

Of Microbes and Wood

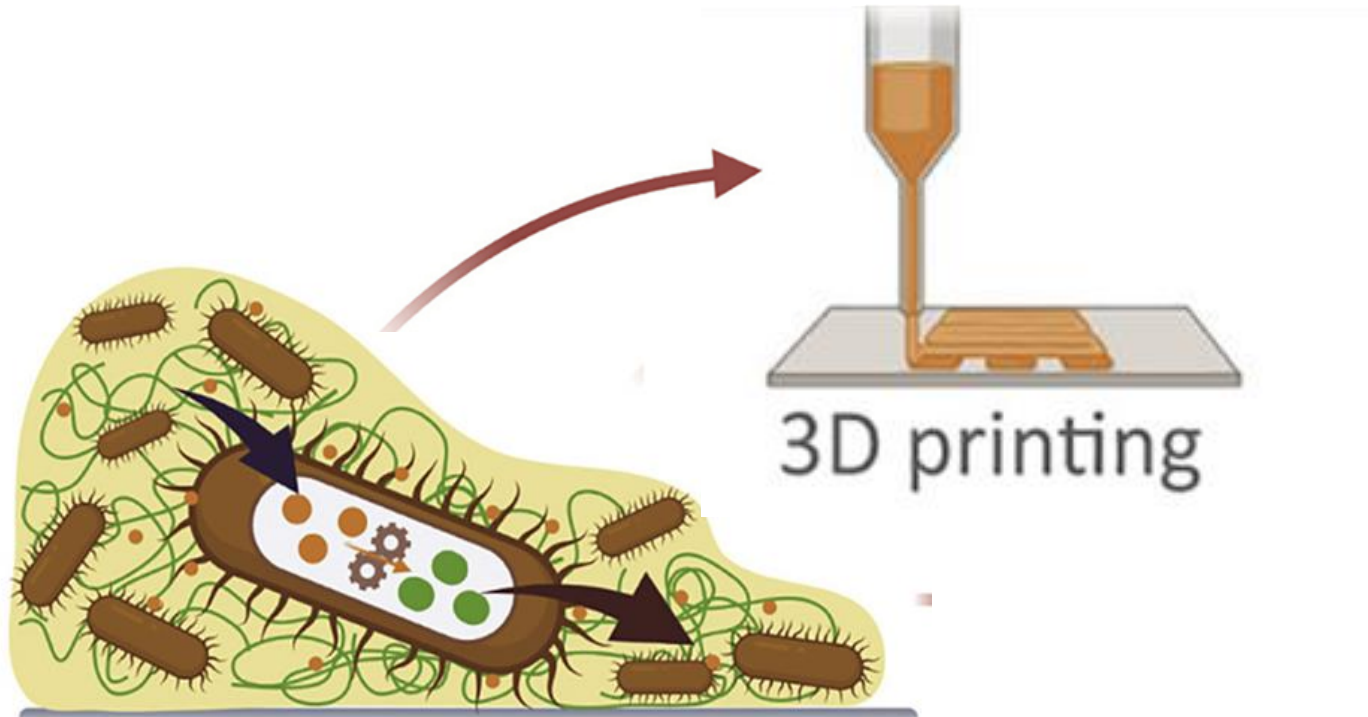
From microbial wood decay to
microbial wood engineering

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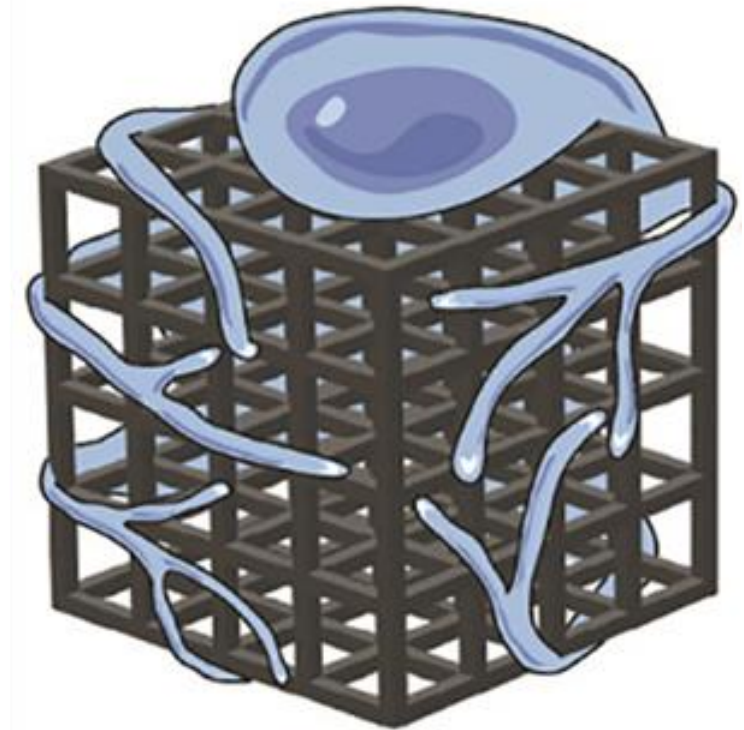
Using Organisms to Make Materials

Biological Engineered Living Materials (Bio-ELMs)



Biofilm

Hybrid Living Materials (HLMs)



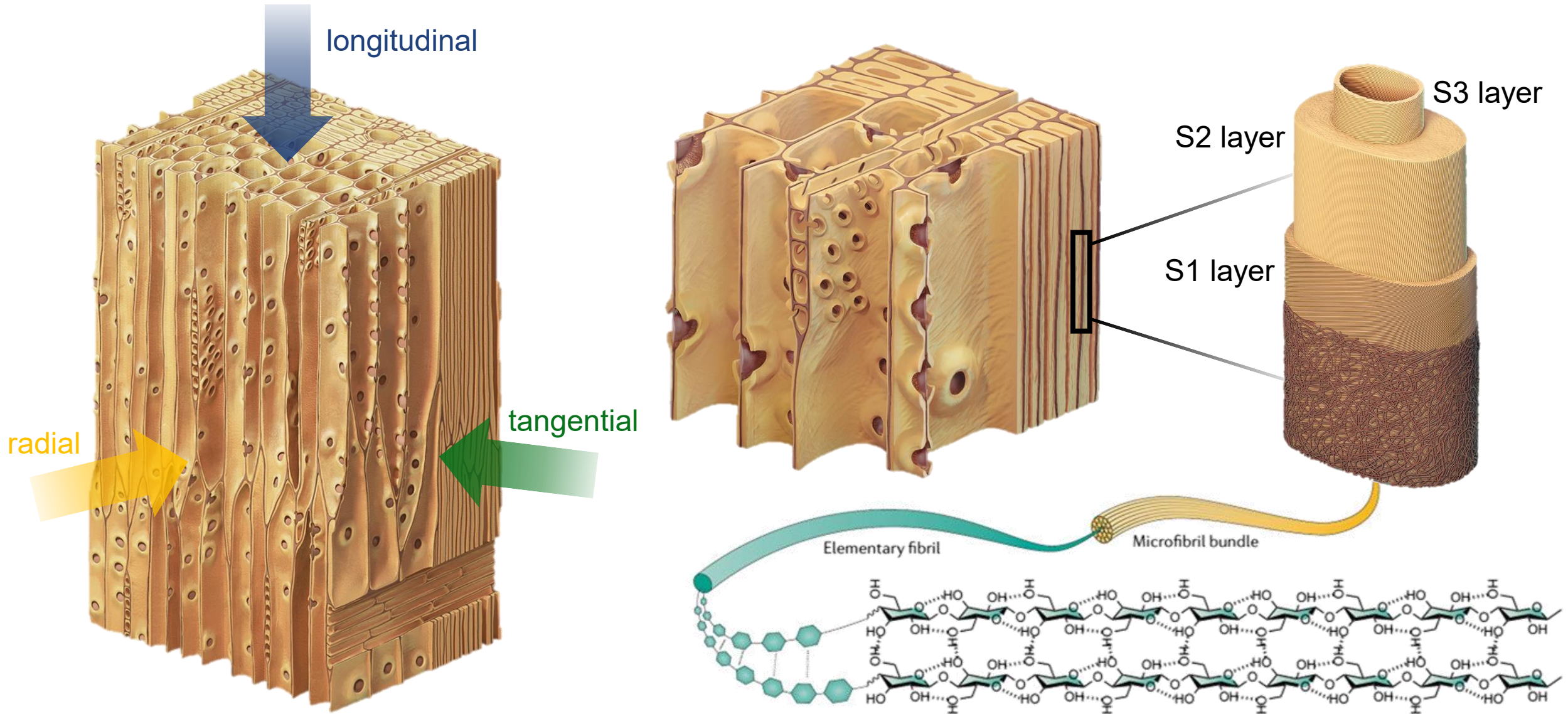
e.g. cells cultured on abiotic scaffold

Wood

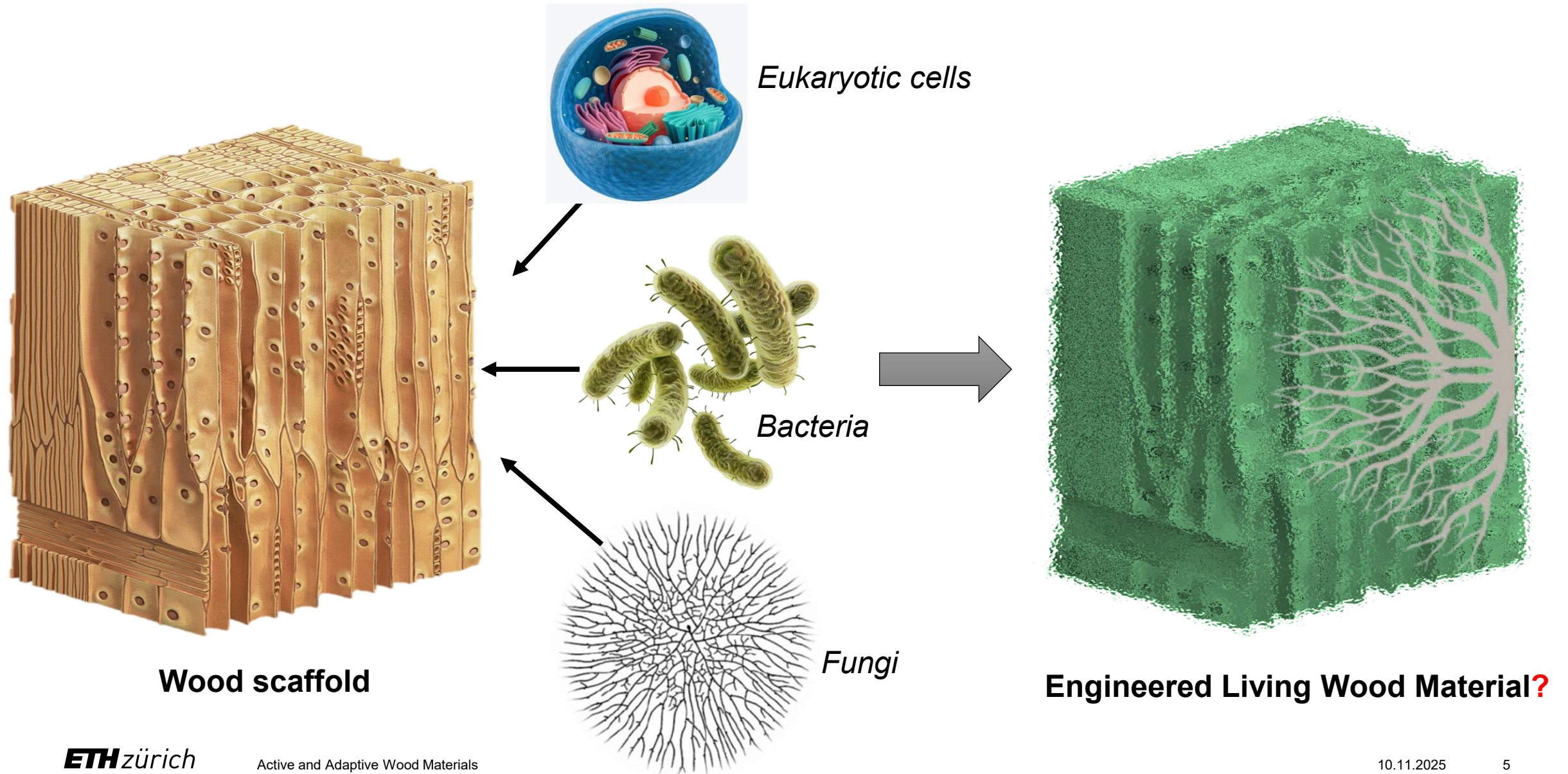
Wood



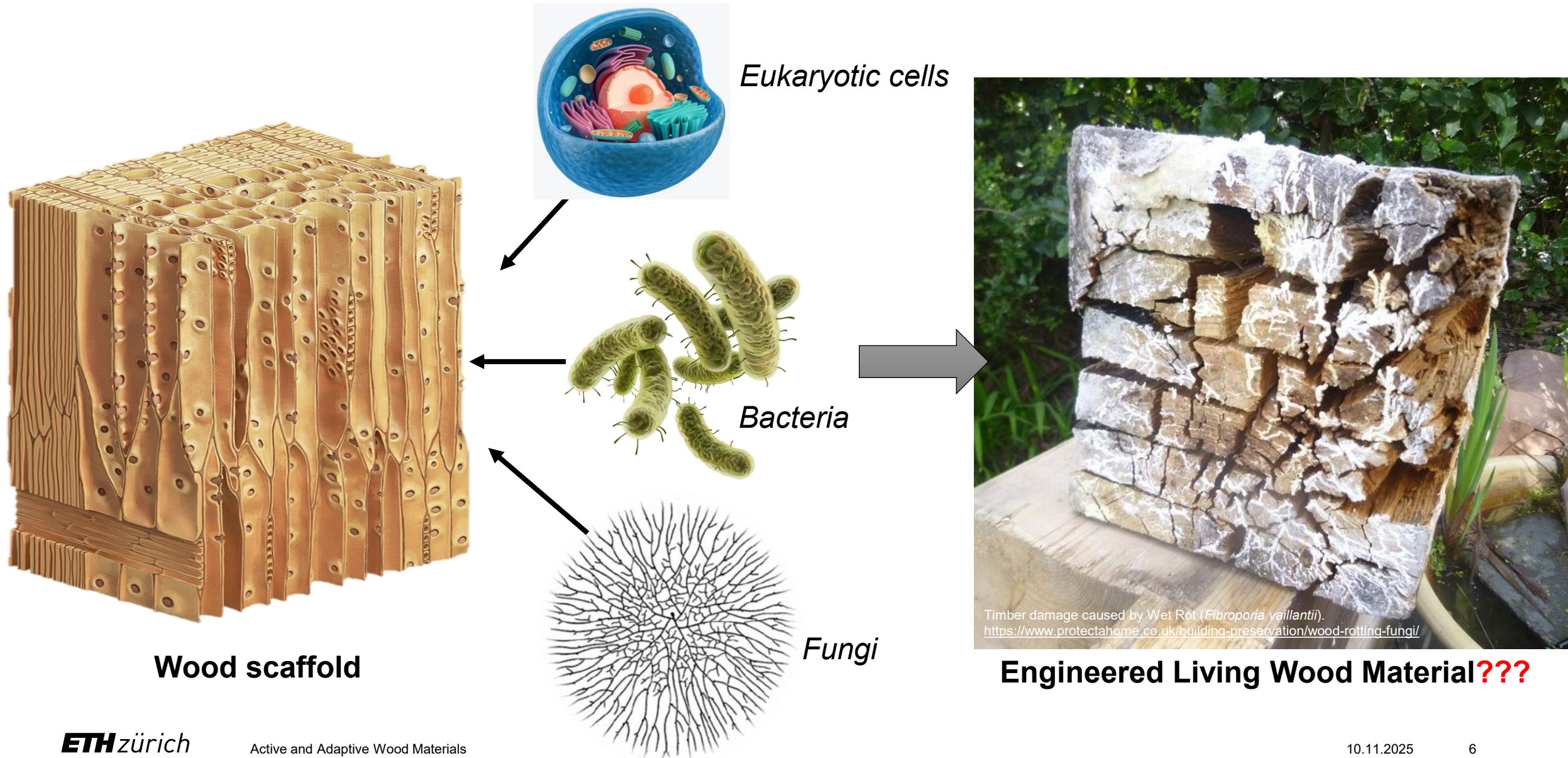
Wood: a unique type of material



Engineered Living Wood Materials: is it all that easy?



Engineered Living Wood Materials: is it all that easy? Not really...



Wood: radically different perspectives on the same subject

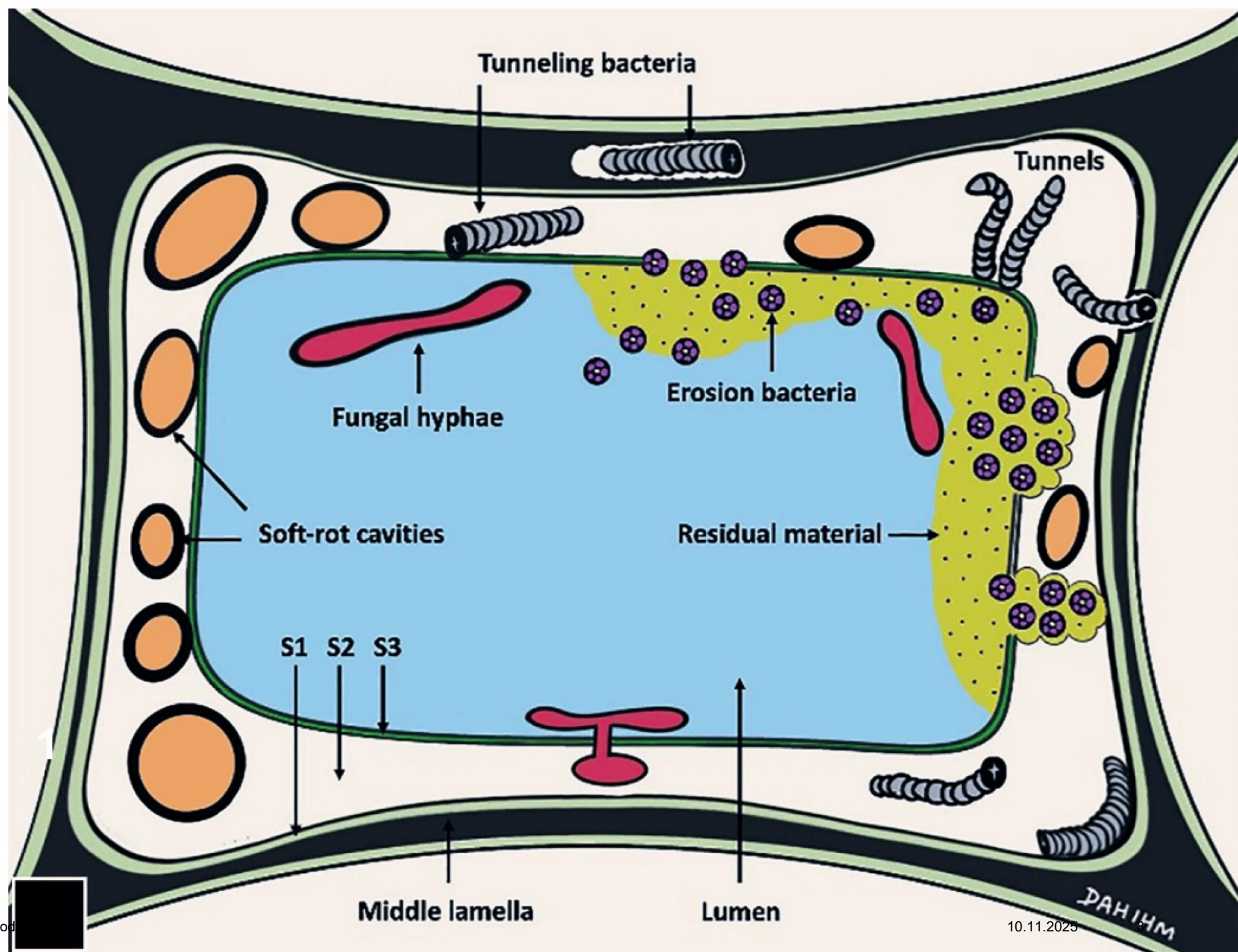
Human perspective



Microbe (e.g. fungi) perspective

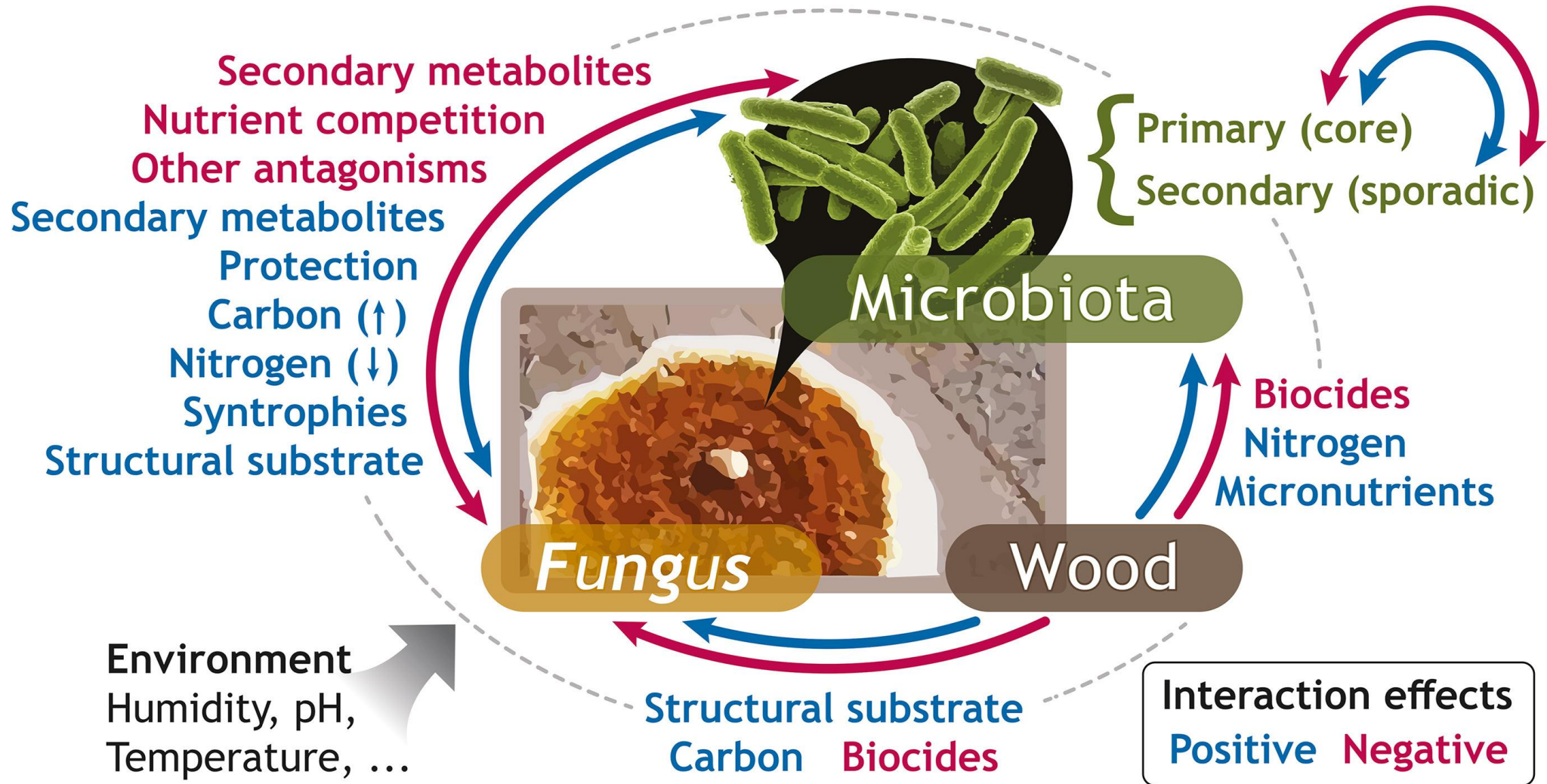


Microbial wood degradation

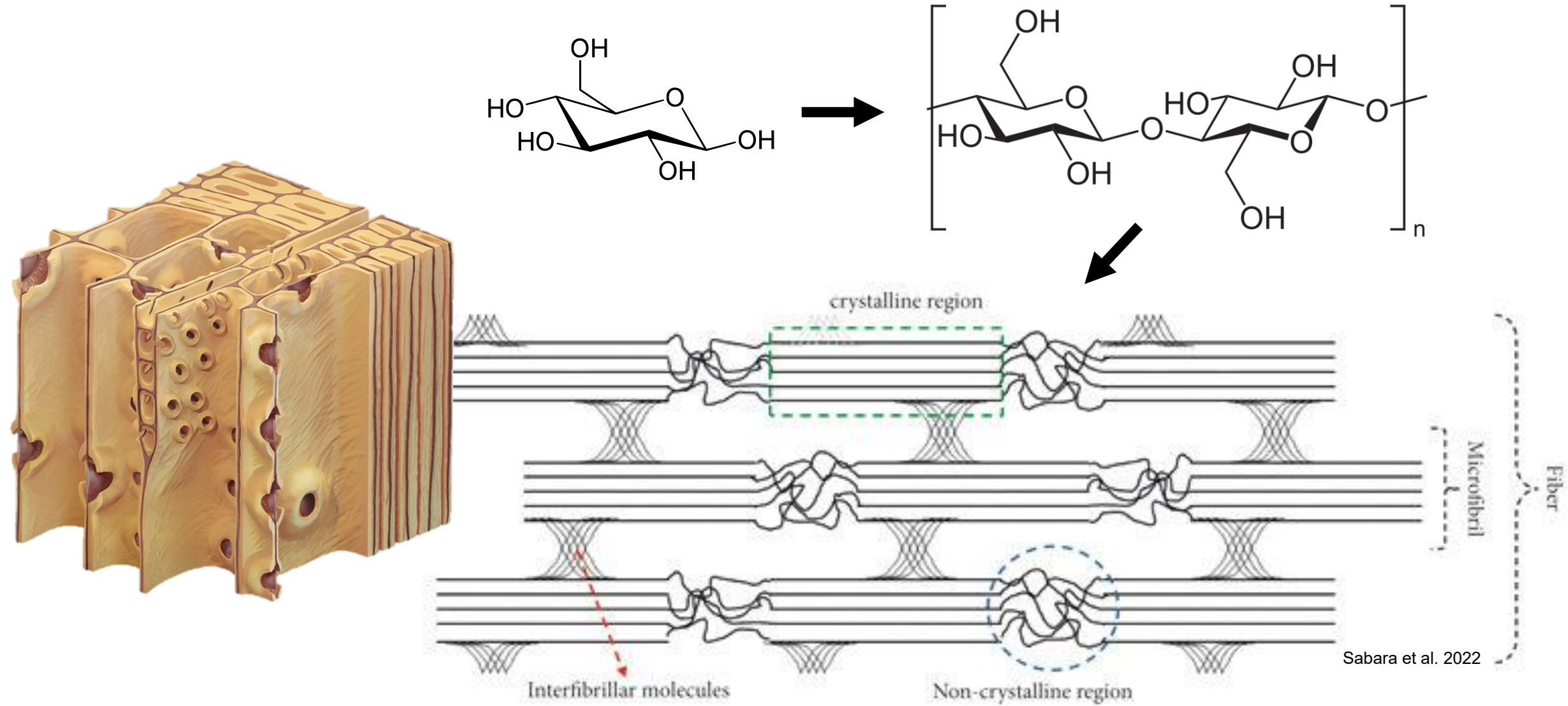


Singh et al. *Forests* 2022, 13(3), 394

Complex interrelations between wood and microbes



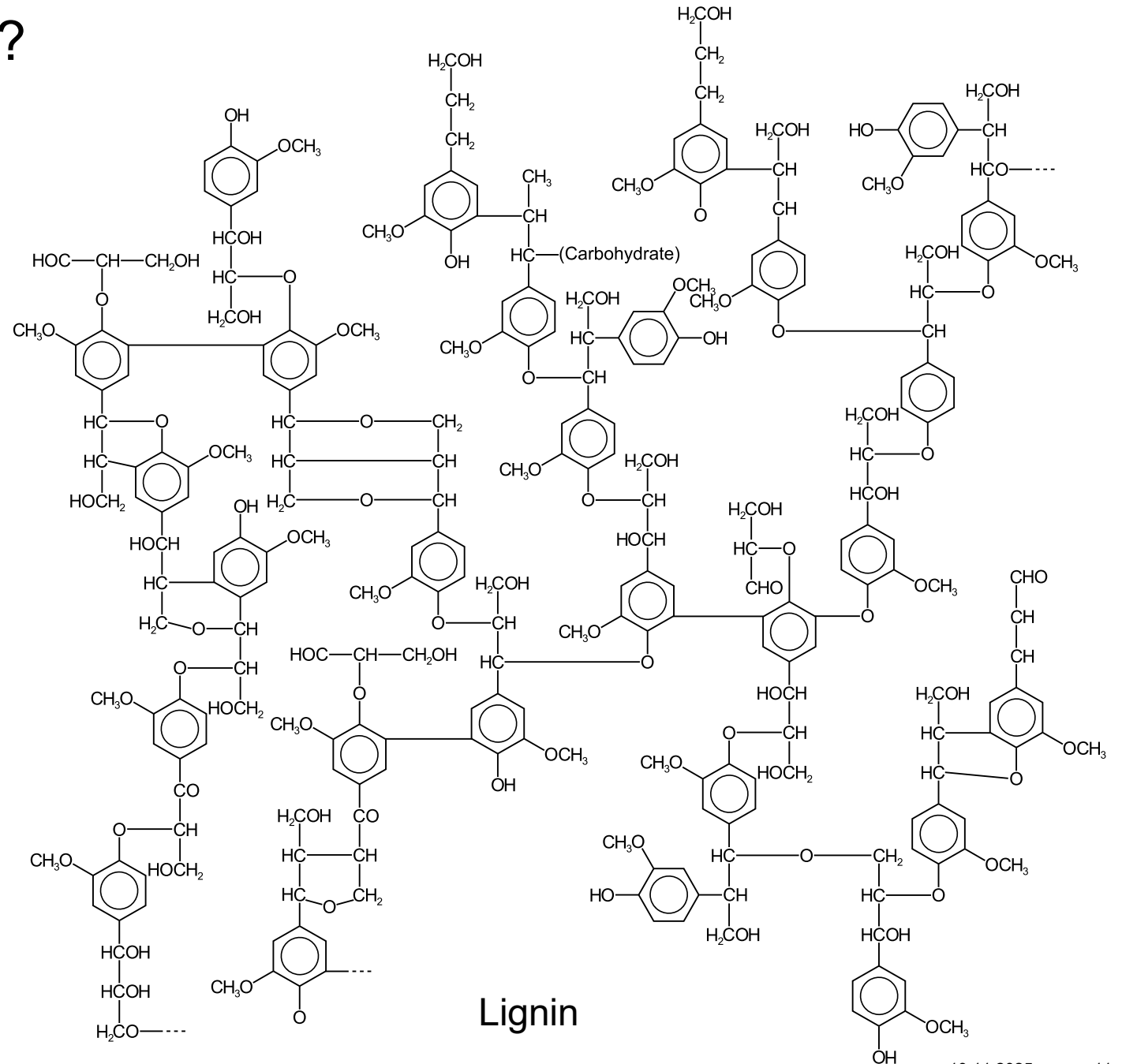
Why do microbes eat wood?



Sabara et al. 2022

Cellulose

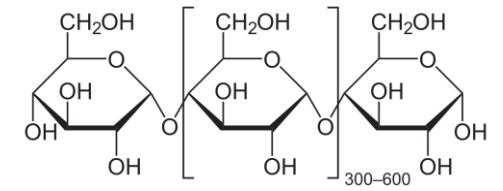
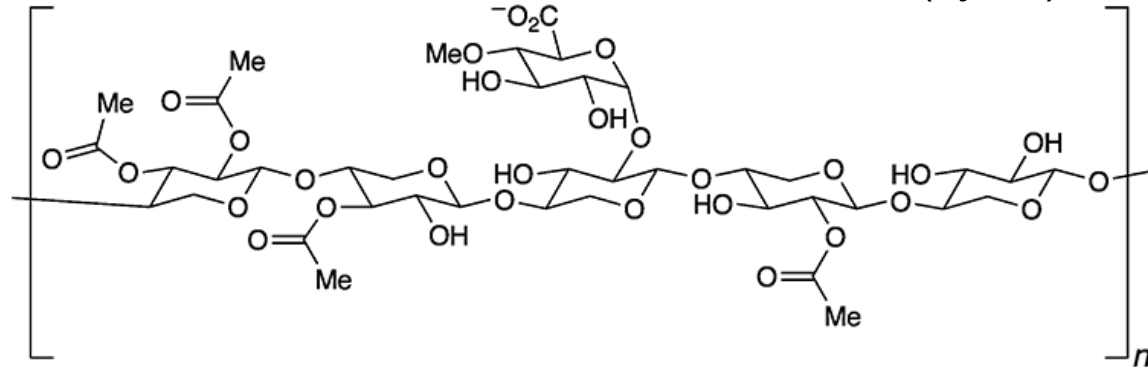
Why do microbes eat wood?



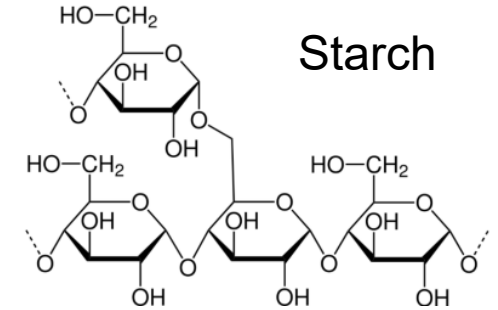
Why do microbes eat wood?



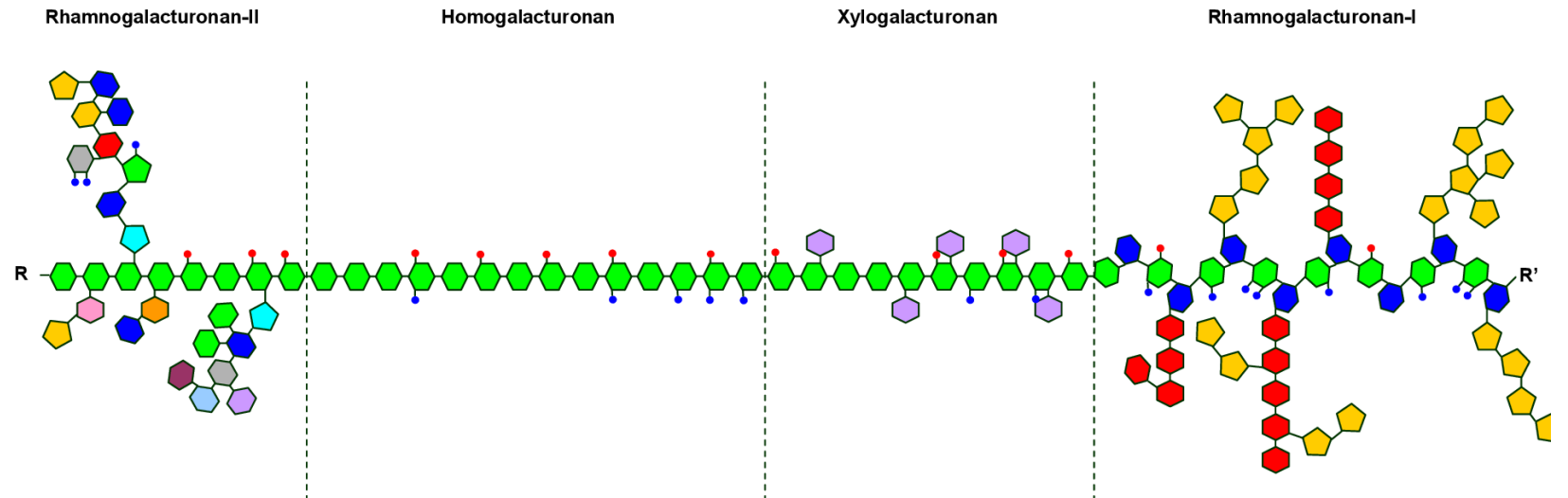
Hemicellulose (xylan)



Starch



Pectin



D-Galacturonic acid	D-Xylose	3-deoxy-D-lyxo-heptulosaric acid
L-Rhamnose	D-Glucuronic acid	2-keto-3-deoxy-D-manno-octulosonic acid
L-Fucose	L-Galactose	Acetyl groups
D-Galactose	D-Apiose	Methyl groups
L-Arabinose	3-C-carboxy-5-deoxy-L-xylose	

Scheller et al. 2007

Wood-degrading fungi

Fungi can cause a variety of decay types in wood: mold, blue stain (sapstain), white rot, brown rot, and soft rot.

«White rot»



«Brown rot»



Rate of colonization, decay, and removal of wood structural components varies greatly between rot types, fungal species, and even strains. Variations in the morphology of decay patterns in wood and wood cell walls reflect local variations in the microdistribution of wood chemical components.

NEVERTHELESS:

a certain fungus causes a specific type of decay and the decay will always be the same no matter what wood type.

Can the appetite of microbes for wood be put to good use?

YES!

Variety (of effects) + Consistency = possibility to choose the type of fungi to achieve a certain property!

Myco-Holz

=

Myco-Wood

(1964)



Original video:
MYKOHOLZ _ MYCOWOOD (2023, DE _ EN)
https://www.youtube.com/watch?v=afUf_6n1BAc

Mycowood pencil



Myco-Wood

Table 1 Overview of the Myko-Holz (myco-wood) related patents from Walter Luthardt

Year of application	Patent document	Content	Relation to <i>Myko-Holz</i>
1944	DD00000000292B1	[DE] Verfahren zur Züchtung holzbewohnender Pilze	Production of edible mushrooms, led to the invention of <i>Myko-Holz</i>
1951	DD000000003114A1	[DE] Verfahren zur Herstellung und Handhabung von Impfmateri- al in der praktischen Pilzkultur	Preparation of the inoculation paste
1951	DD000000002175A1	[DE] Verfahren zur Veredelung von Holz durch Steigerung verschiedener Eigenschaften, wie die der Schnitz- und Spitz- barkeit, Feuerfestigkeit und Schwimmfähigkeit	Evaluation of potential applications of <i>Myko-Holz</i>
1953	DE000000946845B	[DE] Verfahren zur Veredlung von Holz	production of <i>Myko-Holz</i>
1953	DE000000932998B	[DE] Verfahren zur Herstellung und Handhabung von Impfmateri- al in der Pilzkultur	Production of the inoculation paste
1957	DD000000022628A1	[DE] Schüttelkolben zur Anreicherung von Luftsauerstoff in Nährlösungen	Preparation of the inoculation paste
1958	DD000000021771A1	[DE] Glasform aus Holz	Application of <i>Myko-Holz</i> for glass industry
1959	DE000001063336A	DE] Schüttelkolben fuer biologische Zwecke, insbesondere zur Herstellung von Pilzkulturen	Preparation of the inoculation paste
1971	DD000000102553A1	[DE] Verfahren zur Herstellung von Pilzimpfmateri- al zur Übertragung von Pilzkulturen, insbesondere für Kulturen holzbewoh- nender Pilze	Preparation of the inoculation paste

...so, what's the problem?

TIME!

(AND fungi stability, biosafety, energy consumption → climate chambers...)

“Myco-wood”

The incubation of beech wood with the white-rot fungi *Pleurotus ostreatus* and *T. versicolor* has been applied to produce so-called “myco-wood” (Luthardt 1963; Schmidt 1994; Unbehaun et al. 2000). Depending on the incubation time, the fungal treatment reduced the density and loosened the structure of the wood. In consequence, the machinability and rate of water sorption were improved. “Myco-wood” was mostly used for pencils, rulers, drawing tables and wooden forms in the glass industry. During 1958–1965, 55×10^6 “myco-wood” pencils were produced in the former German Democratic Republic. At the moment, this technique is not applied in practice, but production is under consideration by some German pencil producers.

In general, wood mycologically modified by the Luthardt-method is characterized by the following attributes [10]:

- High and uniform porosity
- Low variation in raw density
- Stressless material (thus low warpage)
- Good insulating properties
- Low swelling
- Good heat and cold insulation
- High sound absorption
- High moisture absorption
- High absorption and good impregnability
- Good sharpenability

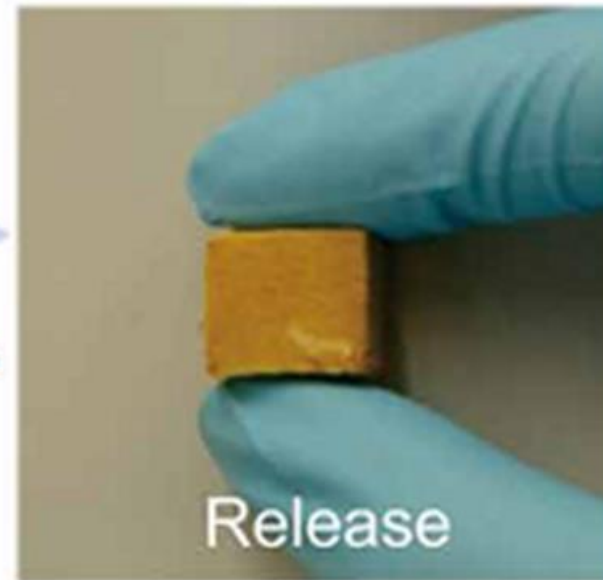
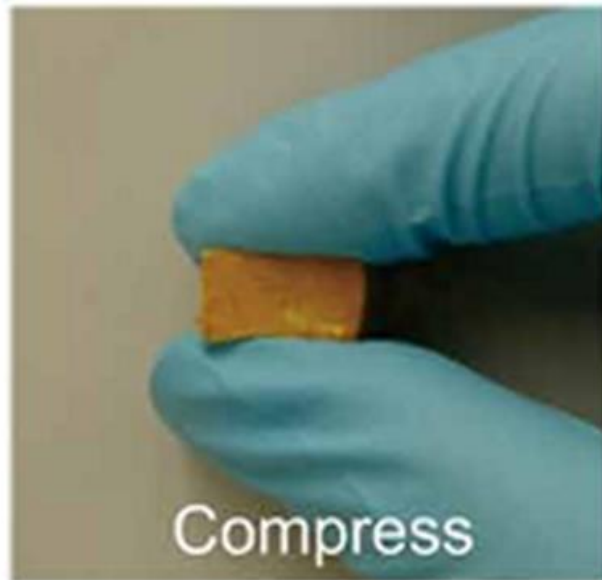
White rot-degraded balsa wood



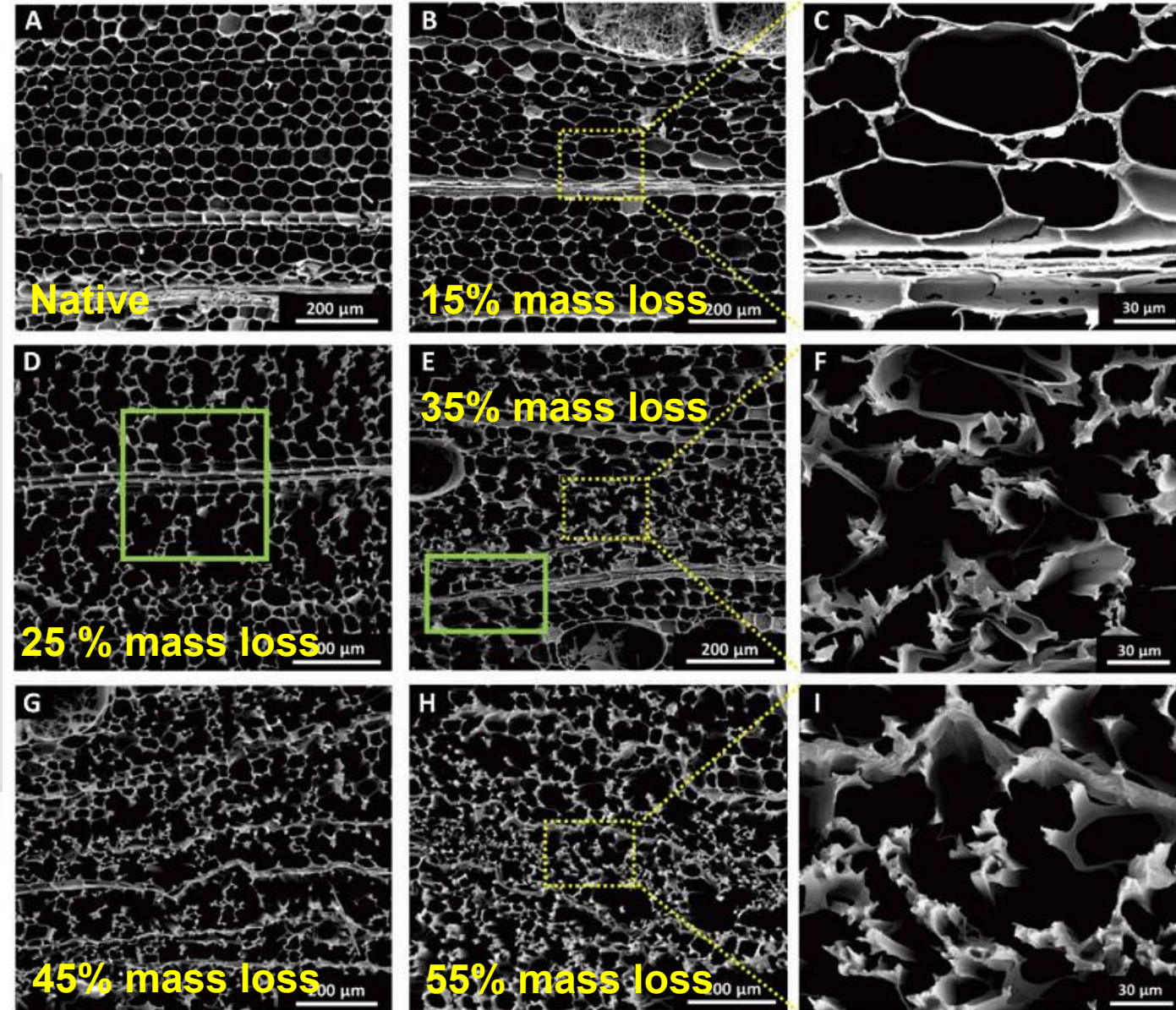
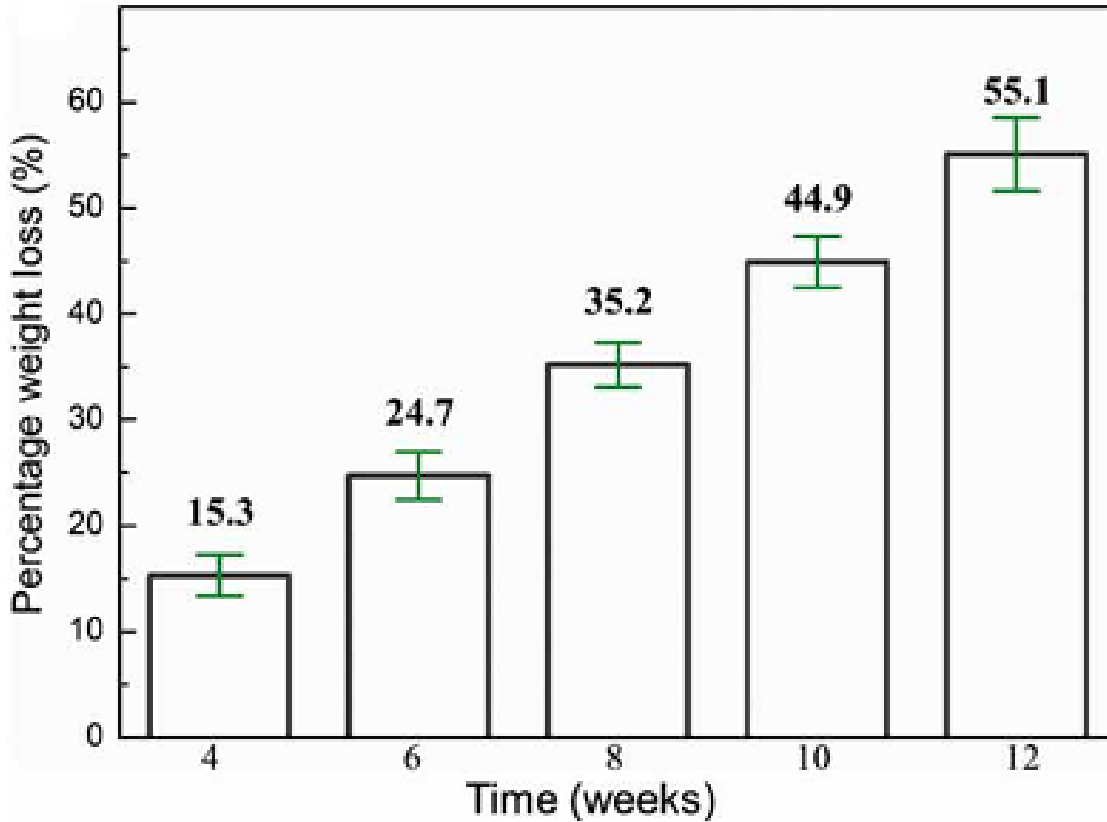
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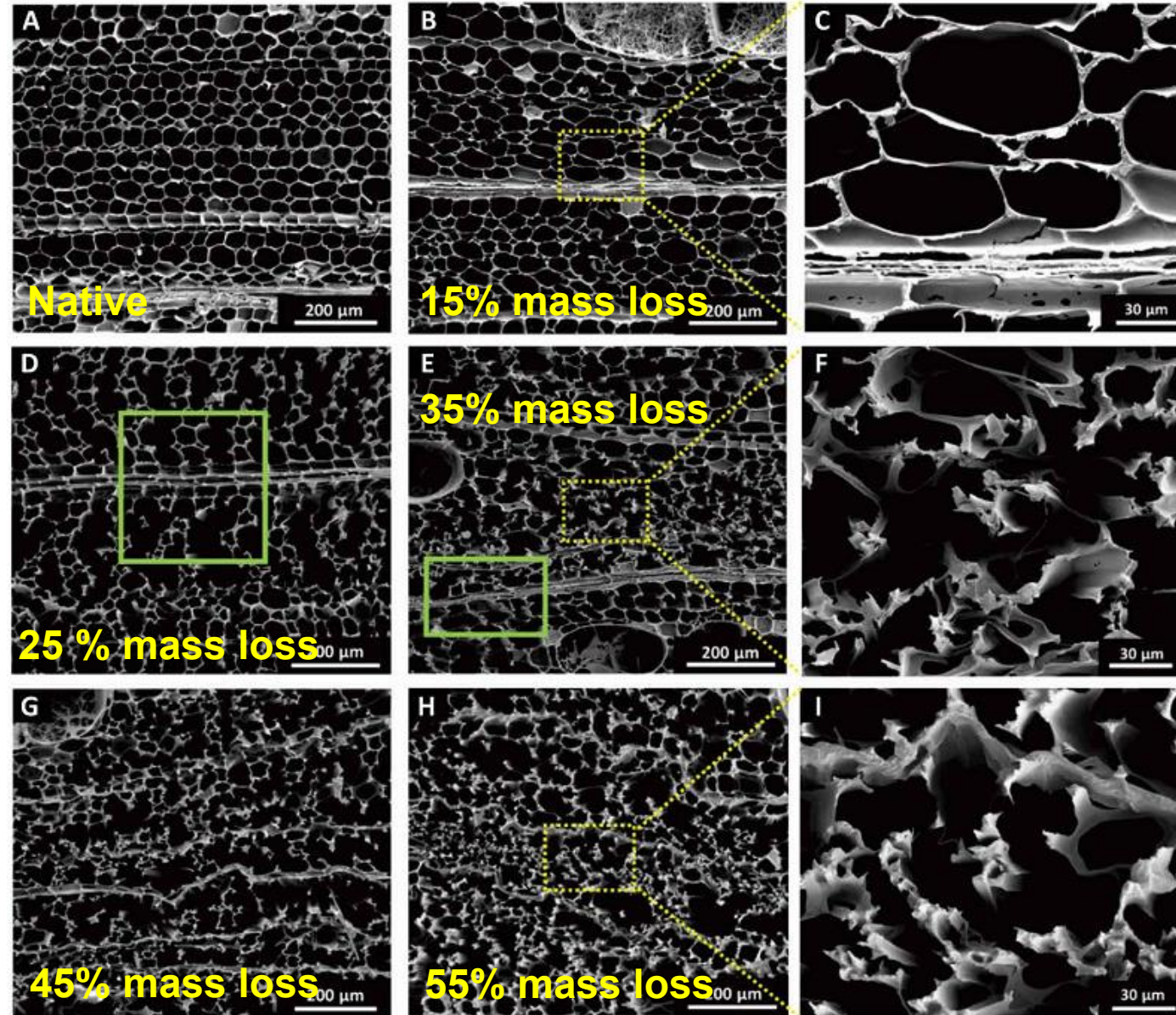
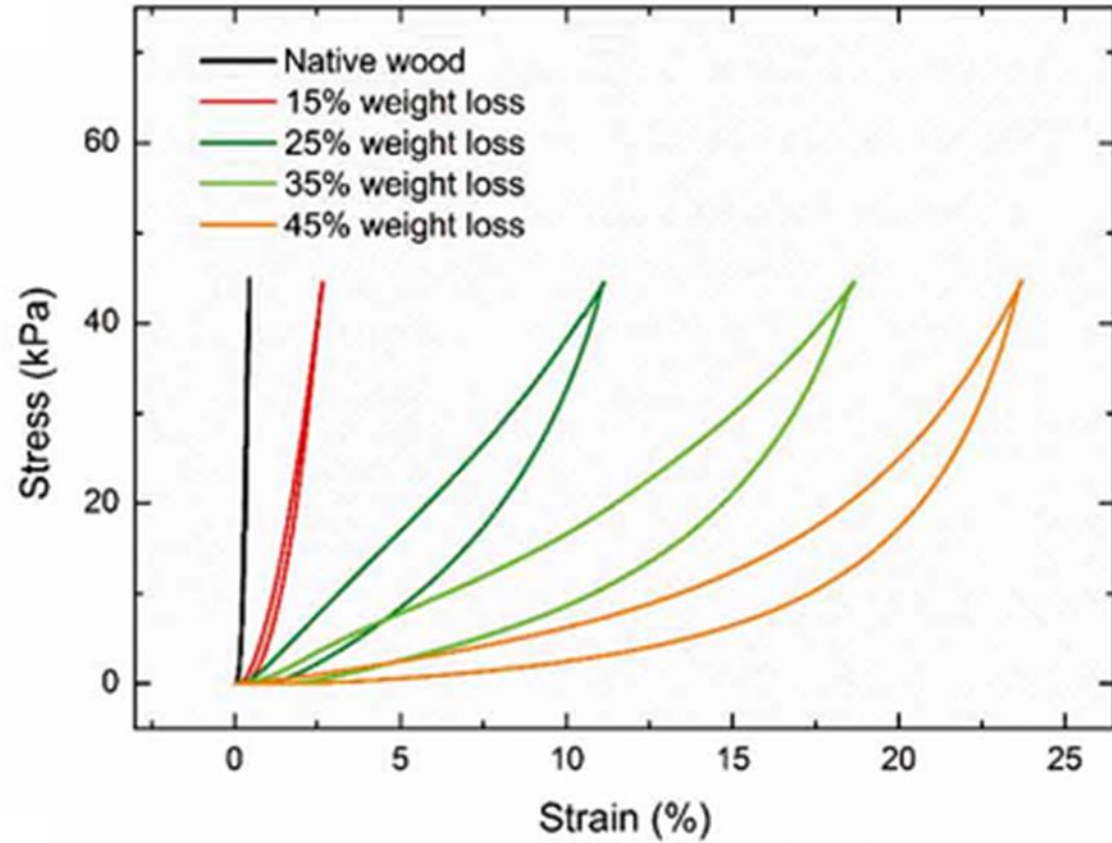
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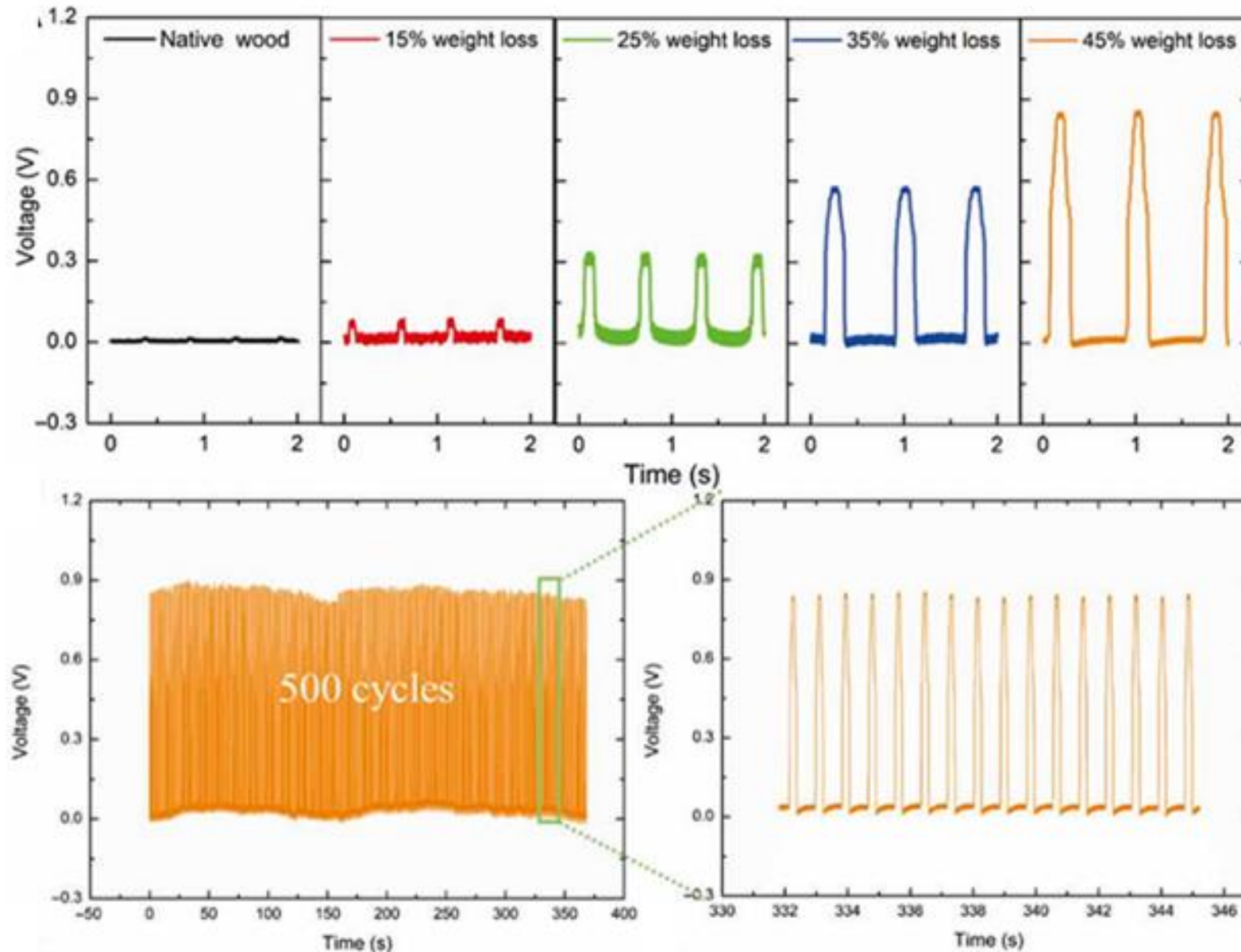
White rot-degraded balsa wood



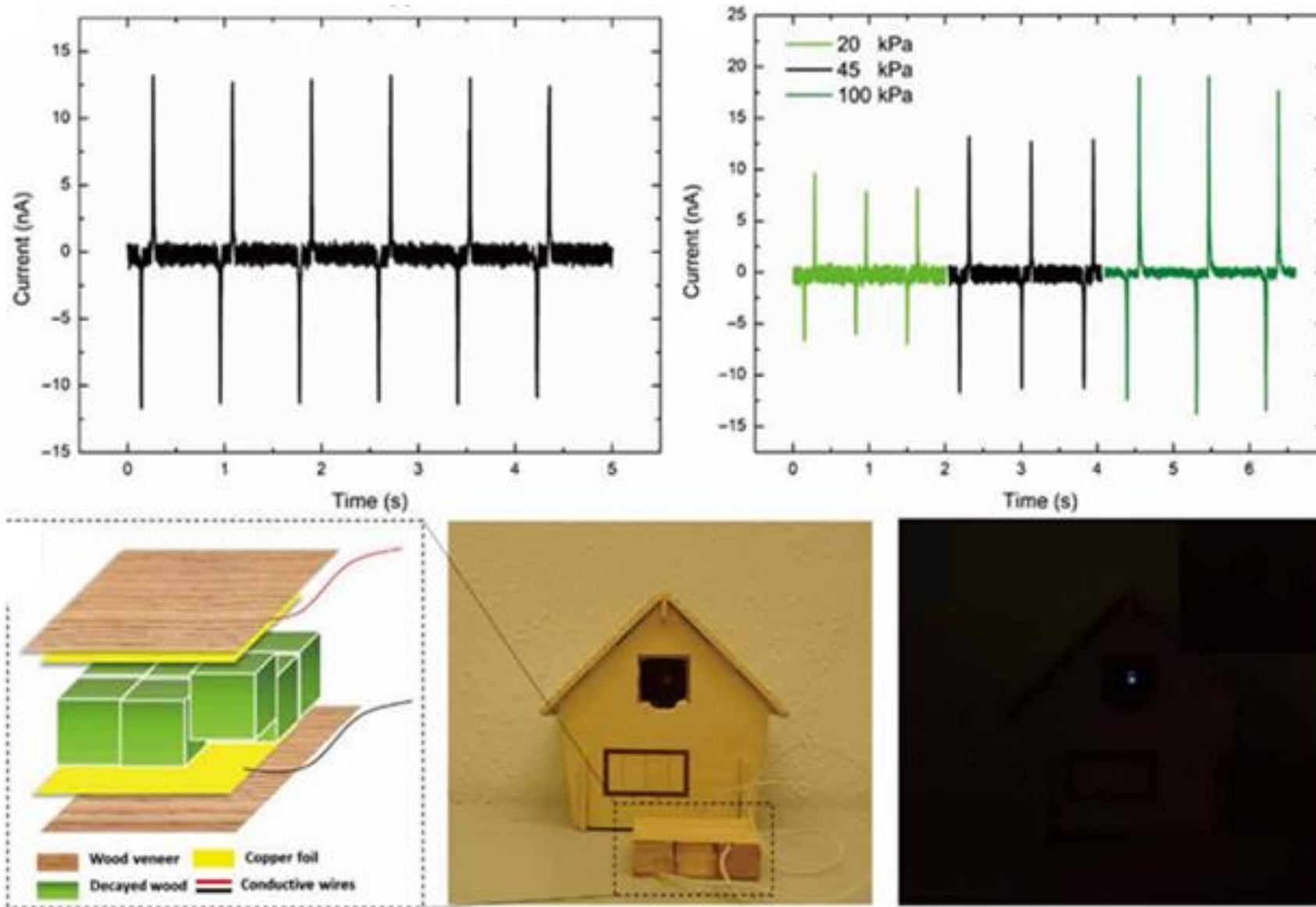
White rot-degraded balsa wood



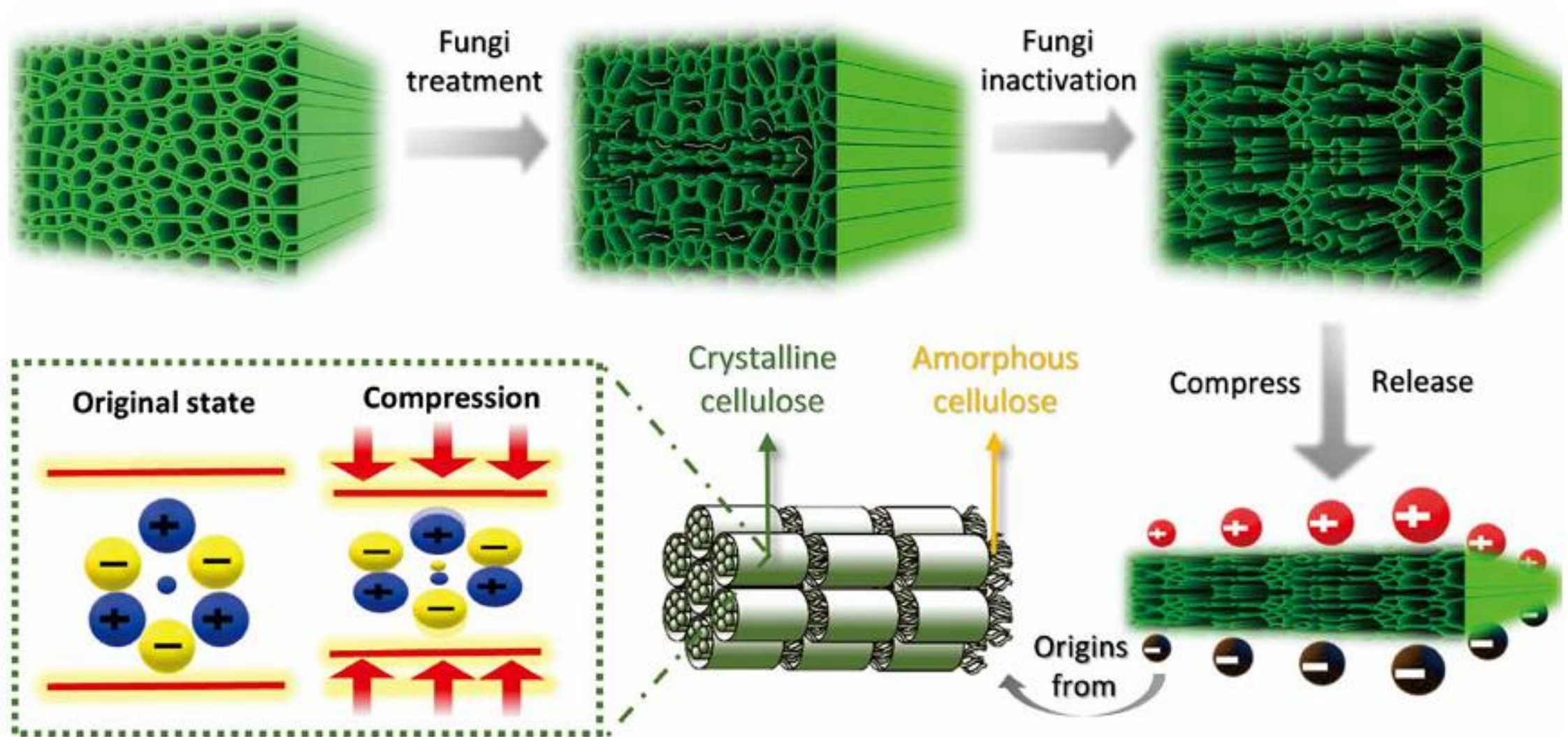
Piezoelectricity from white rot-degraded balsa wood



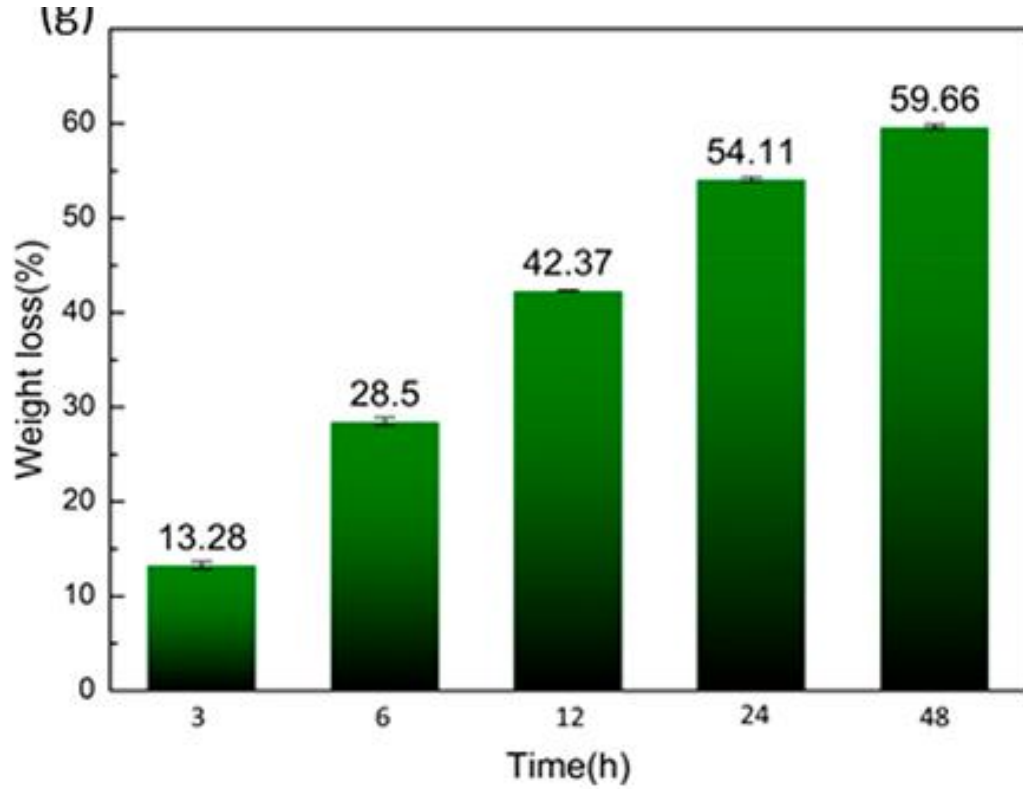
Piezoelectricity from white rot-degraded balsa wood



Piezoelectricity from white rot-degraded balsa wood

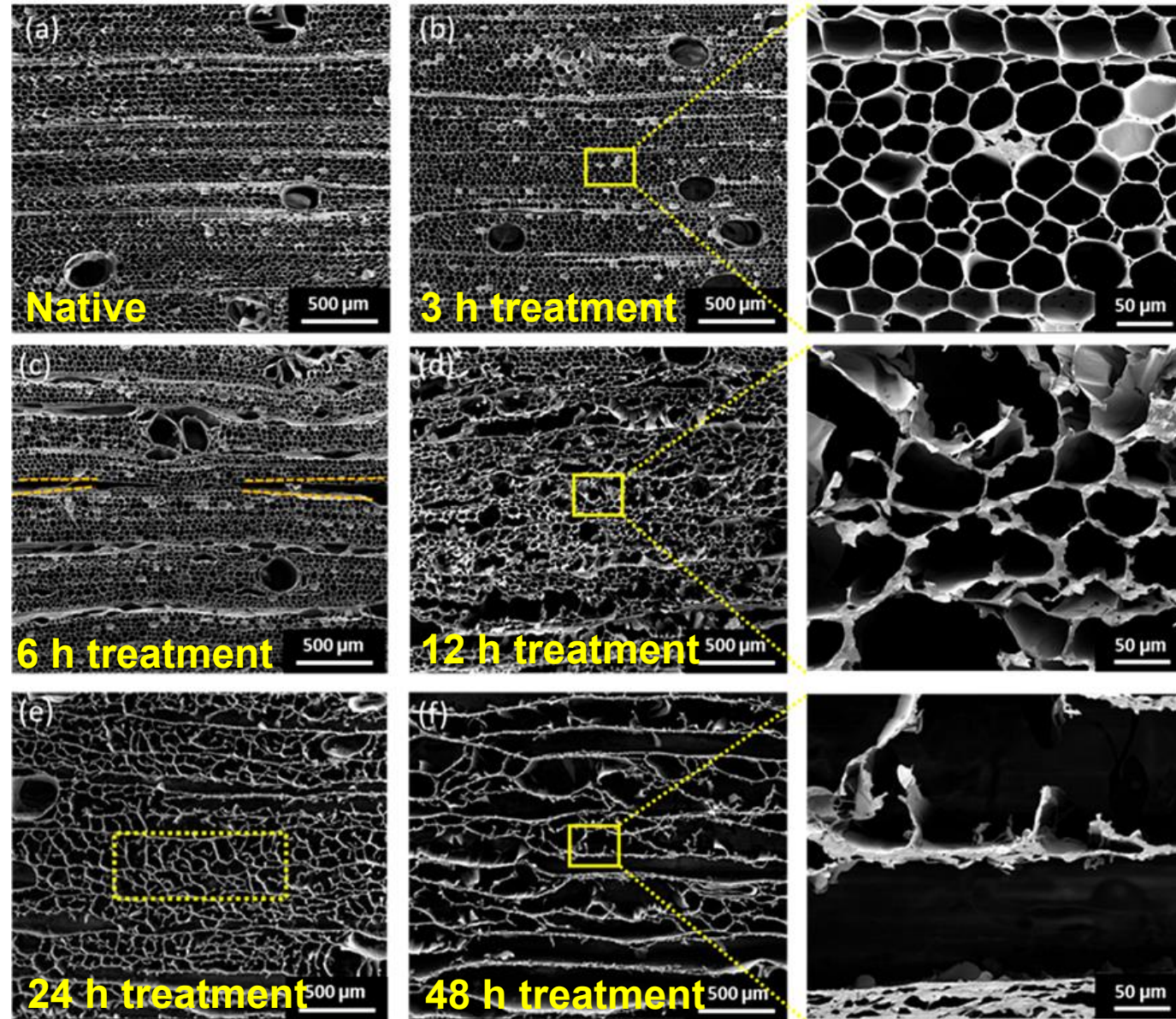


Piezoelectricity from chemically-delignified balsa wood

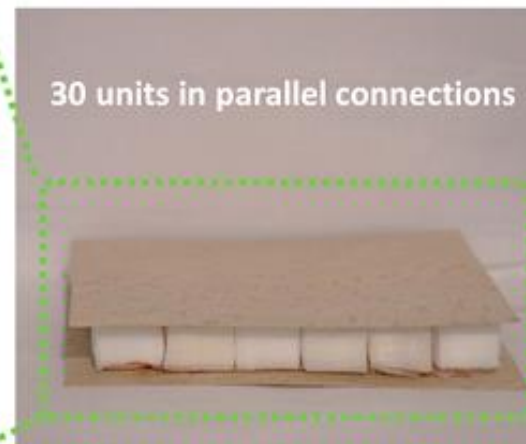
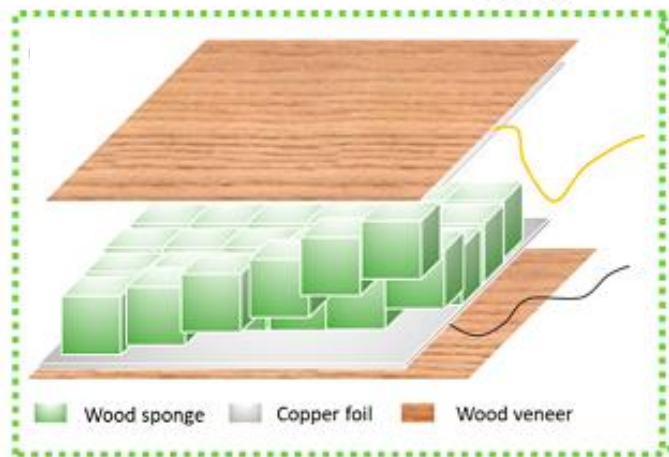
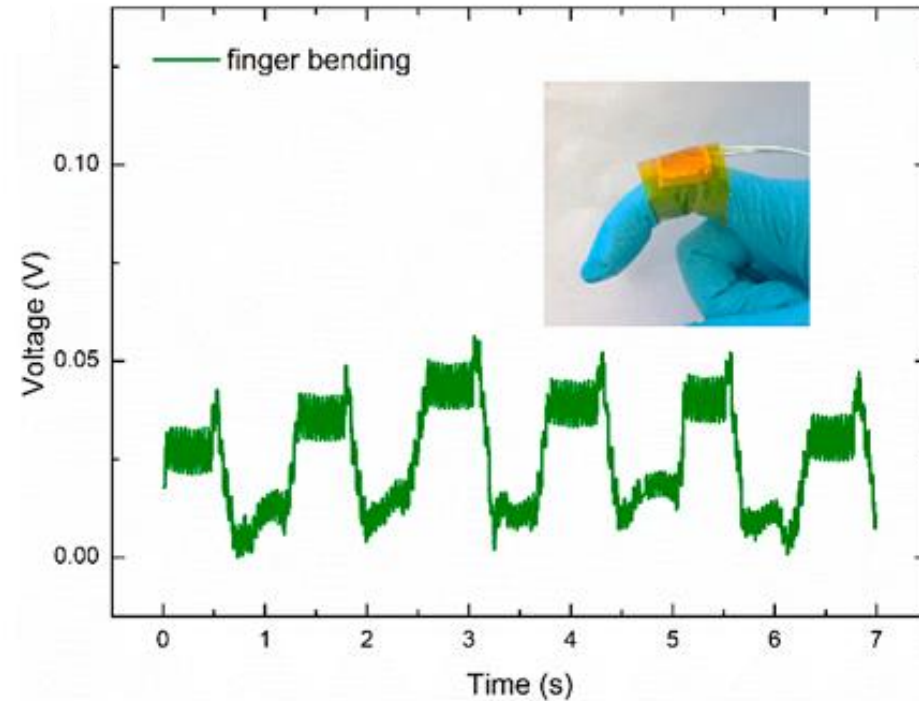
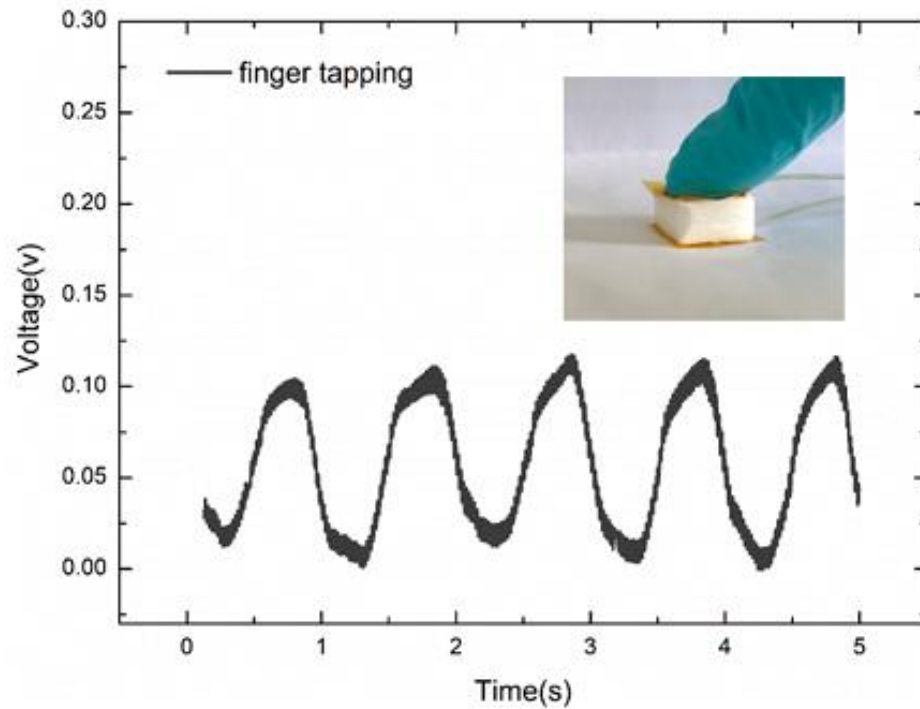


Treatment:

50:50 vol:vol glacial acetic acid and 30% H₂O₂, 80°C



Piezoelectricity from chemically-delignified balsa wood



«Spalted wood»

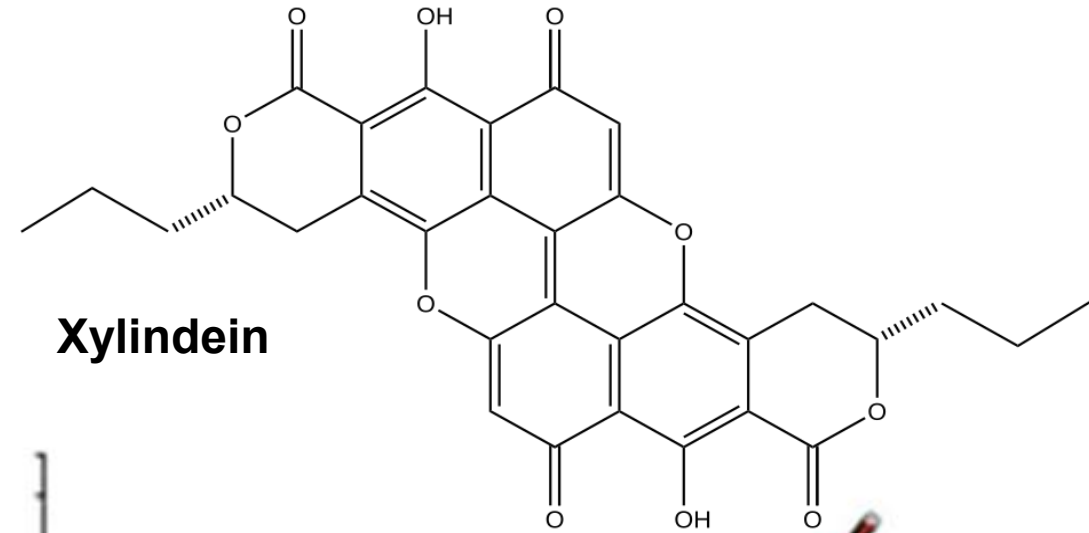
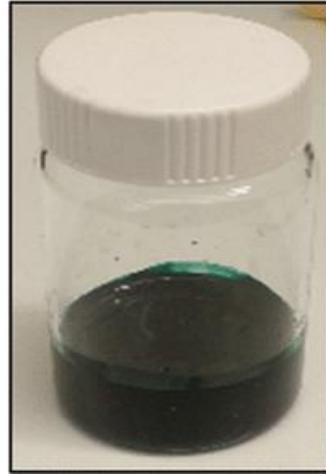
R. Hartig, *Lehrbuch der Baumkrankheiten* 1882, Springer (Berlin), p. 98:



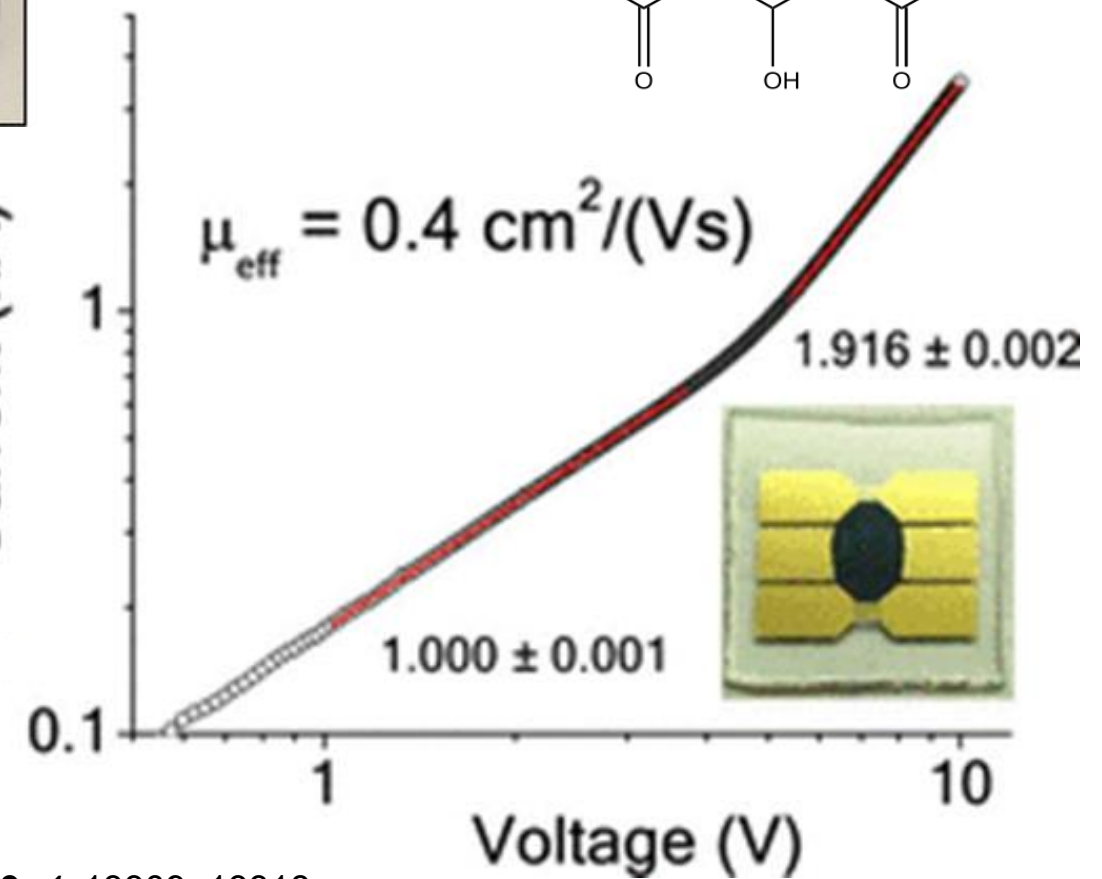
Ein allgemeineres Interesse beanspruchen unter den saprophytischen Holzpilzen noch die *Peziza aeruginosa*, welche zu den Discomyceten gehörend, doch hier schon erwähnt werden möge, da sie die sogen. Grünfäule des Holzes veranlasst. Insbesondere Eichen- und Rothbuchenholz, welches in stark zersetztem Zustande und anhaltend durchfeuchtet auf dem Waldboden längere Zeit gelegen hat, erhält oftmals eine intensiv spangrüne Färbung dadurch, dass das Mycel des genannten Pilzes, welches nebst den schüsselförmigen Fruchträgern intensiv grün gefärbt ist, den Holzkörper durchzieht und in den Wandungen der Holzelemente ebenfalls jenen grünen, extrahirbaren Farbstoff erzeugt.

Es soll derselbe wegen seiner Unzerstörbarkeit eine technische Verwendung finden, und neuerdings bemüht man sich in Hannover, durch künstliche Züchtung grünfaules Holz in grösserer Quantität zu erzeugen.

Chlorociboria aeruginosa



Current (nA)

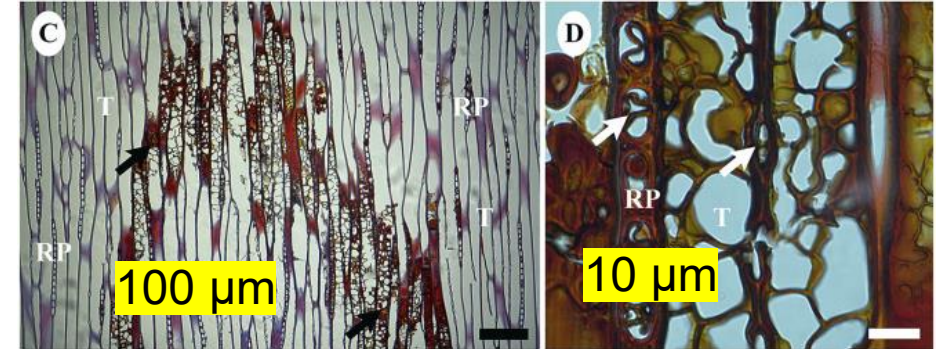


«Spalted wood»

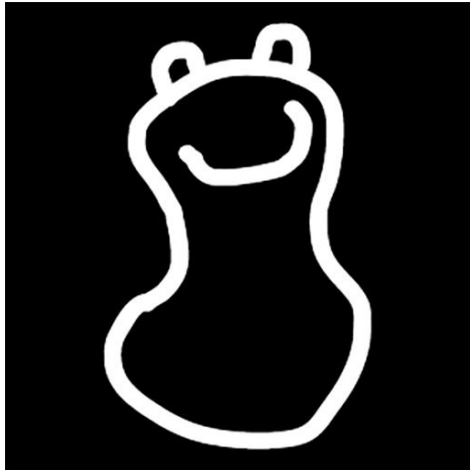
Extracellular secretion of (eu/allo)melanin by fungi minimises degradation of plant cells and supports structural integrity of zone lines and fungus survivability. The melanin appears to fill pit membranes and the lumina of adjoining cells where it helps form a complete and continuous almost impenetrable closely packed barrier.



Armillaria ostoyae in *Picea abies*



«Fungi Force»: a commercial engineered living wood material



<https://www.fungiforce.com>

MOLD BUSTERS®
DECONTAMINATION EXPERTS | EST.2005

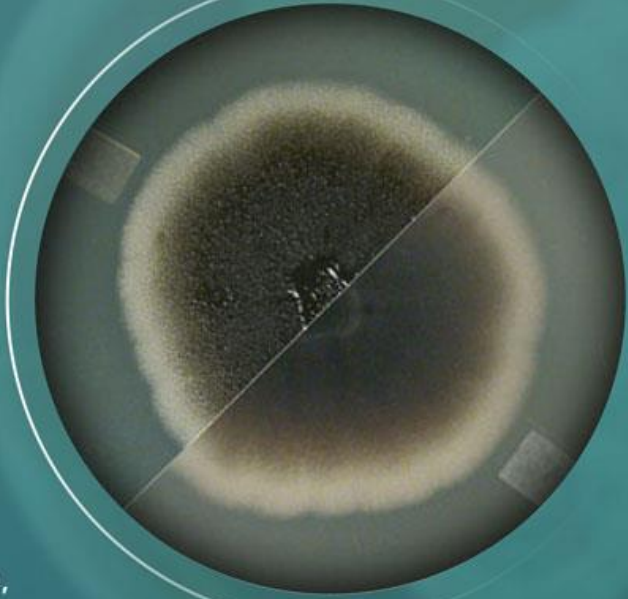
Aureobasidium pullulans (de Bary & Löwenthal) G. Arnaud, 1918

▶ Kingdom: Fungi	▶ Order: Dothideales
▶ Phylum: Ascomycota	▶ Family: <i>Sacrotheciaceae</i>
▶ Class: Dothideomycetes	▶ Genus: <i>Aureobasidium</i>

▶ **Ecology & habitat:** Generalist species (soil, air), epiphyte and endophyte on many plant species

▶ **Human & Animals Pathogenicity:** Contaminant in humidifiers or air conditioners, causes humidifiers or air conditioners ("humidifier lung").

▶ **Importance:** Potential for enzyme production (amylase, proteinase, lipase, cellulase, xylanase, manganese, transferases)



«Fungi Force»: a commercial engineered living wood material



From the Fungi Force website (<https://www.fungiforce.com>):

« Fungi Force is a 100% organic stain or bio stain. It consists of two separate components. On the one hand a biological water-based mould solution and on the other hand natural linseed oil.

Fungi Force is based on a naturally occurring harmless fungus (*Aureobasidium pullulans*). This adheres to the wood and thus forms a matte black solid protective layer against harmful UV-light from the sun. This protective layer also prevents the colonization of wood by other harmful fungi.

Nutrition for the fungus is natural linseed oil, which is also applied to the wood after the fungus has been applied. This oil is moisturizing and protects the wood against the effects of moisture.

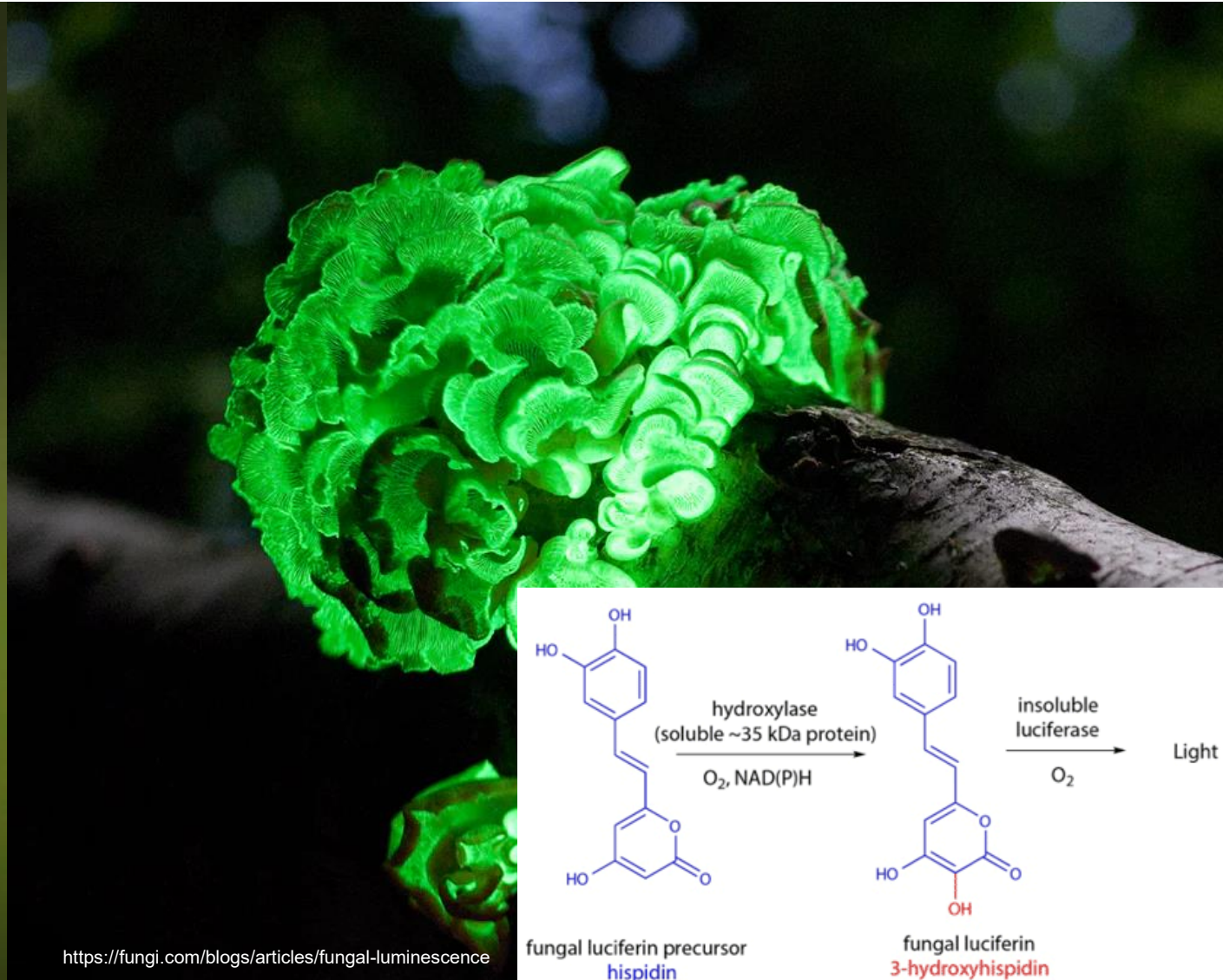
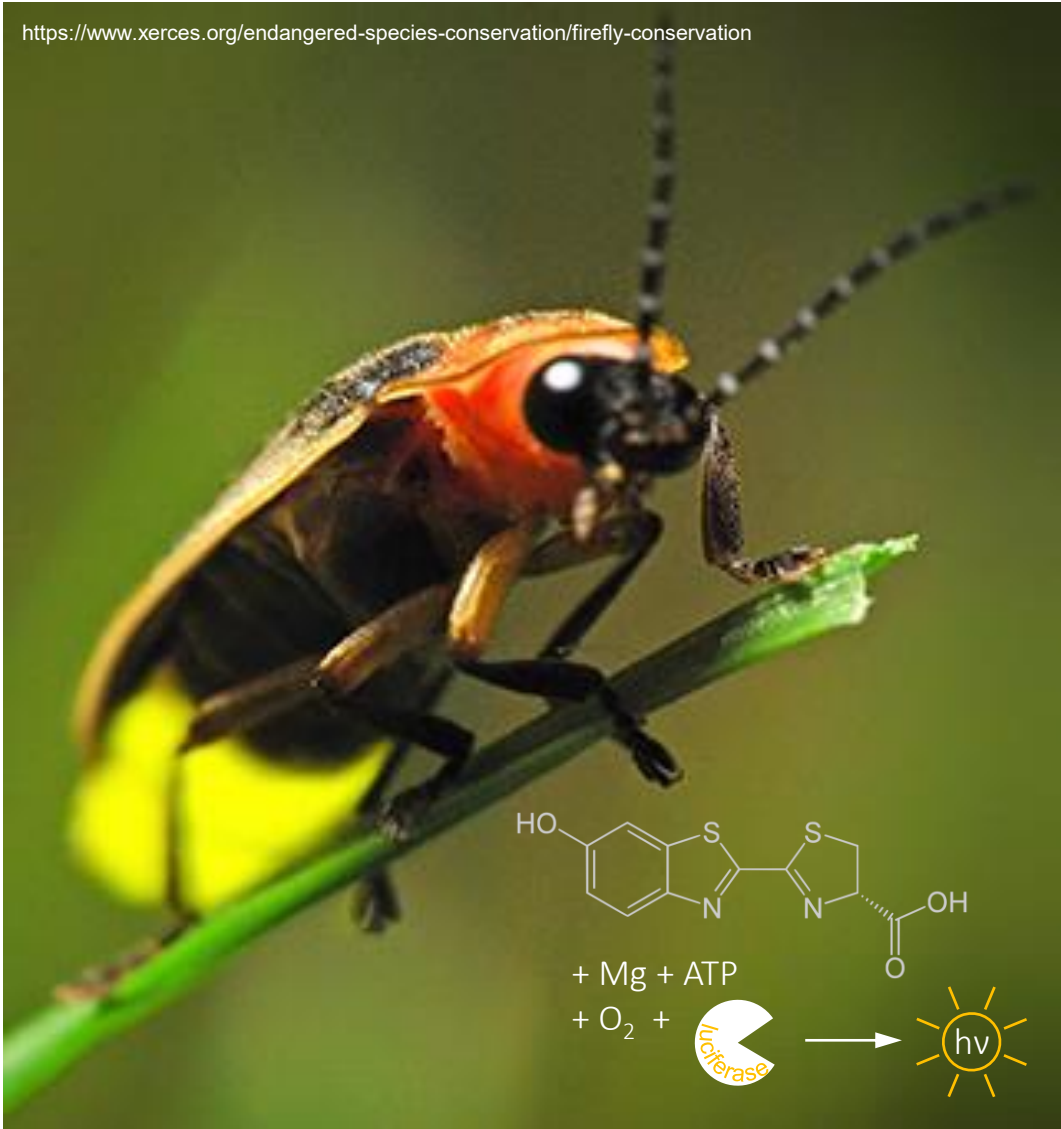
Fungi Force is applied to untreated wood. About a week after application of the fungus layer and the natural linseed oil Fungi Force has attached itself to the wood and the protective layer is dry and does not give off.

Fungi Force requires hardly any maintenance. Annual post-treatment with the oil finish. natural linseed oil to feed the fungus and keep it nice and black is sufficient. After a few years, this maintenance can even take place every other year.

Fungi Force is self-healing. This means that minor damage such as cracks and scratches are repaired by the living mould layer itself. »

Fireflies and Foxfire

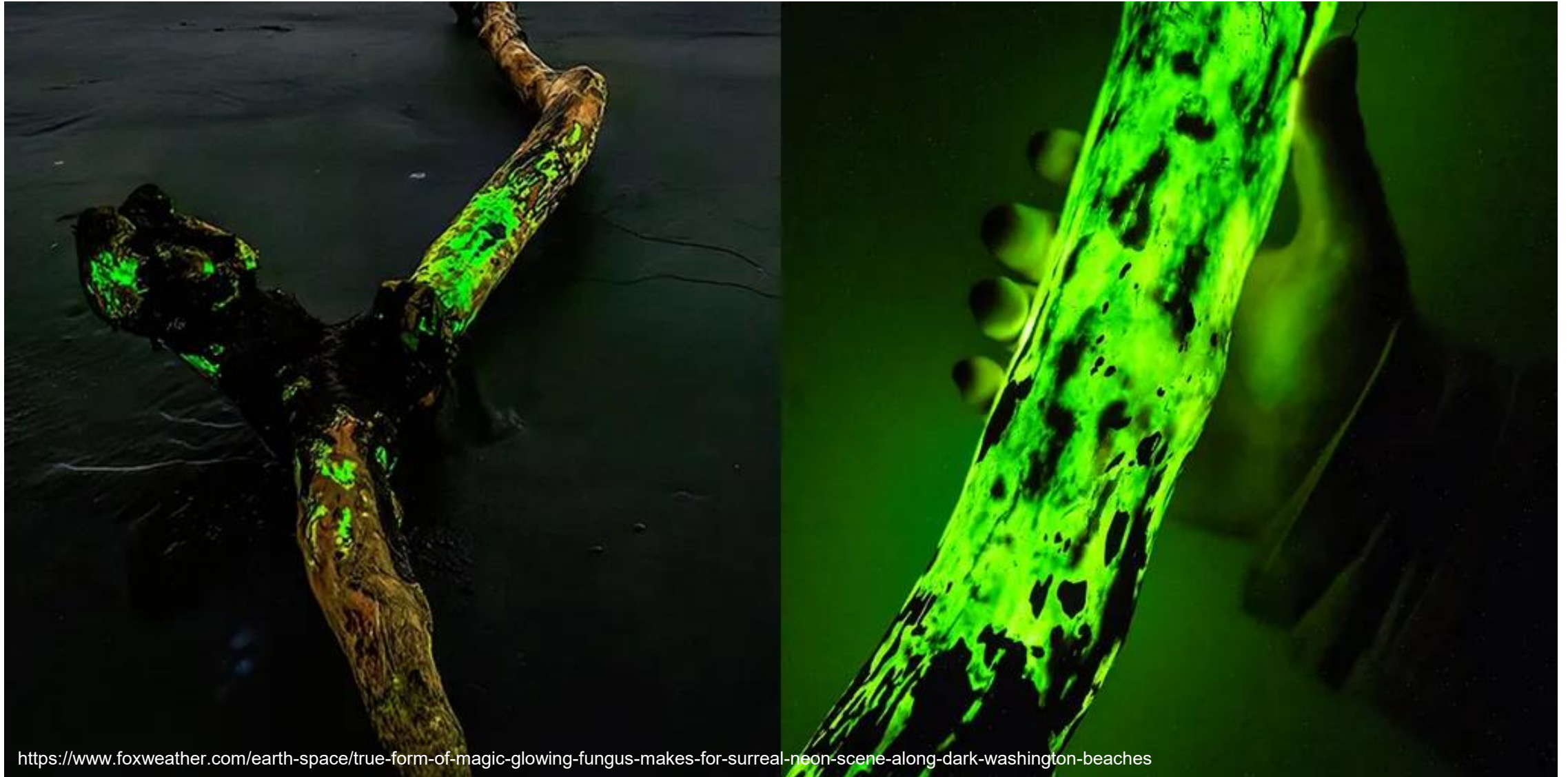
<https://www.xerces.org/endangered-species-conservation/firefly-conservation>



<https://fungi.com/blogs/articles/fungal-luminescence>

Kaskova et al. *Sci. Adv.* 2017, 3(4), e1602847

Bioluminescent Wood: a naturally-occurring Hybrid Living Material



Artificial Bioluminescent wood

« The chemical reaction [responsible for light emission] has not [yet] been artificially induced in wood through manipulation of the conditions for fungi to glow.

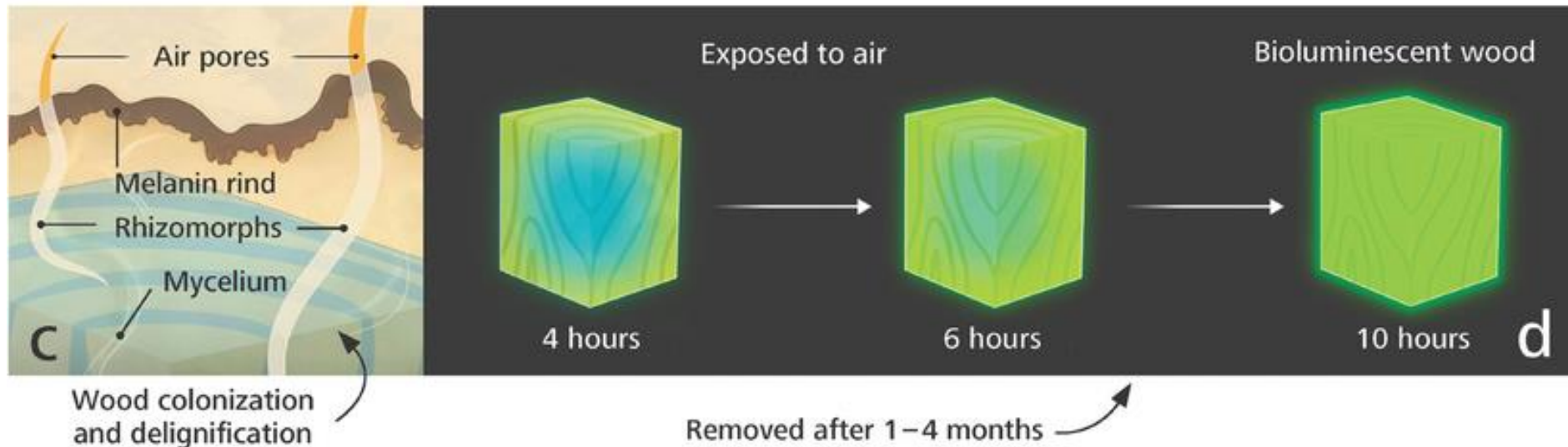
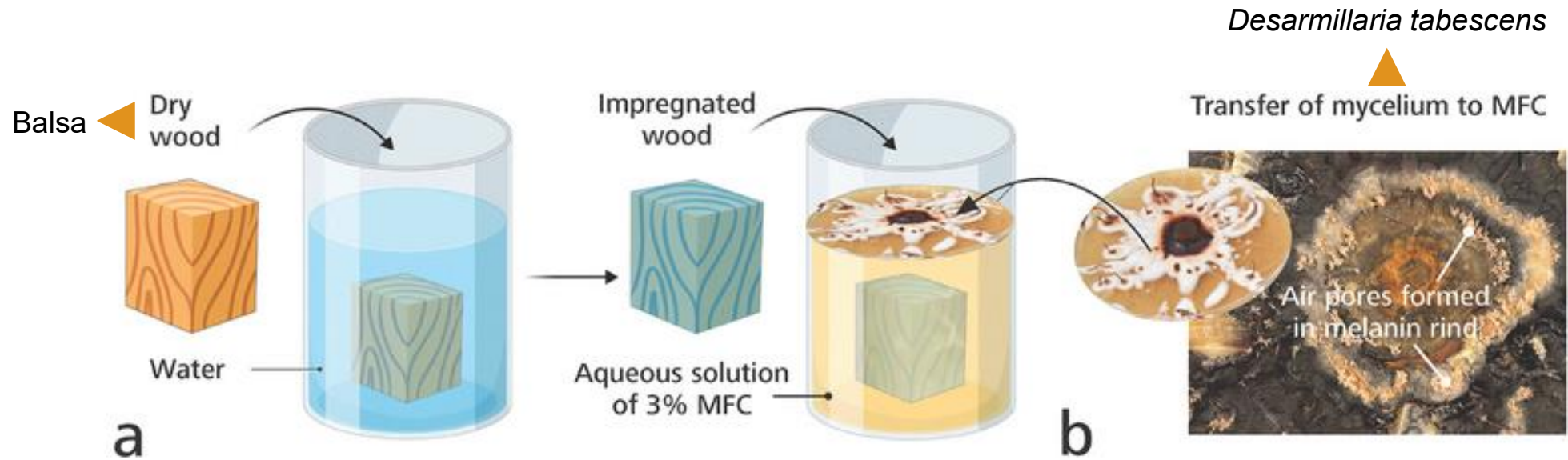
The main reason for this is that the balance between the choice of fungal species, wood species, its moisture content and the environmental conditions required to produce bioluminescent wood is very challenging.

Investigating the production of bioluminescent wood, we hypothesized that using the white rot fungus *Desarmillaria tabescens* will lead to bioluminescence, because of its ability to colonize and degrade lignin in wood with a very high moisture content.

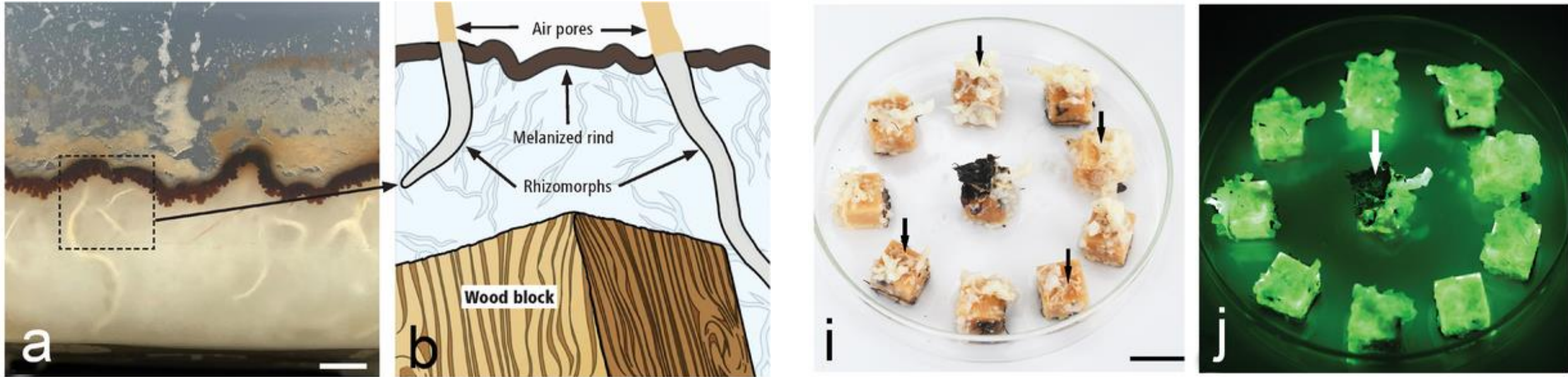
The hybrid material with the highest bioluminescence is produced by soaking the wood blocks before co-cultivating them with the fungus for 3 months.

Regardless of the incubation period, the strongest bioluminescence is evident from balsa wood blocks with a moisture content of 700–1200%, highlighting the fundamental role of moisture content for bioluminescence production. »

Artificial Bioluminescent Wood



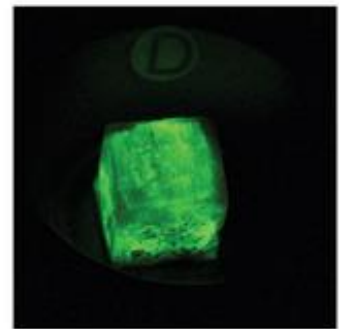
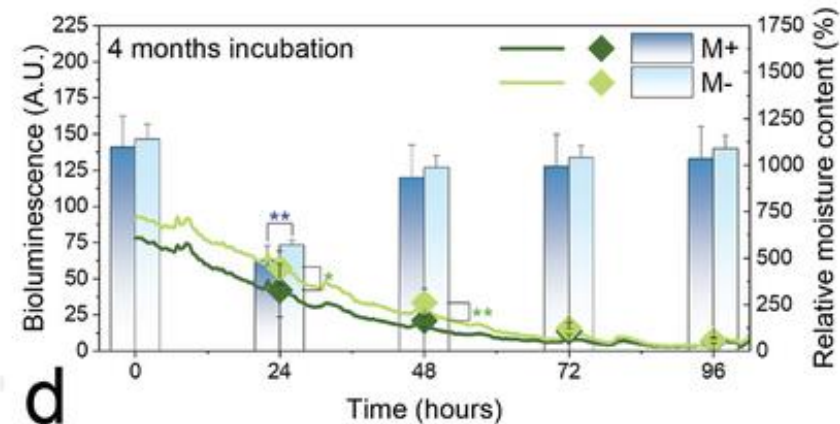
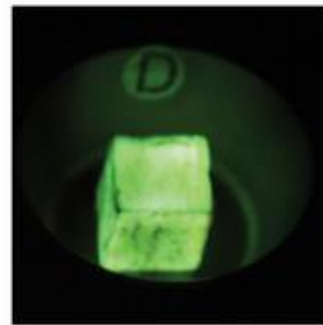
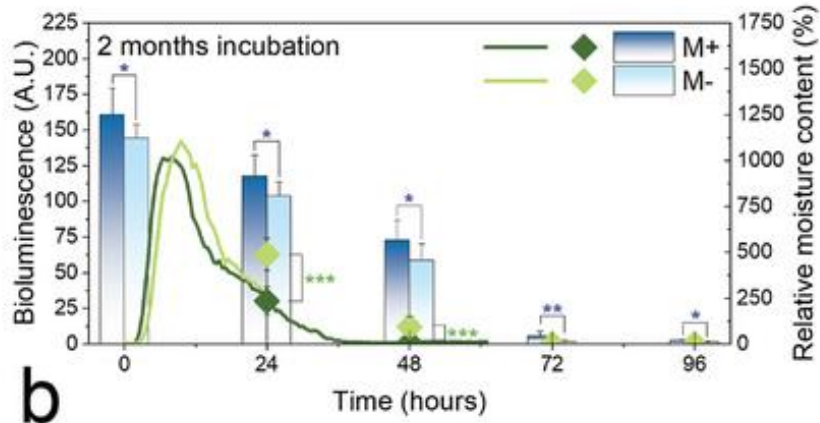
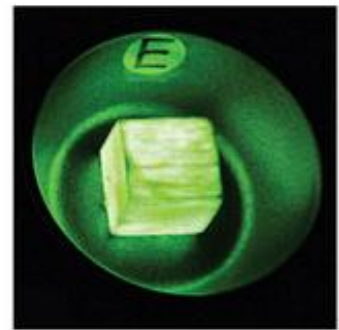
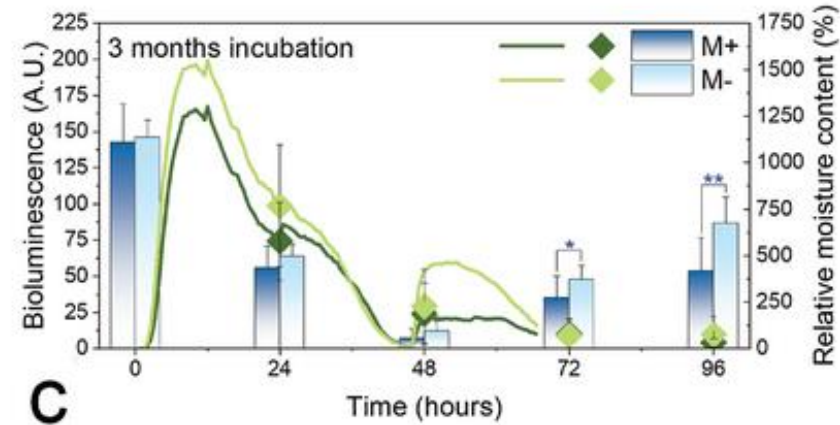
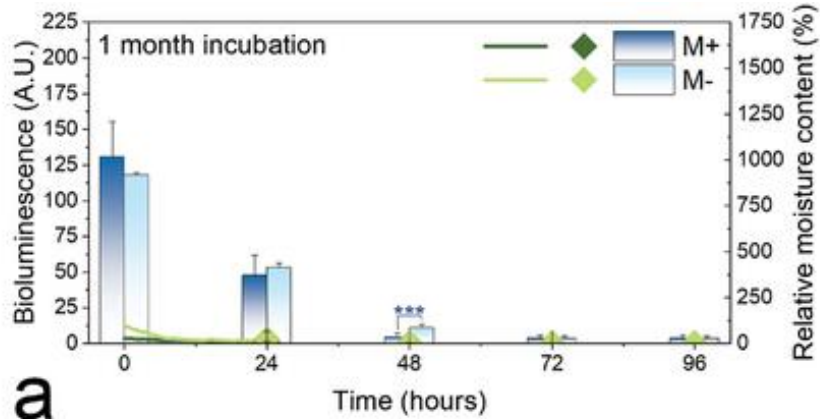
Artificial Bioluminescent Wood



Incubation of wood blocks in 3% MFC with *D. tabescens* for producing bioluminescent wood. a) Non-melanized rhizomorphs (arrow) colonize MFC and the submerged wood blocks. Scale bar = 1 cm. b) Illustration showing wood blocks submerged in MFC and the connection of rhizomorphs and air pores (arrows). At the substrate-air interface, MFC is covered by a melanized rind (arrow). i) Balsa wood blocks incubated for 3 months with *D. tabescens* and exposed to light. Note abundant rhizomorphs (arrows) of *D. tabescens* have strongly colonized balsa wood blocks (scale bar = 1.5 cm). j) Wood blocks in i) exposed to air in the dark showing strong bioluminescence induced by *D. tabescens*. Note dark melanin rind (arrow) absorbing bioluminescence emitted from wood (scale bar = 1.0 cm).

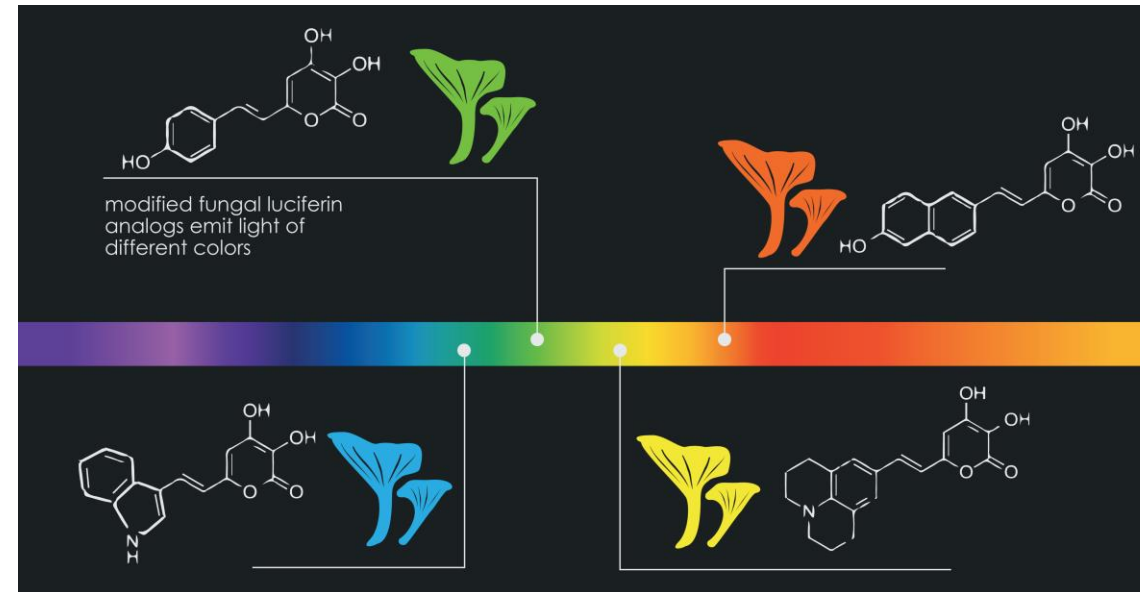
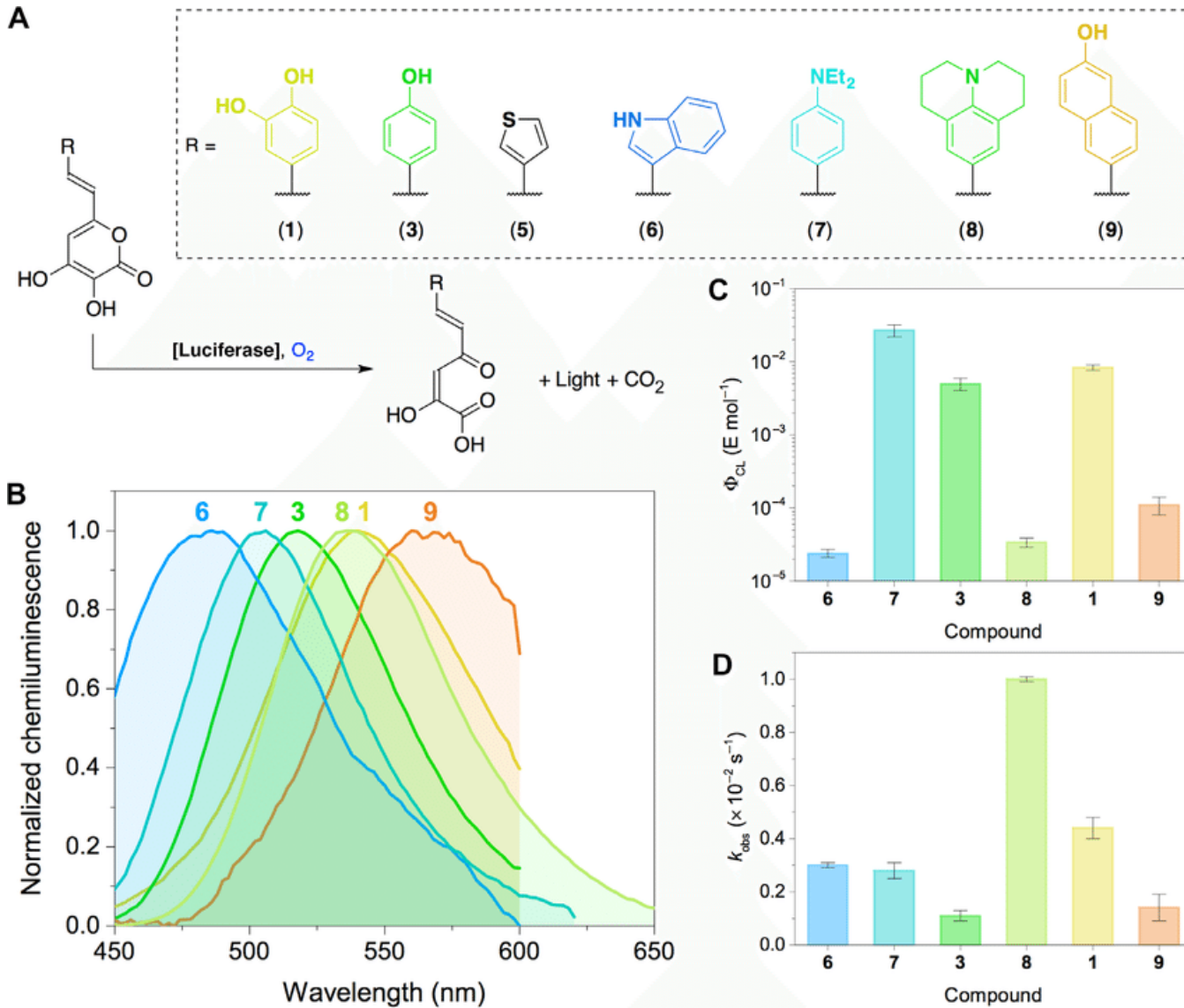
Artificial Bioluminescent Wood

Relationship between bioluminescence and moisture content of wood blocks (incubated for 1, 2, 3, and 4 months) over 96 h in a dark room at RT ([M+] = with malt, [M-] = without malt).

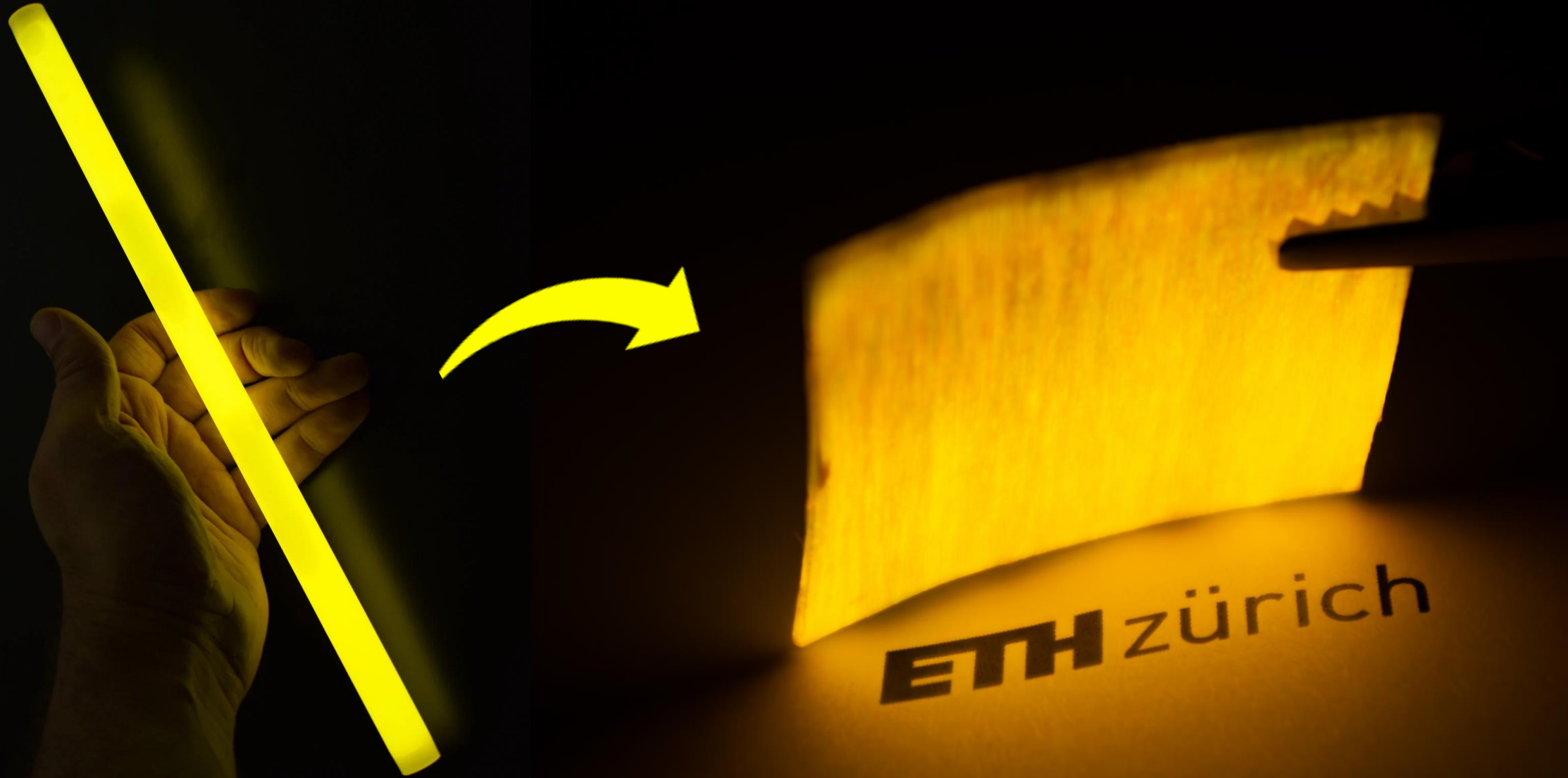


Artificial Bioluminescent Wood

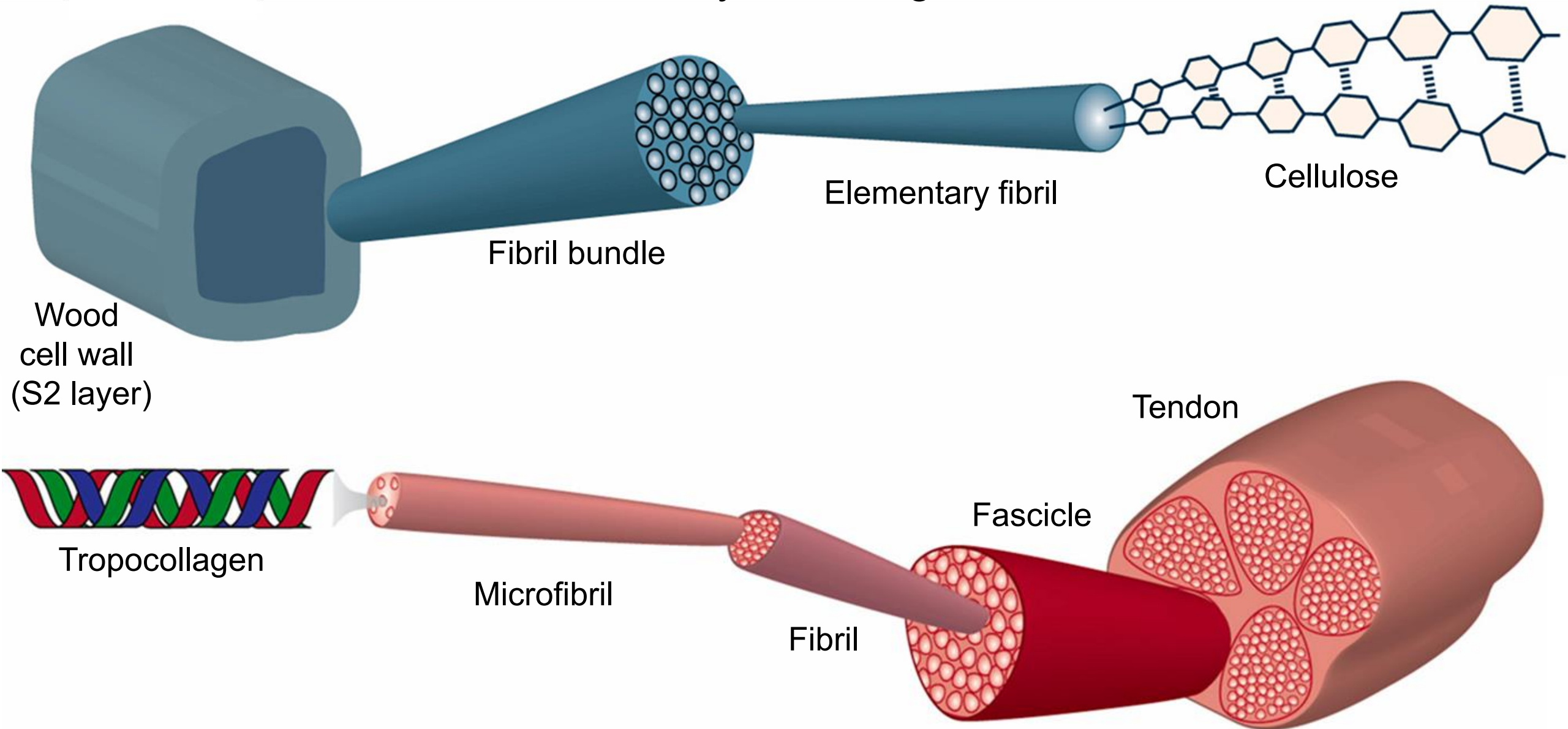
Artificial Bioluminescent Wood: expanding the color palette



Chemiluminescent Wood

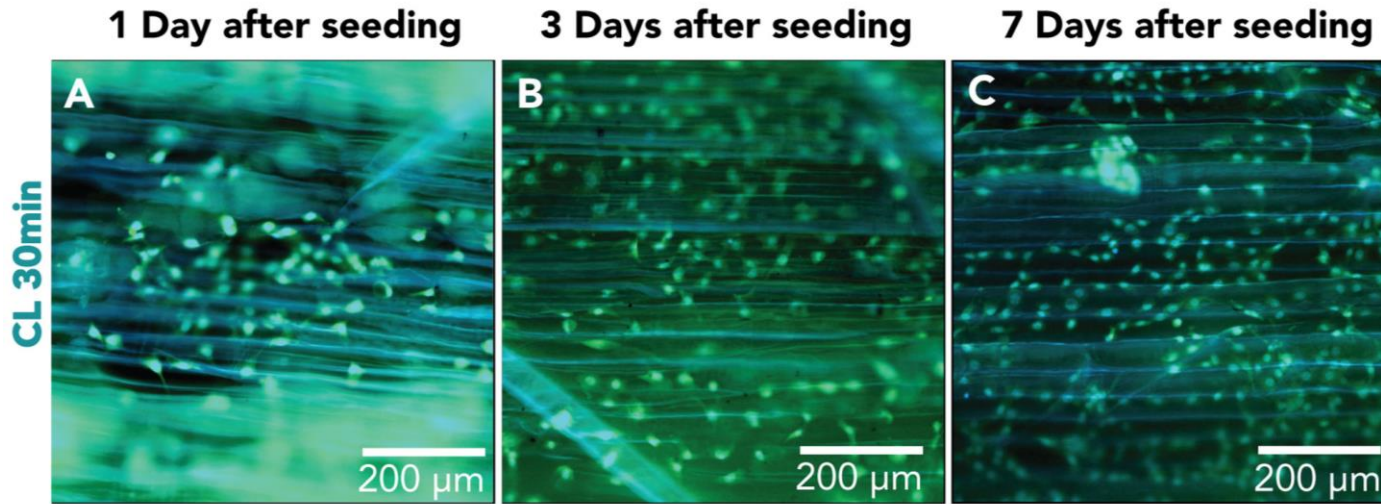


Wood as an abiotic scaffold for Hybrid Living Materials

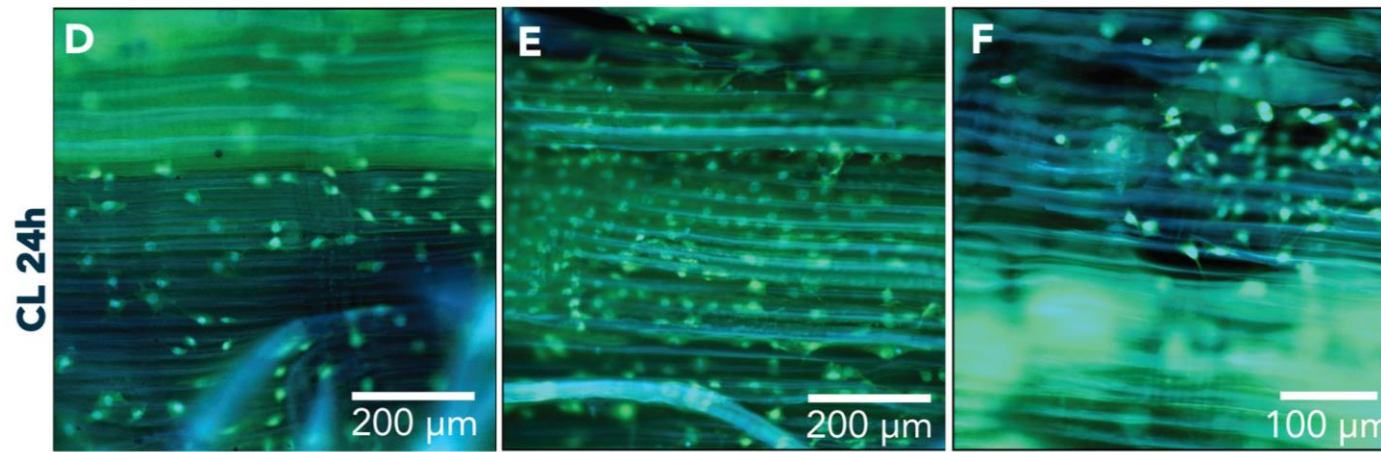


Wood as an abiotic scaffold for Hybrid Living Materials

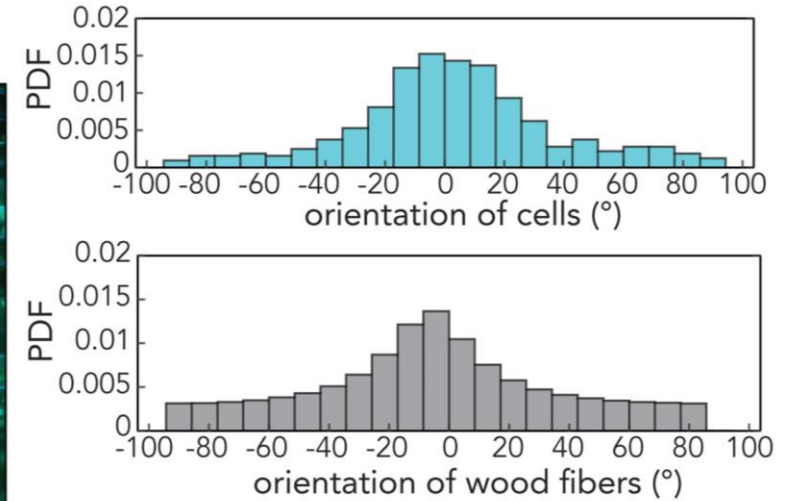
Cell morphology



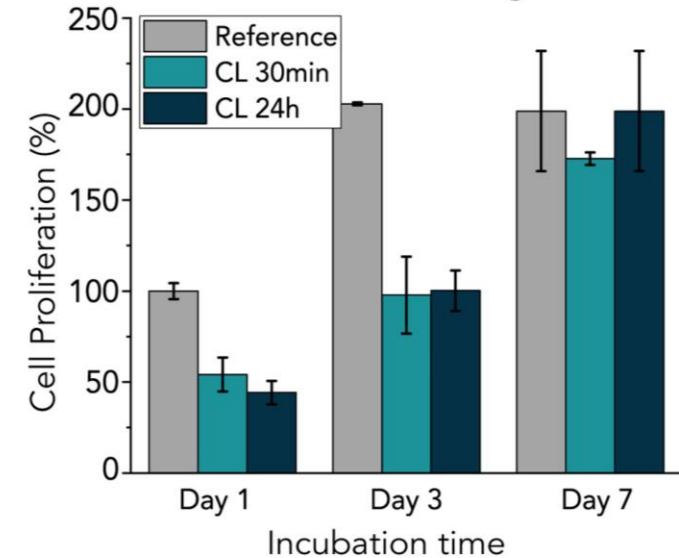
Murine NIH3T3 fibroblasts cultivated on gelatin-impregnated, delignified wood



G Cell orientation



H Cell viability



ETH zürich

EPFL

THANK YOU!



**Active & Adaptive
Wood Materials**