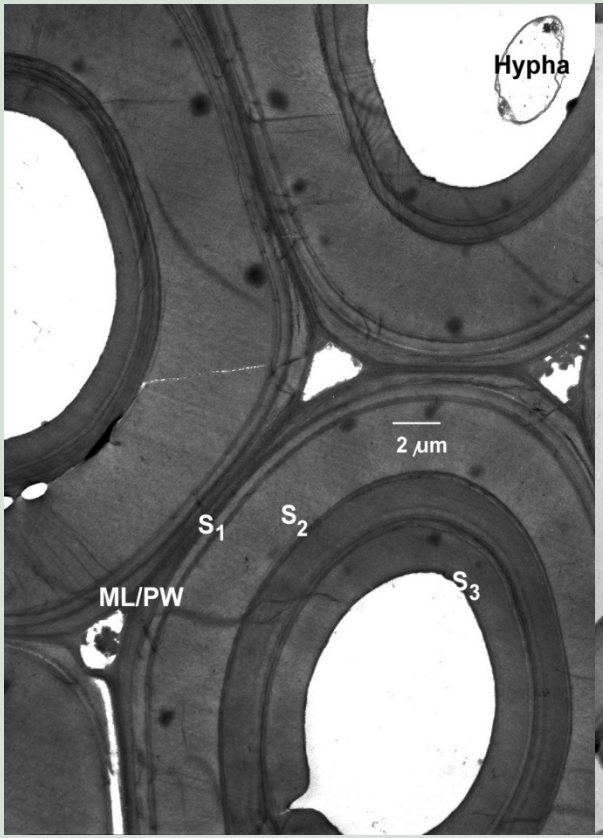
Mass loss (ML,%) and moisture content (MC, %u) of bamboo in metal tubs

Bamboo	Soil contact	*Coniophora puteana*		*Schizophyllum commune*	
		ML (%)	MC (%u)	ML (%)	MC (%u)
Arundinariaamabilis	Yes	15.5	187	15.3	148
	No	**41.2**	57	7.9	54
Bambusa maculata	Yes	9.9	159	11.0	174
	No	36.3	50	4.3	35
Dendrocalamus asper	Yes	5.4	104	5.1	90
	No	27.7	48	4.1	32
Gigantochloa atroviolacea	Yes	6.1	95	6.1	95
	No	42.3	58	4.0	41
Phyllostachys nigra	Yes	9.7	112	16.4	126
	No	40.2	61	6.5	46
Phyllostachys nigra Boryana	Yes	35.3	103	**19.7**	182
	No	38.4	53	5.4	38
Phyllostachys pubescens	Yes	6.3	61	6.3	63
	No	34.8	43	6.0	34

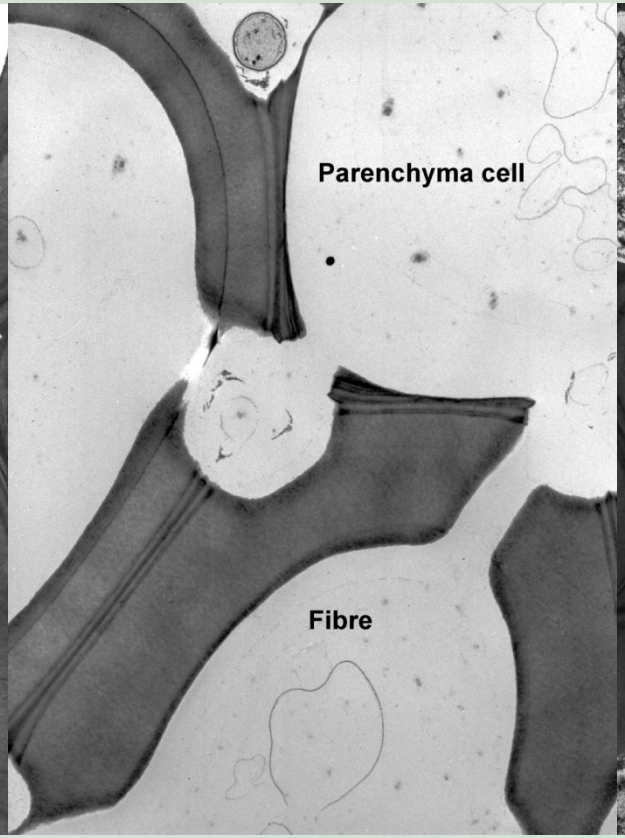
Micromorphological Study of Degradation

- fix bamboo pieces after degraded by fungi
- dehydrate and embed bamboo samples
- section samples on an ultramicrotome with a diamond knife
- examine with transmission electron microscope (TEM)

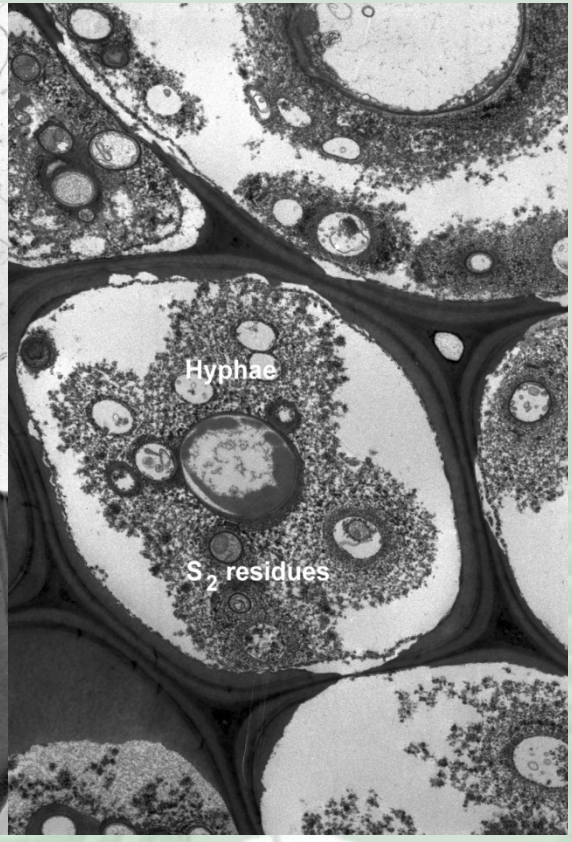
Micromorphological Study of Degradation



Early brown-rot symptoms in *Gigantochloa atroviolacea* by *Coniophora puteana*



Medium white-rot decay in *Bambusa maculata* by *Trametes versicolor*

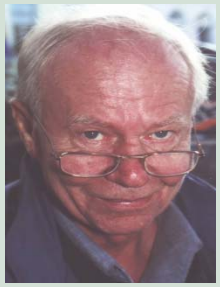


Severe soft-rot degradation in *Bambusa maculata* by *Chaetomium globosum*

Summary

- 150 strains were isolated and 76 isolates were identified.

  Most isolates were ascomycetes related fungi imperfecti, only two basidiomycetes were identified.
- All bamboo species investigated can be colonized by the various groups of fungi, namely moulds, staining and rot fungi.
- The tissue was colonized by the typical thick, brown hyphae and by chlamydospores of blue stain fungi. In some cases, transpressorium was observed, which is the only fungal `organ´ of blue stain fungi penetrating lignified cell walls.
- Considerable degradation occurs by white-, brown- and soft-rot fungi. Soil moisture influenced some fungi.
- Fungi produced the typical wood degradation patterns in the bamboo cell-wall.



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Thank you for your attention!